

Risk Analysis for Phytophthora ramorum Werres, de Cock & Man in't Veld, Causal Agent of Sudden Oak Death, Ramorum Leaf Blight, and Ramorum Dieback

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Executive Summary

This pest risk analysis was conducted by the United States Department of Agriculture, Animal and Plant Health Inspection Service, Plant Protection and Quarantine, Center for Plant Health Science and Technology, Plant Epidemiology and Risk Analysis Laboratory to assess the risk of the importation and domestic spread of *Phytophthora ramorum* Werres, de Cock, & Man in't Veld, 2001. This pathogen is the subject of USDA Emergency Regulations due to its threat to agricultural, horticultural, and natural ecosystems in the United States. The analysis focused on 1) the risks associated with the importation of plants (including plants in APHIS-approved growing media and bare-root plants) and plant products (wood, lumber, chips, bark and other wood products, and greenery) that are hosts of *P. ramorum*; 2) the risks associated with the domestic movement of the pathogen through plants, plant products, soil, other growing media, compost, and water; and 3) mitigation measures to prevent the movement and spread of *P. ramorum* to non-infested areas in the United States.

Diseases caused by an unknown species of *Phytophthora* were first observed in Europe on nursery stock in 1993 and in California forests on *Quercus* spp. and *Lithocarpus densiflorus* in 1995, but the pathogen, *P. ramorum*, was not formally described until 2001. Since initial reports and detections, *P. ramorum* has expanded its geographic distribution in forested areas of California and Oregon and has been detected in hundreds of nurseries in Europe and North America. The pathogen continues to be detected on new hosts and in nurseries outside of quarantined and regulated areas.

Several biological factors affect the risk of introduction and establishment of *P. ramorum*, including the large host range, variation in symptoms, production of multiple spore states, and factors inducing and breaking latency and dormancy. The large host range is mirrored by the complexity of the disease symptoms, which can be grouped into three general disease categories: canker, foliage, and dieback. Hosts can exhibit the symptoms of one or more of these disease categories.

The risk presented by *P. ramorum* is High. The risk is based on six Elements: Climate-Host Interaction, Host Range, Dispersal Potential, Economic Impact, Environmental Impact, and Pest Opportunity.

Climate-Host Interaction. The risk rating is High for this element. The level of certainty for this risk rating is fairly certain. Most of the eastern United States has actual and potential hosts growing in climates conducive to infection. The uncertainty lies in the range of biotic and abiotic factors triggering the establishment of *P. ramorum* in new areas.

Host Range. The large number of hosts in multiple plant families, differential susceptibility, and virulence warrant a High risk rating. The level of uncertainty of this risk element is low because *P. ramorum* already has a large demonstrated host range.

Dispersal Potential. In the United States, both regulated and associated hosts are widely distributed, overlapping, abundant, and susceptible. In addition, the pathogen is polycyclic, infections may remain undetected for years, long-distance dispersal via trade has been

demonstrated, and circumstantial evidence suggests spread by natural means. For these reasons, the risk rating for this element is High and the level of uncertainty is Low based on the evidence of human-assisted and natural movement.

Economic Impact. *Phytophthora ramorum* is impacting the international and domestic movement of plants and plant products (nursery stock, fruit, logs, lumber, *etc*.) and has resulted in restrictions in trade and movement. The risk rating for this element is High and the uncertainty depends on the relationships between the extent of the host range and the value of these plants on the open market.

Environmental Impact. The risk rating for this element is High. The environmental factors include: (1) direct costs of prevention, eradication, or suppression, (2) current-use and future-use values, and (3) indirect ecological consequences (changes in locally important ecological processes such as perturbations of hydrological cycles, *e.g.*, flood control and water supply; waste assimilation; nutrient recycling, conservation, and regeneration of soils; and crop pollination). Assessing the environmental impact is difficult due to the uncertainty of cost estimates that address all of the relevant ecological components; therefore, the uncertainty of this element is High.

Pest Opportunity for Introduction. The rating for Likelihood of Introduction is High. Both natural and human-assisted factors aid in the dispersal of *P. ramorum* to areas where suitable hosts and climatic conditions are conducive to establishing and sustaining a population. Differences in reproductive ability and infection susceptibility of a large number of hosts contribute to the uncertainty. The uncertainty for this rating is High and is based on the variability in detecting *P. ramorum* and the ability to predict the levels of resistance and susceptibility among hosts and potential hosts occurring in non-infested regions.

In addition, the following pathways were analyzed: nursery stock, Christmas trees (cut and living), cut foliage/flowers, wood and wood products, greenwaste and compost, potting media, and soil. Although individual elements for cut Christmas trees and cut foliage/flowers pathways were rated Medium, the overall risk potential for all pathways was High.

Current regulatory efforts (exclusion, eradication, containment, suppression, and sanitation) and potential mitigations for pathways were reviewed. There are considerable challenges in devitalizing this pathogen because it occurs in forests and regulated articles (*e.g.* nursery stock, wood/wood products, compost). In addition, there are a limited number of long-term fungicidal or eradicant treatments and the efficacy of these treatments to inoculum varies.

Exclusion is the most effective mitigation option, but domestic and international trade render this difficult. Eradication of the pathogen via chemicals is problematic, because the pesticides available for control are fungistatic, not fungicidal. Containment and suppression efforts vary based on forest and nursery scenarios. These include forest and water surveys, nursery certification programs, and other methods to reduce inoculum, such as the destruction of host material. Sanitation (pathogen-free water, pots, potting media, benches, tools and equipment, clothing, *etc.*) is required to maintain pathogen-free material.

Pathway mitigation measures include chemical, physical, and cultural and biological treatments. The efficacy of chemical control is dependent upon timing, type of application, and location of the pathogen in or on the plant. Physical control includes heat, heat and vacuum, heat via aerated steam, removal of infected bark and wood, and air drying. Cultural and biological methods include best management practices and the use of biological antagonists.

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I. Initiating Event: Proposed Action

This is an update of the pest risk analysis (PRA) conducted in May 2005 by the United States Department of Agriculture, Animal and Plant Health Inspection Service, Plant Protection and Ouarantine, Center for Plant Health Science and Technology, Plant Epidemiology and Risk Analysis Laboratory (USDA, APHIS, PPQ, CPHST, PERAL) to assess the risk of the importation and domestic spread of *Phytophthora ramorum* Werres, de Cock, & Man in't Veld, 2001. Phytophthora ramorum is the subject of USDA Emergency Regulations due to its threat to agricultural and natural ecosystems in the United States. This analysis will focus on 1) the risks associated with the importation of plants (including plants in APHIS-approved growing media and bare-root plants) and plant products (wood, lumber, chips, bark and other wood products, and greenery) of hosts of P. ramorum; 2) the risks associated with the domestic movement of the pathogen through plants, plant products, soil, other growing media, compost, and water; and 3) mitigation measures to prevent the movement and spread of *P. ramorum* to non-infested areas in the United States. This document consists of four major components: a pest data sheet, an organism assessment, pathway assessments, and mitigation measures. The pathways analyzed are nursery stock, Christmas trees, cut foliage/flowers, wood and wood products, greenwaste and compost, potting media, and soil.

The authority for APHIS to regulate plant pests and plant products is derived from the Plant Protection Act of 2000 (7 USC §7701 *et seq.*); for plant imports, the Nursery Stock, Plants, Roots, Bulbs, Seed and Other Plant Products subpart of the Code of Federal Regulations (7 CFR §319.37); and for regulating domestic interstate movement of items at risk for moving *P. ramorum*, Phytophthora Ramorum (7 CFR §301.92). The risk assessment methodology and rating criteria (APHIS, 2002) and the use of biological and phytosanitary terms are consistent with relevant international standards published by the International Plant Protection Convention (IPPC).

The current pest risk analysis was prepared in response to a need to promulgate regulations addressing the international and domestic movement of *P. ramorum* and its hosts at the genus level. The justification for this approach is based on scientific considerations such as an expanding list of natural hosts (22 families, 42 genera, and over 66 species reported as natural hosts in 2005; 35 families, 70 genera, and over 109 species in 2007), the unknown host specificity of this pathogen, the potential movement of infected asymptomatic plants, the variability of environmental conditions leading to expression of the disease, the expanding list of countries reporting the pathogen (seven European countries reported detections in 2005; 16 countries reported detections in 2007), and recent expansions within the United States, specifically in Humboldt County, CA and Curry County, OR. A compounding problem is the variable resistance observed within a species, e.g., Umbellularia californica (Meshriy et al., 2005). Additionally, although hosts may be present in different countries, they have not been found to be infected in all counties, even when the pathogen is present; for example, Quercus rubra is present in the United Kingdom (Jones et al., 2003) and the Netherlands, but only found infected in the Netherlands (RAPRA, 2007). The analysis addresses the potential risks from products associated with these host genera, including soil, compost, and growing media

The domestic movement of *P. ramorum* is currently regulated under an Interim Rule, "Domestic Quarantine Notices *Phytophthora Ramorum*" 7 CFR §301.92, and an Agriculture Department Emergency Federal Order Restricting Movement of Nursery Stock from California, Oregon, and Washington Nurseries (APHIS, 2007b). The USDA implemented emergency measures to regulate the international movement of regulated articles from Europe; these measures mirrored the federal domestic regulations that went into effect November 1, 2002. Changes in Federal domestic emergency measures are applied to movement from Europe (February 27, 2007).

II. Glossary

The majority of the terms listed are quoted directly from the reference cited.

Baiting – A method of recovering fungi from aquatic and soil/potting media by using various types of organic substrates. Classic baiting techniques for species of *Phytophthora* (Erwin and Ribeiro, 1996) using pears and leaves of hosts are used for *P. ramorum* (APHIS, 2004b).

Chlamydospore – Spore, usually globose but occasionally ovoid, that is delimited from the mycelium by a septum and may be terminal (at the end of the hyphae) or intercalary (formed in the middle of a hyphal strand) with a thickened wall. It "...survives for a long time in soil" (Erwin and Ribeiro, 1996).

Disease Cycle – This is the sequence of events involved in disease development, including the stages of development of the pathogen and the effect of the disease on the host; the chain of events that occurs between the time of infection and the final expression of disease (Shurtleff and Averre, 1997).

Heterothallism (adjective **heterothallic**) – Self-sterility; a sexual condition in which an individual produces only one kind of gamete. Used chiefly in reference to fungi and algae (Shurtleff and Averre, 1997).

Host – A living organism (*e.g.*, a plant) harboring or invaded by a parasite and from which the parasite obtains part or all of its nourishment (Shurtleff and Averre, 1997).

Regulated Host – Host plant that is naturally infected and for which Koch's postulates have been completed, documented, reviewed, and accepted. Some are regulated in part (such as redwood and Douglas-fir) and some are regulated in their entirety (such as tanoak and western starflower) (APHIS, 2007a).

Associated Plant – Host plant that is reported to be naturally infected and from which *P. ramorum* has been cultured and/or detected using polymerase chain reaction (PCR). For each of these, traditional Koch's postulates have not yet been completed or documented and reviewed. These reports must be documented and reviewed by PPQ before a plant becomes an APHIS Regulated Host for *P. ramorum* (APHIS, 2007a).

Experimental Host – Host plant that has indicated susceptibility to infection by *P. ramorum* in experiments.

Host Range – The complete range of plants that may be attacked by a given pathogen (Shurtleff and Averre, 1997).

Hypha(e) – The basic vegetative unit of structure and function of most fungi; a largely microscopic tubular filament that increases in length by growth at its tip. New hyphae arise as lateral branches. Some can become specialized for given functions including producing spores, penetrating host tissues, *etc.* (Erwin and Ribeiro, 1996).

Koch's Postulates – Four rules, proposed by Robert Koch, followed to prove the pathogenicity of a microorganism (Shurtleff and Averre, 1997). The rules below work well for most fungi, bacteria, nematodes, and related organisms (Agrios, 2005). A modification is used for hard to isolate pathogens, such as some viruses (Agrios, 2005, p. 27; Shurtleff and Averre, 1997).

- Rule 1. Organism is consistently associated with a disease syndrome.
- Rule 2. Organism is isolated and grown in pure culture.
- Rule 3. Organism is used to inoculate a healthy host of the same species and the same disease syndrome noted in rule 1 is observed.
- Rule 4. Organism is re-isolated from the inoculated plant and has the same characteristics as the initial isolate (Shurtleff and Averre, 1997).

If all of the above steps...are followed and proved true, then the isolated pathogen is identified as the organism responsible for the disease (Agrios, 1997, p. 40).

Latent Infection – Infection in a plant without visual symptoms (Shurtleff and Averre, 1997). See Latency.

Latency – Stage of an infectious disease, other than the incubation period, where no symptoms are expressed in the host (Shurtleff and Averre, 1997).

Life Cycle – Cyclical progression of stages in the growth and development of an organism (plant, animal, or pathogen) that occur between the appearance and reappearance of the same stage of the organism (Shurtleff and Averre, 1997).

Mating Types – Compatible strains, usually designated + and – or A and B, necessary for sexual reproduction in heterothallic fungi (Shurtleff and Averre, 1997).

Monocyclic – Having one cycle per growing season; no secondary infections (Shurtleff and Averre, 1997).

Mycelium – Tubular strands that make up the body of the fungal microorganism. In *Phytophthora*, mycelium is non-septate, but plugs, often called false septa, can be seen in old mycelium (Erwin and Ribeiro, 1996).

Oomycete(s) (Oomycota, Peronosporomycetes, Chromista) – A class of the Mastigomycotina, typically aquatic, saprobic, or parasitic fungi that produce oogonia, antheridia, and oospores (Shurtleff and Averre, 1997). A fungus-like chromistan that produces oospores; a water mold (Agrios, 2005, p. 895). These organisms are now classified as Peronosporomycetes and placed within the Straminipila (Abad, 2007; Dick, 2001; Dick *et al.*, 1984).

Oospore – Thick-walled, resting spore in the oomycetes that develops from a fertilized oosphere or by parthenogenesis (Shurtleff and Averre, 1997).

Pest Risk Analysis – The process of evaluating biological or other scientific and economic evidence to determine whether a pest should be regulated and the strength of any phytosanitary measures to be taken against it (IPPC, 2002).

Pest Risk Assessment – Determination of whether a pest is a quarantine pest and evaluation of its introduction potential (IPPC, 2002).

Pest Risk Management – The decision-making process of reducing the risk of introduction of a quarantine pest (IPPC, 2002).

Polycyclic – A disease of which many cycles occur in one growing season, resulting in many secondary infections (Shurtleff and Averre, 1997).

Propagule – Any part of an organism capable of initiating independent growth when separated from the parent body (Shurtleff and Averre, 1997). In the case of *P. ramorum*, propagules reported from nature are mycelia, sporangia, chlamydospores, and zoospores. Oospores have been produced in the laboratory.

Soil –The loose surface material of the earth in which plants grow, in most cases consisting of disintegrated rock with an admixture of organic material (NAPPO, 2004).

Sporangium/sporangia – Sac within which zoospores form, especially when water is cooled to about 10°C below ambient temperature. In solid substrates, sporangia usually germinate by germ tubes (Erwin and Ribeiro, 1996).

Sporulate, Sporulation – To form or produce spores (Shurtleff and Averre, 1997).

Zoospore – Spore that forms within the sporangium and exits through the terminal pore, has a tinsel and a whiplash flagellum, and is capable of swimming for several hours (Erwin and Ribeiro, 1996).

III. Pest Data Sheet

A. Identity

Name: Phytophthora ramorum Werres, de Cock, & Man in't Veld (2001)

Synonym: none

Taxonomic position: Straminipila: Peronosporomycetes (Oomycetes): Pythiales: Pythiaceae:

Phytophthora (Dick, 2001; Dick et al., 1984)

Disease names: Sudden oak death (Ramorum bleeding canker in the United Kingdom),

Ramorum leaf blight, Ramorum twig blight or dieback

B. Hosts

The host range (Table 1) for *Phytophthora ramorum* is broad and continues to expand. As of December 1, 2007, 40 plant species and all the species in five genera are designated as proven hosts, with an additional 65 species listed as associated plants by USDA (APHIS, 2007a). The difference between proven hosts and associated plants is a successful demonstration of Koch's Postulates (see Glossary).

Proven Hosts: These hosts are regulated because Koch's Postulates have been demonstrated, documented, and reviewed. The parts of the host that are regulated depend on the tissues infected by the pathogen. Damage to the timber, tourism, and nursery industries, and the environment has been documented (Davidson *et al.*, 2003b). Details for selected hosts are listed below.

<u>Caprifoliaceae</u>: The Caprifoliaceae includes important nursery and landscape species worldwide, particularly the genus *Viburnum*. One of the first hosts detected in Europe was *Viburnum x bodnantense* (Werres *et al.*, 2001). Lane *et al.* (2003) reported the first infection of *V. tinus* by *P. ramorum*. Plants displayed severe aerial dieback, stem base discoloration, and partial root decay. Flower blight has also been reported (DEFRA, 2006).

<u>Ericaceae</u>: This family encompasses another important group of nursery and landscape plants, *e.g.*, *Kalmia* spp., *Pieris* spp., and *Rhododendron* spp. (Tooley *et al.*, 2004). In addition,

members of this family are important environmental, wildland, understory, and small fruit production plants, *e.g.*, *Calluna vulgaris*, *Kalmia latifolia*, *Rhododendron* spp., and *Vaccinium* spp., respectively.

Fagaceae: This family includes a variety of forest species. Members of the red/black oak group section Lobatae, *Quercus agrifolia*, *Q. parvula* var. *shrevei*, and *Q. kelloggii* (Rizzo *et al.*, 2002a, b), although not major timber species, are important to the environment and tourism. The red/black oak group includes several important timber species on the east coast, *Q. rubra* and *Q. falcata* (Table 1), which have both been found naturally infected in Europe (Brasier *et al.*, 2004c; RAPRA, 2007). *Q. chrysolepis*, a member of section Protobalanus, is also a natural host (Davidson *et al.*, 2003a, 2003b; Murphy and Rizzo, 2003). Three species of the white oak group (section Quercus) have been found to be susceptible: *Q. ilex* (naturally infected), *Q. alba*, and *Q. robur* (experimental hosts) (Brasier *et al.*, 2002; Jones *et al.*, 2003; Tooley and Kyde, 2007). *Quercus cerris*, a member of section Cerris native to Europe, Asia, and Africa, is a natural host of *P. ramorum* (RAPRA, 2007). Another member of the Fagaceae, *Lithocarpus densiflorus*, is unique in that its stems (trunks), twigs, and foliage are susceptible. This species is very common in northern California and southern Oregon (Barrett, 2006) and is important for wildlife food and habitat (Barrett *et al.*, 2006).

<u>Pinaceae/Taxodiaceae</u>: Forest trees include important timber species, *e.g.*, *Sequoia sempervirens* (Taxodiaceae) and *Pseudotsuga menziesii* (Pinaceae). Only needles and twigs are regulated because infection in the field is limited to succulent growth (Chastagner *et al.*, 2004, 2006b; Davidson *et al.*, 2002a; Goheen *et al.*, 2006b; Maloney *et al.*, 2002a, b). Additionally, species used as Christmas trees or nursery stock, *e.g. P. menziesii* var. *menziesii*, are regulated for interstate movement (APHIS, 2007a, b).

<u>Lauraceae</u>: *Umbellularia californica* can be an important source of inoculum. Occurrence of *U. californica* is highly correlated with sudden oak death incidence in *Quercus* and *Lithocarpus* in California (Kelly and Meentemeyer, 2002; Meshriy *et al.*, 2005; Swiecki and Bernhardt, 2002a, b), but not in Oregon (Hansen *et al.*, 2005). Variation in susceptibility of *P. ramorum* has been observed in populations of *U. californica* (Meshriy *et al.*, 2006).

<u>Theaceae</u>: This family includes *Camellia* spp., which are important nursery and landscape plants. *Camellia* is regulated at the genus level because of the large number of species and hybrids determined to be hosts (APHIS, 2007b; Beales *et al.*, 2004a; Parke *et al.*, 2004a; Shishkoff, 2006). *Phytophthora ramorum*-infected *Camellia* plants have been detected in domestic and international trade (Bulluck *et al.*, 2006; RAPRA, 2007). Linderman and Davis (2007a) demonstrated that although there were variations in lesion size and sporulation among cultivars of *Camellia*, all cultivars tested were susceptible.

Associated Plants: Species symptomatic in a natural setting from which *P. ramorum* has been isolated but for which Koch's postulates have not been demonstrated, documented, and reviewed are designated as Associated Plants (Table 1). Taxa are moved from the Associated Plant List to the Proven Host List when Koch's Postulates are demonstrated and reviewed (APHIS, 2007a). Details for selected host families are listed below.

<u>Oleaceae</u>: This family contains important horticultural plants. Associated plant species in this family found naturally infected, are *Fraxinus latifolia*, *Osmanthus decorus*, *O. delavaya*, *O. fragrans*, and *O. heterophyllus*. All are foliar and shoot dieback hosts (RAPRA, 2007).

<u>Magnoliaceae</u>: Members of this family are important ornamental and forest plants. Manglietia insignis, Magnolia grandiflora, M. maudiae, M. stellata, M. ernestii, Magnolia x loebneri, Magnolia x soulangeana, and Parakmeria lotungensis are primarily foliar hosts.

Experimental Hosts: A database of experimental hosts is currently available on the Risk Analysis for *P. ramorum* website (RAPRA, 2007). Pathogenicity tests have been conducted by inoculating intact leaves, detached leaves, or both (Garbelotto *et al.*, 2003; Parke *et al.*, 2002b, 2002c, 2006a; Tooley *et al.*, 2004), log sections (Brasier *et al.*, 2002; Hansen *et al.*, 2005), and saplings (Rizzo *et al.*, 2002b; Tooley and Kyde, 2007), and by infested media (Parke *et al.*, 2006b). These screening techniques are used to predict potential hosts (Parke *et al.*, 2006a), but unless hosts are found naturally infected they will not be added to the Proven Hosts or Associated Plants List.

Table 1. Proven Hosts and Plants Associated with *Phytophthora ramorum* as listed by the United States Department of Agriculture as of December 1, 2007¹ coupled with the disease(s) and affected plant part(s).

Scientific Name	Common Name	Disease(s)	Plant Part(s) Infected	
Proven Hosts				
Aceraceae				
Acer macrophyllum	Bigleaf maple	Leaf blight	Leaf	
Acer pseudoplatanus	Planetree maple	Canker	Trunk	
Caprifoliaceae	•	•	•	
Lonicera hispidula	California honeysuckle	Leaf blight	Leaf	
Viburnum spp.	Viburnum Canker		Stem, Flower	
Ericaceae				
Arbutus menziesii	Madrone	Leaf blight, Dieback	Branch, Leaf	
Arctostaphylos manzanita	Manzanita	Leaf blight, Canker	Stem, Leaf, Twig, Branch	
Calluna vulgaris	Heath	Twig		
Kalmia spp.	Mountain laurel	Leaf, Twig		
Pieris spp.	Andromeda, Pieris	Leaf, Twig		
Rhododendron spp.	Rhododendron	Leaf blight, Dieback	Leaf, Twig, Stem	
faccinium ovatum Huckleberry		Canker, Dieback, Leaf blight	Main stem, Branch, Leaf	
Fagaceae				
Castanea sativa	Sweet chestnut	Stem necrosis or canker; Leaf blight and necrosis	Leaf, Stem	
Fagus sylvatica	European beech	Canker	Trunk	
Lithocarpus densiflorus	Tanoak	Canker, Leaf blight	Stem, Branch, Leaf	
Quercus agrifolia	Coast live oak	Canker	Stem	
Quercus cerris	European turkey oak	Canker	Trunk	
Quercus chrysolepis	Canyon live oak	Canker	Sapling, Stem	
Quercus falcate	Southern red oak	Canker	Bole	
Quercus ilex	Holm oak	Dieback	Sprout	
Quercus kelloggii	California black oak	Canker	Stem	
Quercus parvula var. shrevei	Shreve oak	Canker	Stem	
Griseliniaceae		1	I.	
Griselinia littoralis	Griselinia	Leaf necrosis	Leaf	
Hamamelidaceae				

Scientific Name	Common Name	Disease(s)	Plant Part(s) Infected	
Hamamelis virginiana	Witch hazel	Leaf blight, Dieback	Leaf, Twig	
Parrotia persica	Persian ironwood	Leaf necrosis		
Hippocastanaceae				
Aesculus californica	California buckeye	Leaf blight	Leaf, Twig	
Aesculus hippocastanum	Horse chestnut	Canker	Bole	
Lauraceae				
Laurus nobilis	Bay laurel	Leaf blight	Leaf	
Umbellularia californica	California bay laurel, Oregon myrtlewood, Pepperwood	Leaf blight	Leaf	
Liliaceae				
Maianthemum	False Solomon's seal	Loofblight	Leaf	
racemosum	Faise Solomon's seal	Leaf blight	Lear	
Magnoliaceae				
Magnolia doltsopa	Michelia	Necrosis	Leaf	
Oleaceae				
Fraxinus excelsior	European ash	Canker	Trunk	
Syringa vulgaris	Lilac	Leaf Blight	Leaf	
Pinaceae			•	
Pseudotsuga menziesii	D 1 "	D1: 1.	D1. 37 . 11 . /1 . 2	
var. menziesii	Douglas-fir	Blight	Branch, Needle (leaf)	
Primulaceae	l	1		
Trientalis latifolia	Western starflower	Blight	Leaf	
Pteridaceae				
Adiantum aleuticum	Western maidenhair fern	Leaf necrosis	Leaf	
Adiantum jordanii	California maidenhair fern	Leaf necrosis	Leaf	
Rhamnaceae		Edux Hadroom	2441	
Frangula californica	California coffeeberry	Blight	Leaf	
Frangula purshiana	Cascara	Blight	Leaf	
Rosaceae	Cascara	Diigitt	Dear	
Heteromeles arbutifolia	Toyon	Leaf blight, Dieback	Branch, Leaf	
Photinia fraseri	Red tip photinia	Leaf blight	Leaf	
Rosa gymnocarpa	Wood rose	Leaf blight	Leaf	
Salicaceae	W 000 1030	Dour origin	Deur	
Salix caprea	Goat willow	Leaf blight, Dieback	Leaf, Twig	
Тахасеае	Goat winow	Lear origin, Dieback	Lear, I wig	
Taxus baccata	European yew	Dieback	Twigs at buds	
Taxodiaceae	European yew	Dieback	1 Wigs at ouds	
Sequoia sempervirens	Coast redwood	Needle blight	Needle, Twig, Sprout	
	Coast redwood	Needle blight	Needle, Twig, Sprout	
Theaceae		1	Leaf, Petiole, Flower	
Camellia spp.	Camellia	Leaf blight; Less frequently, dieback	bud, Shoot, Twig	
	llated only as Nursery Stock)			
Aceraceae			 	
Acer circinatum	Vine maple	Leaf necrosis	Leaf	
Acer davidii	Striped bark maple	Leaf blight	Leaf	
Acer laevigatum	Evergreen maple	Chlorotic leaves, Leaf necrosis	Leaf	
Anacardiaceae		1	r	
Toxicodendron diversilobum Poison oak		Canker	Stem	
Apiaceae				
Osmorhiza berteroi	Sweet cicely	Leaf necrosis	Leaf	
Aguifaliagas			-	
Aquifoliaceae				
Ilex purpurea	Oriental holly	Leaf blight, Leaf tip dieback	Leaf	
_	Oriental holly	Leaf blight, Leaf tip dieback	Leaf	
Ilex purpurea	Oriental holly Oregon grape	Leaf blight, Leaf tip dieback Leaf blight	Leaf Leaf	

Scientific Name	Common Name	Disease(s)	Plant Part(s) Infected
Betulaceae			
Corylus cornuta	California hazelnut	Leaf blight	Leaf
Calycanthaceae			
Calycanthus occidentalis	Spicebush	Leaf necrosis	Leaf
Celastraceae			
Euonymus	Spreading euonymus	Shoot tip dieback, Leaf blight	Shoot tip, Leaf
kiautschovicus	spreading edonymus	shoot up ureaun, zeur engar	oncov up, zeur
Cornaceae			
Cornus kousa x Cornus	Cornus Norman Haddon	Shoot tip dieback	Shoot tip
capitata	Comas i (orman fraudon	Shoot up dicoden	вност пр
Dryopteridaceae			
Dryopteris arguta	California wood fern	Leaf blight	Frond
Ericaceae			
Arbutus unedo	Strawberry tree	Leaf blight	Leaf
Arctostaphylos	Manzanita	Leaf necrosis	Leaf
Columbiana		Lear necrosis	Leai
Arctostaphylos uva-ursi	Bearberry, Kinnikinnick	Leaf necrosis	Leaf
Gaultheria shallon	Salal, Oregon wintergreen	Leaf blight	Leaf
Leucothoe axillaries	Fetterbush, Dog hobble	Leaf blight	Leaf
Leucothoe fontanesiana	Drooping leucothoe	Leaf blight	Leaf
Fagaceae			
Castanopsis	G	Leaf chlorosis, Leaf necrosis (drip tip	T C C1 4
orthacanthus	Castanopsis	and mid-rib), Shoot tip die-back	Leaf, Shoot
Quercus acuta	Japanese evergreen oak	Canker	Trunk
Quercus petraea	Sessile oak	Canker	Trunk
Quercus rubra	Northern red oak	Canker	Trunk
Garryaceae			
	Silk tassel tree,		
Garrya elliptica	Coast silk tassel	Leaf necrosis	Leaf
Hamamelidaceae			
Corylopsis spicata	Spike witch hazel	Leaf necrosis	Leaf
Distylium myricoides	Myrtle-leafed distylium	Leaf blight	Leaf
Hamamelis x intermedia,			
(H. mollis & H.	Hybrid witch hazel	Leaf blight	Leaf
japonica)	11) 0114 111011 114201	Edul oligin	2441
Hamamelis mollis	Chinese witch hazel		
Loropetalum chinense	Loropetalum	Leaf blight	Leaf
Lauraceae	zoropeunum	Eval ongue	2441
Cinnamomum camphora	Camphor tree	Shoot tip die-back, Stem necrosis or canker, Leaf chlorosis	Shoot, Stem, Leaves
Liliaceae			
Clintonia andrewsiana	Andrew's clintonia bead lily	Leaf blight	Leaf
Magnoliaceae	J		
Manglietia insignis	Red lotus tree	Leaf blight, Tip dieback	Leaf, Shoot
Magnolia grandiflora	Southern magnolia	Leaf blight	Leaf
Magnolia stellata	Star magnolia	Leaf blight	Leaf
Magnolia x loebneri	Loebner magnolia	Leaf blight	Leaf
Magnolia maudiae	Michelia	Leaf blight	Leaf
Magnolia ernestii	Michelia	Leaf blight	Leaf
Magnolia x soulangeana	Saucer magnolia	Leaf chlorosis	Leaf
Parakmeria lotungensis	Eastern joy lotus tree	Leaf blight	Leaf
Myrsinaceae	Lastern joy rotus tree	Lear ongin	L-Ca1
•	Ardisia	Leaf blight	Leaf
Ardisia japonica	AIUISIä	Lear ongut	Leai
Myrtaceae	Caribble	Loofoblessia	I £
Eucalyptus haemastoma	Scribbly gum	Leaf chlorosis	Leaf
Nothofagaceae	D.11.11. C. d. 1. 1	01	T. 1
Nothofagus oblique	Roble beech, Southern beech	Canker	Trunk

Scientific Name	Common Name	Disease(s)	Plant Part(s) Infected	
Oleaceae				
Fraxinus latifolia	Oregon ash	Leaf blight	Leaf	
Osmanthus decorus	Osmanthus	Leaf blight	Leaf	
Osmanthus delavayi	Delavay osmanthus, Delavay tea olive	Shoot tip dieback, stem necrosis, or canker	Shoot, Stem	
Osmanthus fragrans	Sweet olive	Leaf necrosis, Shoot tip dieback, Stem necrosis or canker	Leaf, Shoot, Stem	
Osmanthus heterophyllus	Holly olive	Leaf necrosis	Leaf	
Pinaceae				
Abies concolor	White fir	Leaf necrosis	Leaf	
Abies grandis	Grand fir	Dieback, Leaf necrosis and bleeding canker	Branch, Needle	
Abies magnifica	Red fir	Leaf blight, tip dieback	Leaf, shoot	
Pittosporaceae				
Pittosporum undulatum	Victorian box	Leaf blight	Leaf	
Rhamnaceae				
Ceanothus thysisflorus	Blueblossom	Leaf necrosis, stem necrosis or canker, canker	Leaf, Stem, Trunk	
Rosaceae				
Physocarpus opulifolus	Ninebark	Leaf necrosis, dieback	Leaf	
Prunus laurocerasus	English laurel, Cherry laurel	Leaf necrosis	Leaf	
Prunus lusitanica	Portuguese laurel cherry	Leaf blight	Leaf	
Pyracantha koidzumii	Formosa firethorn	Leaf blight	Leaf	
Rosa (specific cultivars) Royal Bonica ("MEImodac") Pink Meidiland ("MEIpoque") Pink Sevillana ("MEIgeroka")	Hybrid roses	Leaf blight	Leaf	
Rosa rugosa	Rugosa rose	Leaf blight	Leaf	
Rubus spectabilis	Salmonberry	Leaf blight	Leaf	
Taxaceae				
Taxus brevifolia	Pacific yew	Dieback	Needle, Twig	
Taxus x media	Yew	Stem base rot	Stem	
Torreya californica	California nutmeg	Leaf necrosis, Shoot tip dieback	Leaf, Shoot	
Theaceae				
Schima wallichii	Chinese guger tree	Shoot tip dieback	Shoot	
Winteraceae		<u>-</u>		
Drimys winteri	Winter's bark	Leaf blight, Dieback	Leaf, Twig	

Most current version is posted at: http://www.aphis.gov/plant_health/plant_pest_info/pram/

C. Geographic Distribution

Asia: No record
Africa: No record
Caribbean: No record

Central America: No record

Oceania: No record

South America: No record

Europe: Belgium, the Czech Republic (eradicated, Běhalová, 2006), Denmark, Finland (imported plants only), France, Germany, Ireland, Italy, Netherlands, Norway, Poland, Slovenia, Spain (Mallorca, Islas Baleares), Sweden, Switzerland, and the United Kingdom (Steeghs, 2007).

North America:

Mexico: No record.

Canada: Infected ornamental plants in nurseries and landscape plantings have been detected and destroyed in British Columbia.

United States: Fourteen counties in California (Alameda, Contra Costa, Humboldt, Lake, Marin, Mendocino, Monterey, Napa, San Francisco, San Mateo, Santa Clara, Santa Cruz, Solano, and Sonoma) and 116 square miles in Curry County, Oregon are currently under quarantine for *P. ramorum* (7 CFR §301.92; OSOS, 2007a, b). Infected nursery stock has been detected and destroyed in the following states: Alabama, Arkansas, Arizona, California, Colorado, Connecticut, Florida, Georgia, Louisiana, Maryland, Mississippi, North Carolina, New Jersey, New Mexico, New York, Oklahoma, Oregon, Pennsylvania, South Carolina, Tennessee, Texas, Virginia, and Washington (APHIS, 2005a). Since January 10, 2005, all nursery stock shipped interstate from California, Oregon, and Washington has been regulated to prevent movement of this pathogen (APHIS, 2004a, 2007b).

Table 2. Plant genera with detections of *Phytophthora ramorum* in Europe.

Country		Reference			
Country	Nursery Outdoor Unknown				
Belgium	Rhododendron,			RAPRA,	
Beigiuiii	Viburnum			2007	
				Běhalová,	
Czech	Viburnum			2006;	
Republic	Viburnum			RAPRA,	
				2007	
Denmark	Rhododendron		Rhododendron,	RAPRA,	
Delilliark	Кпоиоиенигоп		Viburnum	2007	
				Lilja <i>et al</i> .,	
				2007;	
Finland	Rhododendron		Rhododendron	RAPRA,	
Tillialiu	Knouodenaron		Knoaoaenaron	2007;	
				Rythönen et	
				al., 2007	
				RAPRA,	
France	Camellia, Pieris,			2007	

Rhododendron,

Country	Detection Location								
Country	Nursery	Outdoor	Unknown	Reference					
	Viburnum								
Germany	Rhododendron, Viburnum	Pieris, Rhododendron	Rhododendron, Viburnum						
Ireland	Rhododendron		Rhododendron, Viburnum						
Italy	Rhododendron								
Netherlands	Rhododendron, Taxus, Viburnum	Fagus, Quercus, Rhododendron	Viburnum						
Norway	Rhododendron	Rhododendron, Viburnum]					
Poland	Calluna, Photinia, Pieris, Rhododendron								
Slovenia	Kalmia]					
Spain	Arbutus, Camellia, Rhododendron, Viburnum		Aesculus, Arbutus, Camellia, Rhododendron, Syringa, Taxus, Viburnum						
Sweden	Rhododendron		Rhododendron						
Switzerland	Rhododendron, Viburnum	Viburnum							
United Kingdom	Camellia, Garrya, Grisellinia, Hamamelis, Kalmia, Laurus, Leucothoe, Magnolia, Osmanthus, Parrotia, Pieris, Rhododendron, Syringa, Taxus, Viburnum	Acer, Aesculus, Castanea, Castanopsis, Cinnamomum, Cornus, Cydonia, Drimys, Eucalyptus, Fagus, Fraxinus, Grisellinia, Hamamelis, Kalmia, Laurus, Magnolia, Michelia, Notofagus, Pieris, Quercus, Rhododendron, Schima, Syringa, Umbellularia, Viburnum	Arbutus, Camellia, Hamamelis, Kalmia, Leucothoe, Lonicera, Magnolia, Pieris, Quercus, Rhododendron, Salix, Syringa, Taxus, Viburnum						

D. Biology and Epidemiology

The disease cycle associated with *P.ramorum* (Fig. 1) is complex because of the variety of habitats where the pathogen occurs, the diversity of plants attacked, and the variation in plant response to infection (Davidson *et al.*, 2003c). *Phytophthora ramorum* incites multiple diseases, with host dependent symptomology: bleeding canker (sudden oak death), *e.g.*, on several members of Fagaceae; ramorum leaf blight, *e.g.*, on *U. californica*; and ramorum dieback, *e.g.*, on *Q. ilex*. Wilt symptoms have been observed on shoot tips of various hosts of *P. ramorum* and Parke *et al.* (2007) recently demonstrated a possible mechanism for a vascular wilt disease in *L. densiflorus*.

Phytophthora ramorum produces sporangia, zoospores, and chlamydospores in culture and in nature (Davidson *et al.*, 2003c; Parke *et al.*, 2002a; Werres *et al.*, 2001), and oospores under laboratory conditions (Boutet and Chandelier, 2007; Werres and Zielke, 2003). Sporangia are semi-papillate, caducous, and range in length from 20-80 μm (Rizzo *et al.*, 2002b; Werres *et al.*, 2001). Chlamydospores are produced on hyphal tips and are hyaline, becoming brown with age and when produced on host tissue (Rizzo *et al.*, 2002b; Werres *et al.*, 2001). Chlamydospores range in size from 40-80 μm (Rizzo *et al.*, 2002b) and 20-91 μm (Werres *et al.*, 2001). Hyphae

of this species are nodose, highly branched, contorted, and form a dendritic pattern. *Phytophthora ramorum* is a poor saprophytic competitor (Rizzo *et al.*, 2002b).

Phytophthora ramorum is a heterothallic organism with two mating types, A1 and A2 (Werres et al., 2001). Originally, A1 isolates were found only in Europe (Werres et al., 2001) and A2 isolates only in the United States (Rizzo et al., 2002b). The two mating types coincided with genetic differences and were determined to be distinct populations (Brasier, 2003; Brasier et al., 2003; 2006a, b; Kroon et al., 2004). In 2003, an A2 isolate that matched the European population was detected on imported European nursery stock in Belgium (Werres and De Merlier, 2003). Also in 2003, A1 isolates were detected on nursery stock in Oregon, Washington, and British Columbia that matched the European A1 population (Hansen et al., 2003a).

Ivors *et al.* (2006) identified three lineages of *P. ramorum*, one from Europe and two from North America. These three lineages are based on microsatellite profiles and designated EU1, NA1, and NA2; the names correspond to the continent where the lineage was originally found (Table 3; COMTF, 2007). The EU1 lineage, originally found in Europe, consists predominantly of A1 isolates but also contains three A2 isolates from Belgium nurseries (RAPRA, 2007). The NA1 lineage consists of A2 isolates that were detected in forests in California and Oregon or in nurseries in the United States and Canada. The NA2 lineage is rare and consists of A2 isolates. These NA2 isolates were found in or traced to nurseries in Washington and California. These different lineages have limited molecular variation, suggesting that they were introduced separately from a more variable original population (Ivors *et al.*, 2006).

Table 3. Summary of characteristics of the three lineages of *Phytophthora ramorum*.

Lineage	Mating Type	Location
NA1	A2	Forests in California and Oregon; Nurseries in U.S. and Canada
NA2	A2	Nurseries in North America
EU1	A1	Nurseries and wildlands in Europe; Nurseries in U.S. and Canada
EU1	A2	Nurseries in Belgium

Oospores have not been detected in nature, but have been observed in culture when *P. ramorum* strains are paired with other *Phytophthora* species representing opposite mating types (Boutet and Chandelier, 2007; Brasier and Kirk, 2004; Brasier *et al.*, 2006a, b; Rizzo *et al.*, 2002b; Werres *et al.*, 2001). Boutet and Chandelier (2007) reported that gelling qualities of culture media and genotype influenced the formation of gametangia. A European A1 strain producing very few chlamydospores was found to be a better mating partner than other A1 strains. This research suggests that these oospores are the result of selfing and not hybridization between mating partners (Boutet and Chandelier, 2007). Oospores were reported on hyphae produced from a pairing of U.S. isolate PR6-2 with EU isolate BBA 9/95 on green *Rhododendron* twigs (Zielke and Werres, 2002).

In culture, *P. ramorum* had optimum growth at 20°C (Werres *et al.*, 2001), reduced growth at -1°C, and did not survive at -25°C (DEFRA, 2004c). However, one North American A2 isolate was found to grow optimally at 25°C (DEFRA, 2004c).

There are a number of studies on infection by *P. ramorum* (detached leaves, stems, roots, plants, and log segments). For example, detached leaf assays of *Rhododendron* found a positive correlation between lesion development and number of degree-days; the maximum temperature tested, 25°C, resulted in the largest lesions (DEFRA, 2004c). Garbelotto *et al.* (2003) found that 9-12 hours of leaf wetness at 18-22°C are necessary to obtain significant infections on *U. californica* leaves. Brasier *et al.* (2007) demonstrated infection by zoospores through intact bark on log segments of *F. sylvatica*, *Q. robur*, and *A. pseudoplatanus*. Parke and Lewis (2007) observed *P. ramorum* penetrating *Rhododendron* roots at primordia, emerging laterals, and wound sites. They also noted that *P. ramorum* did not need stomata to infect leaves and that infections near the midrib resulted in more rapid disease development than infections at other leaf sites.

Hosts of *P. ramorum* usually fall into one of two disease categories based on the plant part infected: "canker hosts" or "leaf and twig hosts" (Davidson *et al.*, 2003b). The pathogen is polycyclic (Fig. 1) on most leaf and twig hosts (Davidson *et al.*, 2003a, b, 2005). Infections in leaf and twig hosts are rarely fatal, but they can serve as a reservoir of the pathogen and a source of inoculum (DEFRA, 2004c; Parke *et al.*, 2002b, c; Rizzo *et al.*, 2002b). Sporangia and chlamydospores are produced abundantly on several foliar and dieback hosts, including *U. californica* (Davidson *et al.*, 2002b), *Rhododendron*, and *K. latifolia* (DEFRA, 2004c). Differences in sporulation ability and susceptibility to infection have been reported for foliar and dieback hosts (DEFRA, 2004c; Dodd *et al.*, 2002; Hansen *et al.*, 2005; Hüberli *et al.*, 2002; Linderman and Davis, 2007a; Parke *et al.*, 2002a, b, c, 2006a; Tooley and Kyde, 2007; Tooley *et al.*, 2004).

In field tests, chlamydospores within host material were shown to overwinter down to -9°C in the United Kingdom (DEFRA, 2004c) and to oversummer in California (Fichtner *et al.*, 2004, 2006b, 2007a). Chlamydospore survival increased with depth of burial in both studies (DEFRA, 2004c; Fichtner *et al.*, 2006b).

Canker hosts exhibit infections on basal stems (trunks of trees, stems of *Viburnum*) and often die. Sporulation was not observed on canker surfaces of these hosts (Davidson *et al.*, 2003b, c), although exudates have tested positive with PCR (Tjosvold *et al.*, 2002a). However, if the inner bark (cambium) is exposed and free water is present, the pathogen can sporulate on exposed surfaces (Davidson *et al.*, 2003b, c). The pathogen has been recovered from inner bark (Davidson *et al.*, 2003b), wood chips (Davidson *et al.*, 2003b; Shelly *et al.*, 2005b), and firewood stored for six months (Shelly *et al.*, 2005a). Sporulation was stimulated in baiting trials when inoculated "logs" were kept at 12°C prior to baiting (Garbelotto, 2002). More recent studies have demonstrated that *P. ramorum* can occupy the xylem beneath phloem lesions, perennate in xylem tissue, and spread in xylem tissue ahead of phloem lesions (Brown and Brasier, 2007; Parke *et al.*, 2007).

The disease incidence of sudden oak death in California and Oregon is clustered. Spatial analysis in California indicated that diseased plants were clustered within 100 and 300 m of each other (Meentemeyer and Kelly, 2002). Disease incidence was correlated with proximity to forest edge, potential topographic moisture, abundance of *U. californica*, and potential solar radiation. However, Condeso and Meentemeyer (2007) found that elevation, temperature, and amount of

contiguous forest were correlated with disease incidence. In addition, the temperature range correlated with the highest disease incidence, 0-10°C, was lower than the optimal range observed for zoospore production (15-20°C) under laboratory conditions by Davidson *et al.* (2005). Long-distance dispersal includes movement of infected plant material (wood, green material products, and nursery stock), soil, water (rain, runoff, streams, rivers, irrigation water) (Davidson *et al.*, 2002b, c), animals, and aerial dissemination (of sporangia, zoospores and possibly chlamydospores) during major weather events. It is postulated that long-distance dispersal through aerial dissemination is responsible for spread of the NA1-A2 mating type in California and Oregon (Hansen *et al.*, 2002).

E. Detection and Identification

Symptoms

Different diseases are attributed to *P. ramorum*: sudden oak death, stem or bole cankers, ramorum dieback, twig blight, and ramorum leaf blight (Table 1). Wilt symptoms have been observed on shoot tips of various hosts of *P. ramorum* (Parke *et al.*, 2007; Storer *et al.*, 2002). Symptomology has been addressed by Davidson *et al.* (2003b); Garbelotto *et al.* (2002a, 2003); Goheen *et al.* (2006b); Parke *et al.* (2003, 2004b); Storer *et al.* (2002); and Tjosvold *et al.* (2004).

Prominent hosts in the nursery trade include *Rhododendron*, *Camellia*, *Pieris*, and *Viburnum*. Symptoms on *Rhododendron* mirror those incited by other species of *Phytophthora* and by certain environmental factors. This makes inspection for the disease more complicated (Davidson and Shaw, 2003) and detection more challenging (Jones *et al.*, 2003).

With *Lithocarpus* species, drooping or wilting of new growth may occur before other symptoms appear (Storer *et al.*, 2002). Parke *et al.* (2007) recently demonstrated a possible mechanism for a vascular wilt disease in *L. densiflorus*. Cankers typically occur in the lower 3 meters and are restricted to above the soil line. Occasionally cankers have been found 20 meters above ground. Earlier research suggested that cankers girdled the tree, resulting in death. Current research indicates that infection by *P. ramorum* caused vessel blockage, resulting in wilt, and potentially, in tree mortality (Parke *et al.*, 2007). Bleeding (oozing) symptoms of the canker are easier to detect during dry weather and become more difficult to detect during the rainy season, when the ooze is washed off.

New *Phytophthora* species were described as a result of field analyses and surveys for *P. ramorum*: *P. nemorosa* E. M. Hansen and Reeser and *P. kernoviae* Brasier, Beales & S. A. Kirk (2005). Species new to the United States have also been found: *P. hedraiandra* and *P. pseudosyringae*. *Phytophthora nemorosa* and *P. pseudosyringae* occupy a similar ecological niche to *P. ramorum* in the United States (Hansen *et al.*, 2003b) and *P. kernoviae* a similar niche in the United Kingdom (Brown and Brasier, 2007; DEFRA, 2004c, 2005a). *Phytophthora hedraiandra* was found on *Viburnum tinus* during nursery surveys in Minnesota (Schwingle *et al.*, 2007), from *Viburnum* in the Netherlands (de Cock and Lévesque, 2004), and from *V. tinus* in Spain (Moralejo *et al.*, 2006). These closely related species occupy the same niches and cause similar symptoms, thus confusing *P. ramorum* detection.

Isolation, Detection and Characterization

Phytophthora ramorum can be isolated directly or indirectly (baiting with pear fruit or host leaves) from infected host material, soil, and water (Davidson et al., 2002a, 2003b; Goheen et al., 2002c; Maloney et al., 2002a; Rizzo et al., 2002a, b; Werres et al., 2001). Recovery rates vary with season and host (Davidson and Shaw, 2003; Hayden et al., 2004), and are facilitated with the use of the selective medium PARP (Davidson et al., 2003b). Additionally, preliminary results indicate that exposure of infected woody material to a cool temperature, 12°C (Garbelotto, 2002), and plating the samples on PARP immediately following collection in the field (Storer et al., 2002) will facilitate recovery of the pathogen. Samples are incubated in the dark at 20° to 22°C and examined within seven days.

Morphological and molecular comparisons of U.S. and European isolates indicate that the two mating types are the same species (Ivors *et al.*, 2004; Man in't Veld *et al.*, 2002; Zielke and Werres, 2002). Pogoda and Werres (2002) found that growth rate and colony morphology were related to aggressiveness. Slow vegetative growth, exhibited by many U.S. isolates, was correlated with mild twig infection. Although the European isolates have greater genetic diversity than the U.S. isolates, they are more phenotypically similar.

Polymerase chain reaction (PCR) methods are used for the detection and identification of this pathogen (Hayden *et al.*, 2004; Martin and Tooley, 2001; Martin *et al.*, 2002, 2004). Hayden *et al.* (2004) found PCR and isolation frequency varied with season and host, but PCR detection was more sensitive than isolation. Maloney *et al.* (2004) first detected *P. ramorum* in madrone by PCR and later were able to isolate the pathogen. *Arctostaphylos manzanita* was found to be positive by PCR (Rizzo *et al.*, 2002a) long before isolation attempts were successful.

Molecular analysis found that 83 isolates (65 U.S., 18 European) were identical at three DNA regions (ITS, *cox* II, and *nad* 5) (Ivors *et al.*, 2004). Amplified Fragment Length Polymorphism (AFLP) analysis indicated that a single clonal lineage dominated the U.S. isolates. Two U.S. isolates from an Oregon nursery differed at those regions. Microsatellite analysis of over 200 isolates revealed seven loci that discriminated between U.S. and European isolates (Prospero *et al.*, 2005, 2007). Microsatellite analysis of 151 isolates of *P. ramorum* revealed three distinct clades: the U.S. population, the European population, and one unique population (Ivors *et al.*, 2006).

Two molecular detection methods have been validated by USDA APHIS (CPHST, Beltsville, MD) for use in regulatory determinations of *P. ramorum*. The APHIS nested PCR protocol is based on Hayden *et al.* (2004), with a multiplex PCR quality control component from Winton and Hansen (2001). APHIS also uses a real-time PCR protocol based on Hughes *et al.* (2006). Nested PCR occasionally has cross-reactions with *P. lateralis* (Blomquist and Kubisiak, 2003) and *P. foliorum* (Donahoo *et al.*, 2006). Safeguards are included in the USDA protocol to identify and prevent misdiagnosis. New procedures are being evaluated for inclusion for use in the regulatory program, including work by Schena *et al.* (2006), Martin *et al.* (2004) and Bilodeau *et al.* (2007).

ELISA can be used to detect species of *Phytophthora* (Brown and Brasier, 2007; Bulluck *et al.*, 2007). This method may be used to facilitate the processing of large number of samples and is a part of the USDA protocol (APHIS, 2006). All ELISA positive samples must be tested by

approved PCR methods to confirm the presence of *P. ramorum*. Approved PCR methods can be run by the Molecular Diagnostic Laboratory, which is part of the National Identification Services, or by the laboratories approved by the National Plant Protection Laboratory Approval Program.

Monitoring

The National Phytophthora ramorum Survey, a nursery inspection program, ended in 2006; however, nursery surveys continue under a Federal Order (effective January 10, 2005) restricting movement of infected plants from California, Oregon, and Washington. This order also required that both host and non-host nurseries be inspected to move nursery stock interstate and that traceforward and trace-back activities be conducted once positive nursery material is detected.

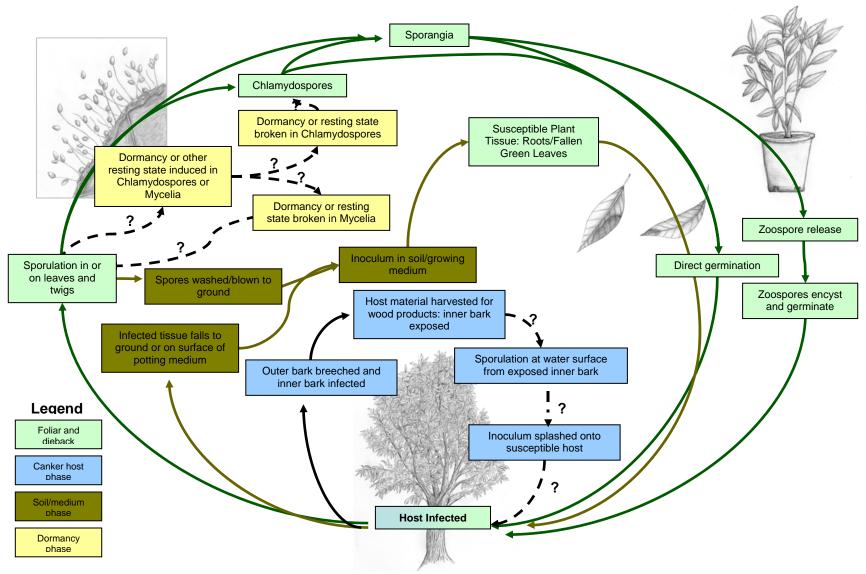


Fig. 1. Possible disease cycle for sudden oak death, ramorum leaf blight, and ramorum dieback. Color is used to designate different hosts and phases.

In addition, individual States may continue surveys through the USDA Cooperative Agricultural Pest Survey (CAPS) program. The USDA Forest Service continues to conduct a national survey program of forests and streams with a focus on areas adjacent to nurseries (http://fhm.fs.fed.us/sp/sod/sod.shtm).

Aerial surveys of forests are conducted annually to survey for damaged *L. densiflorus* in Oregon (Anon., 2007; Goheen *et al.*, 2002d) and for *Quercus* spp. and *L. densiflorus* in California (Bell and Fischer, 2006; Levien *et al.*, 2002). Maps and additional data can be found at:

http://www.oregon.gov/ODF/PRIVATE_FORESTS/fh.shtml (Oregon Reports) http://www.fs.fed.us/r6/nr/fid/data.shtml (Oregon data and maps) http://www.fs.fed.us/r5/spf/fhp/fhm/sod/index.shtml (California) http://kellylab.berkeley.edu/SODmonitoring/ (California)

Airborne Digital Acquisition and Registration (ADAR) imagery based on red, green, blue, and near-infrared wavelengths was tested for capability to map species (Kelly and Meentemeyer, 2002). Results were variable, but more promising for species mapping than for locating moisture-stressed trees. The USDA Forest Service currently uses an advanced digital sketching mapping system for aerial surveys (Anon., 2007).

Aerial surveys are used, in conjunction with risk models, *e.g.*, Meentemeyer *et al* (2004), to target areas for ground surveys (Bell and Fischer, 2006). Field visits resulted in ten new *P. ramorum* detections. The positive detection in southern Monterey County in the Willow Creek watershed is the farthest south the pathogen has been found to date. This watershed was targeted for field survey because a stream bait tested positive for *P. ramorum* in 2005.

In 2006, a pilot survey was conducted to evaluate existing stream baiting and lab diagnostic methods for inclusion in the 2007 national *P. ramorum* early detection survey protocol. Streams in eleven States were surveyed, including ramorum-endemic (CA, OR), nursery-confirmed or nursery-introduced (GA, MD, NC, PA, TN, VA, WA), and states where *P. ramorum* has yet to be detected (KY, WV) (Oak *et al.*, 2007).

IV. Organism Risk Assessment

A. Prior Risk Assessments, Current Status, and Interceptions

On February 14, 2002, regulations were published to control the movement of *P. ramorum* from twelve infested counties in California and an area under eradication in Oregon. Various agencies within USDA (Agriculture Research Service, Forest Service, and Animal and Plant Health Inspection Service), universities and other institutions continue to conduct research and developmental studies to better identify hosts, methods of detection, and effective treatments. There are no chemical treatments currently available to eliminate the pathogen in nursery stock. In April 2004, a Federal Order was issued to address a concern of *P. ramorum* moving via nursery stock from California, Oregon, and Washington. On December 21, 2004, APHIS issued an emergency Federal Order that bolstered the Agency's initial *P. ramorum* restrictions by regulating the interstate movement of plants for planting, including houseplants and propagative

materials, from all commercial nurseries in California, Oregon, and Washington. The Federal Order, which became effective on January 10, 2005, was enacted in response to detections of *P. ramorum* at commercial nurseries in California, Oregon, and Washington that are outside quarantined areas, and addresses a number of concerns regarding the adequacy of previous Federal *P. ramorum* restrictions.

The USDA Forest Service conducted a risk assessment in 2001 with revisions in 2003, and 2005 (Kliejunas, 2001, 2003, 2005). Other risk analyses have been produced by the Canadian Food Inspection Agency (Cree *et al.*, 2001; Cree, 2002; Rioux *et al.*, 2006), the UK Department of Environment, Forestry and Rural Affairs (Jones, 2002; Jones *et al.*, 2003), the Oregon Department of Agriculture (ODA, 2001) and USDA APHIS (Cave *et al.*, 2005). Currently, a PRA is under development by members of the European Union (RAPRA, 2007).

Phytophthora spp. are difficult to detect by visual inspection, because disease symptoms are not always distinctive and the defining characteristics of the disease are not visible to the naked eye. There have been 12 interceptions at U.S. ports since January 1, 1985; of these, four were identified to species (none were *P. ramorum*).

B. Consequences of Introduction

This portion of the assessment considers negative outcomes that may occur when the hosts of *P. ramorum* provide a pathway of entry into the United States from infested countries as well as domestic movement of infested plant material. The potential consequences are evaluated using five Risk Elements (APHIS, 2002): Climate-Host Interaction, Host Range, Dispersal Potential, Economic Impact, and Environmental Impact. These risk elements reflect the biology, host range, and climatic and geographic distribution of this pathogen, and are supported by biological information. For each risk element, a rating of Low, Medium, or High is assigned (APHIS, 2002). Additionally, specific pathways, *i.e.*, plants for planting, wood, soil, potting media, cut flowers/foliage, greenwaste, and compost, will be evaluated using these Elements.

Risk Element 1: Climate-Host Interaction

This risk element considers ecological zonation and the interactions of *P. ramorum* with its hosts in a variety of environments. When introduced into new areas, pests are expected to behave as they do in their native areas if the potential host plants and suitable climate are present. Broad availability of suitable climates and a wide distribution of suitable hosts are assumed to increase the impact of a pest introduction. The rating for this risk element is based on the number of United States Plant Hardiness Zones (USDA, 2003).

Phytophthora ramorum has a high probability of encountering favorable climatic conditions throughout the ranges of potential hosts. Modeling of environmental conditions suggests that there are many areas in the United States which have both favorable conditions for disease development and susceptible hosts (Linderman et al., 2007; Magarey et al., 2004, 2007; Smith et al., 2002) (Fig. 2). Climate potential was higher on the West Coast and east of the Mississippi River than in the Central Plains. The risk to the more arid Central Plains states increases when humid microclimates, such as in plant nurseries or irrigated landscapes, are created. This

occurred during 2003 and 2004 in California when nurseries outside the quarantine zone and in a warmer and more arid environment shipped infected nursery stock (Magarey *et al.*, 2004).

The risk rating is High for the Climate-Host Interaction Risk Element. The level of certainty for this risk rating is fairly certain. Most of the eastern United States has actual and potential hosts growing in climates conducive to infection. The uncertainty lies in the range of biotic and abiotic factors triggering establishment of *P. ramorum* in new areas.

Risk Element 2: Host Range

The risk posed by a plant pest depends on both its ability to establish a viable, reproductive population and its potential to damage plants. This risk element assumes that the consequences of pest introduction are positively correlated with the pest's host range. Aggressiveness, virulence, and pathogenicity also may be factors. The consequences related to host range are rated in accordance with the ability of the pathogen to attack a single species or multiple species within a single genus, a single plant family, or multiple families.

The host range of this pathogen continues to expand through detections in the field. APHIS currently regulates over 109 plants in 35 families and 70 genera (Table 1). The potential host range is also increasing (APHIS, 2007b; DEFRA, 2006; Hansen *et al.*, 2005). Experimental evidence demonstrates that several eastern forest species would be more susceptible than western forest species. In addition, differences in host susceptibility are documented for forest and nursery species and may impact disease development in new environments (DEFRA, 2004c; Meshriy *et al.*, 2005; Tooley *et al.*, 2004).

Brasier *et al.* (2002) screened several forest species by inoculating the inner bark of logs with U.S. and European *P. ramorum* isolates. This study suggested the most susceptible species in the United Kingdom are *Q. rubra*, *Q. cerris*, *Q. ilex*, *F. sylvatica*, *C. sativa*, *P. sitchensis*, *P. menziesii* var. *menziesii*, and *C. lawsonia*. Since this study, several of those species have been found naturally infected by *P. ramorum* during surveys in Europe: *Q. rubra* in the Netherlands, and *Q. falcata*, *Q. ilex*, *Q. cerris*, *F. sylvatica*, and *A. hippocastanum* in the United Kingdom (DEFRA, 2006).

Tree species in the red oak/black oak group appear to be highly susceptible to *P. ramorum*. Greenhouse studies have compared susceptibility of regulated *Quercus* species to non-regulated *Quercus* species. Based on adjusted lesion area, two- to three-year old seedlings of *Q. rubra*, *Q. montana* (syn. *Q. prinus*), and *Q. pagoda* were found to be more susceptible to *P. ramorum* than the regulated host, *Q. agrifolia* (Tooley and Kyde, 2007). *Quercus phellos*, *Q. nigra*, *Q. virginiana*, and *Juglans nigra* were equally susceptible and *A. saccharum* was less susceptible than *Q. agrifolia* (Tooley and Kyde, 2007). In foliar inoculations, *Q. montana* (*Q. prinus*) was more susceptible that *L. densiflorus*; other *Quercus* species were significantly less susceptible (Tooley and Kyde 2007).

Certain white oak species (*Q. douglasii*, *Q. lobata*, and *Q. robur*) are not as susceptible to *P. ramorum* as red oak species (Brasier *et al.*, 2002; Rizzo *et al.*, 2002a). Lesions on young white oak trees were similar in size to those on the wounded non-inoculated trees. However,

two- to three-year old seedlings of *Q. alba* were more susceptible to *P. ramorum* than those of the red oak species, *Q. agrifolia* (Tooley and Kyde, 2007).

The large number of hosts in multiple plant families, differential susceptibility, and virulence warrant a risk rating for Host Range of High. The level of certainty for this risk rating is High. *Phytophthora ramorum* already has a large documented host range. The uncertainty for the rating for this element lies in not knowing the extent of the host range.

Risk Element 3: Dispersal Potential

Pests may disperse after introduction into new areas. The dispersal potential indicates how rapidly and widely the pests may spread. This factor is related to the pest's reproductive potential, inherent mobility, and external dispersal facilitation modes within the importing country or region. Factors for rating the dispersal potential include: the presence of multiple generations per year or growing season, the relative number of offspring or propagules per generation, any inherent capabilities for rapid movement, the presence of natural barriers or enemies, and dissemination enhanced by wind, water, vectors, or human assistance.

The scattered pattern of sites where *P. ramorum* has become established suggests that it has a mechanism of long-distance dispersal. Strong winds common during heavy rains along the California coast may move the easily detached sporangia great distances (Hansen *et al.*, 2002). Initial survey results in California and Oregon indicate *P. ramorum* is in streams and rivers adjacent to and far from known infested areas (Murphy *et al.*, 2006; Hansen *et al.*, 2005).

Inoculum has been detected seasonally from soil on hiking trails and from soil on hikers' boots (Davidson *et al.*, 2002c, 2005; Tjosvold *et al.*, 2002b). The concerns about soil and litter movement by equipment have prompted California authorities to request that vehicles and other equipment, including tents and shoes, be washed prior to leaving a *P. ramorum*-infested area (COMTF, 2004a).

In 2004, confirmed positive sites from the trace-forward, national, and other surveys totaled 176 in 22 States (APHIS, 2005b, c). The total included three residential finds (Georgia, South Carolina), and one detection (PCR only) in the environs (New York) which prompted repeated testing. This area was finally released in 2007 (DA-2007-03, February 15, 2007). As of January 10, 2005, all nursery stock shipped interstate from California, Oregon, and Washington is regulated to prevent movement of this pathogen (APHIS, 2007b). In 2004, 665 Christmas tree plantations in Oregon and 100 in Washington were surveyed and found negative for *P. ramorum* (COMTF, 2004b).

Many of the hosts on the regulated host and associated plants lists are major nursery, forest, and understory species (Davidson *et al.*, 2003b), and the host range is expanding. Evidence exists that several eastern forest species would be as susceptible as those affected in California and Oregon. Additionally, environmental conditions in areas in the eastern United States are predicted to be more conducive to disease development than in the majority of the western United States (Magarey *et al.*, 2004, 2007).

Newly established populations may go undetected for years. The disease was first noted in California in 1995 (Garbelotto *et al.*, 2001). With the rate of oak death, researchers suggest that the pathogen was introduced at least five years before the first detection (Rizzo and Garbelotto, 2003).

In the United States, both regulated and associated hosts are widely distributed, overlapping, abundant, and susceptible. In addition, the pathogen is polycyclic; infections may remain undetected for years. Long-distance dispersal has been documented via trade and natural means. For these reasons, the rating for *P. ramorum* is High for Dispersal Potential. The level of uncertainty for this rating is low based on the evidence of human-assisted and natural movement.

Risk Element 4: Economic Impact

Introduced pests cause a variety of direct and indirect economic impacts, such as reduced yield, reduced commodity value, loss of foreign or domestic markets, and non-crop impacts. Factors considered during the ranking process included the following: effect on yield or commodity quality; plant mortality; ability to act as a disease vector; increased costs of production, including pest control costs, lower market prices, effects on market availability, increased research or extension costs, or reduction in recreational land use or aesthetic value; ability of the pest to attack the hosts or products with significant commercial value, to directly cause tree mortality, or to predispose the host to mortality by other organisms; impact of the pest on the value of the affected host (*e.g.*, by lowering its market price, increasing cost of production, maintenance, or mitigation, or reducing value of property where it is located); and lack of effective control measures.

The USDA had spent more than \$55 million by the end of 2005 on regulatory, research, and educational issues related to *P. ramorum* (Table 4). This value does include state and local government and industry expenditures.

California's oak woodlands contain about 5 billion cubic feet of wood valued at over \$275 million (Kliejunas, 2003). The nearby California timberlands contain 5.8 billion cubic feet of oaks, which are worth over \$500 million for forest products alone (Kliejunas, 2003). Oak products exported from California from 1996-2000 averaged almost \$50 million per year (USITC, 2005).

Phytophthora ramorum presents a potential economic threat to eastern U.S. oaks. Two oak species native to the eastern United States, *Q. rubra* and *Q. falcata*, were found naturally infected in Europe (Brasier et al., 2004b; EPPO, 2004). Susceptibility of other eastern U.S. tree species (*Q. alba*, *Q. laurifolia*, *Q. nigra*, *Q. pagoda*, *Q. phellos*, *Q. montana* (syn. *Q. prinus*), *Q. virginiana*, *A. saccharum*, and *J. nigra*) has been experimentally demonstrated (Brasier et al., 2002; Linderman et al., 2007; Tooley and Kyde, 2007) and represents a potential economic threat to commercial timber production in the United States exceeding \$30 billion (Kliejunas, 2003). The export value of red oak logs and lumber was over \$300 million dollars in 2002 (USITC, 2005).

In coastal central California, oak woodland suitable for residential development has been estimated at \$20,000 per acre; rangeland with at least 40 oaks per acre was worth 27 percent more than open land (Standiford, 2000). In southwestern Oregon, mature black oak trees can increase property values by \$5,000-30,000 (Osterbauer, 2003).

Current regulations require debarking of the logs in order to send them to pulp mills outside quarantine areas (COMTF, 2003). Hardwood hosts are used for firewood, wood chips for pulping, compost, non-grade lumber, and charcoal. Higher value uses include custom furniture, flooring, cooperage, and tool handles (Shelly *et al.*, 1996).

The U.S. nursery industry is also at risk. Nursery crops include woody perennial plants, such as ornamental trees, shrubs, and vines, which are primarily used for landscaping. In 2006, the U.S. domestic production of nursery crops was valued about \$12.9 billion. Imports for these crops were \$341 million and exports were \$287 million (Jerado, 2007).

Tourism is also affected, as visitors to parks and forests may find that access to selected areas is restricted during certain seasons to prevent movement of the pathogen or to protect visitors from falling limbs from trees killed by *P. ramorum*. When visitors are requested or required to take precautions to prevent its movement, park and forest staff may be required to provide educational information, staff cleaning areas, and provide appropriate supplies and equipment to remove soil from shoes and vehicles (COMTF, 2004a).

The presence of *P. ramorum* has resulted in restrictions in foreign and domestic trade. Australia, Canada, Korea, New Zealand, the European Union, and Switzerland have placed restrictions on the movement of affected plants and plant parts from the United States (EXCERPT, 2007; Rizzo and Garbelotto, 2003). In addition, the United States has placed restrictions on the movement of propagative material from the European Union (Aley, 2007).

The evidence to date is that *P. ramorum* impacts the domestic movement of plants and plant products (nursery stock, fruit, logs, lumber, *etc.*) and has restricted international trade. For these reasons, the economic impact of *P. ramorum* is rated High. Uncertainty depends on the relationships between the extent of the host range and the value of these plants on the open market.



Fig. 2. Overall risk index for the establishment of *Phytophthora ramorum* in the continental United States based on introduction potential (includes number of plants shipped from an infested nursery), climate potential (leaf wetness, temperatures, and relative humidity based on 30-year averages), economic potential, and host strength (quantity and diversity of potential hosts) (Magarey *et al.*, 2004).

Table 4. Summary of USDA funding for *Phytophthora ramorum*, Fiscal Years 2000-2005.

	Funding, by USDA agency (Dollars in Millions)										
Fiscal year	Forest Service	Animal and Plant Health Inspection Service	Agricultural Research Service	Cooperative State Research, Education and Extension Service	Total						
2000	\$0.12	\$0.00	\$0.00	\$0.00	\$0.12						
2001	4.20	0.00	0.00	0.00	4.20						
2002	0.97	0.90	0.00	0.00	1.87						
2003	3.70	2.00	0.62	0.30	6.62						
2004	3.70	19.50	1.30	0.30	24.80						
2005	4.40	12.40	1.00	0.12	17.92						
Total	\$17.09	\$34.8	\$2.92	\$0.72	\$55.53						

Source: United States Government Accountability Office Report to the Chairman, Committee on Resources, House of Representatives report on Invasive Forest Pests, April, 2006, Page 109 (http://www.gao.gov/new.items/d06353.pdf)

Risk Element 5: Environmental Impact

The ratings for this risk element are based on three aspects: the potential of the pest to disrupt native ecosystems and habitats exhibited within its current geographic range, the need for additional chemical or biological control programs due to the presence of the pest, and the potential of the pest to directly or indirectly impact species listed as Threatened or Endangered (50 CFR §17.11-12) by infesting or infecting a listed plant. When a pest is known to infest or infect other species within the same genus, and host specificity data does not exist for the listed plant, then the listed plant is assumed to be a potential host.

In forests, more than 20 non-indigenous species of plant pathogens attack woody plants (Liebhold *et al.*, 1995). Two of the most destructive plant pathogens are *Cryphonectria parasitica* and *Ophiostoma ulmi*, the causal agents of chestnut blight and Dutch elm disease, respectively. Before the introduction of chestnut blight, approximately 25% of eastern U.S. deciduous forest consisted of American chestnut trees (*Castanea dentata*) (Liebhold *et al.*, 1995). These trees have all but disappeared. In urban and forest environments, species and cultivars of *Ulmus* have been destroyed by *O. ulmi*. The environmental costs of prevention, eradication, or suppression of this pathogen include indirect ecological consequences (perturbations of hydrological cycles, *e.g.*, flood control and water supply, waste assimilation, nutrient recycling, conservation and regeneration of soils, crop pollination) and must address both current-use value and future-use values.

Quercus species are the most important and widespread of the hardwood trees in the North Temperate Zone (Pavlik et al., 1991, as cited in Kliejunas, 2003). These woodlands yield important benefits, such as water and watershed protection, grazing, wildlife food and habitat, recreation, and wood products (Monahan and Koenig, 2006; Thomas, 1997); are known for their scenic beauty; and contribute to tourism and high property values. The loss of keystone Quercus species in these forests would be detrimental to forest health. In addition, the effects on rare and endangered plant species in these regions are unknown. Phytophthora ramorum is expected to cause significant direct environmental effects, such as extensive ecological disruption or large-

scale reduction of biodiversity. This pathogen has already caused environmental damage with the death of thousands of *Quercus* and *Lithocarpus* trees. The loss of one particular oak species, *Q. agrifolia*, has been shown to negatively impact the populations of five California bird species (Monahan and Koenig, 2006). Barrett *et al.* (2006) have indicated that dozens of wildlife species would be negatively affected by the loss of *L. densiflorus*, *Q. kelloggii*, and *Q. agrifolia* and the associated loss of food, nesting, and den sites.

A number of genera on the APHIS Host and Associated Plants Lists have species on the U.S. Fish and Wildlife Service Threatened and Endangered Species List (USFWS, 2007). These are *Arctostaphylos confertiflora*, *A. glandulosa* ssp. *crassifolia*, *A. hookeri* var. *ravenii*, *A. morroensis*, *A. myrtifolia*, *A. pallida*, *Prunus geniculata*, *Q. hinckleyi*, and *R. chapmanii*.

The rating for Environmental Impact is High. The uncertainty lies with the difficulty in producing estimates for the costs of *P. ramorum* that address all of the relevant ecological components. These include: (1) the environmental costs of prevention, eradication, or suppression due to herbicide use; (2) the effects on endangered species; and (3) the indirect ecological consequences (changes in locally important ecological processes such as perturbations of hydrological cycles, *e.g.*, flood control and water supply, waste assimilation, nutrient recycling, conservation and regeneration of soils, and crop pollination and habitat destruction).

C. Likelihood of Introduction

The Likelihood of Introduction for a pest is rated relative to three factors and is based on APHIS PRA guidelines (APHIS, 2002). The first factor, Entry Potential, is based on the volume of materials moved domestically and internationally, the value of these shipments, and the likelihood that the pathogen will survive post-harvest treatments and shipment. The second factor, Establishment and Spread Potential, includes the likelihood that the pathogen will be imported or moved to an area suitable for survival and will encounter host material. The third factor, Detection Potential, is an estimation of the likelihood that the pathogen will not be detected at ports-of-entry or during domestic inspections.

Subelement 1: Entry Potential

The rating for this risk element is based on the volume and value of domestic shipments and imports from Europe and Canada and on the ability of the pathogen to survive post-harvest treatments and shipment. The volume of plants for planting from Europe increased from approximately 33 million plants in 2000 to 47 million plants in 2003, and dropped to 38 million plants in 2004 (Table 5); the drop was possibly a result of restrictions on the imports of regulated hosts of *P. ramorum*.

Live plants are grown, shipped, and sent to areas conducive to their survival. Plant products, such as cut flowers and foliage will also be treated in ways not detrimental to the survival of *P. ramorum*. For example, *P. ramorum* has been detected in nursery stock shipped from California to 21 other states and eradicated in nurseries in which it was detected. In addition, models (Kluza *et al.*, 2007; Magarey *et al.*, 2004, 2007; Smith *et al.*, 2002) have indicated that most of the eastern United States has both potential hosts and favorable conditions (Fig. 3).

Living plants are not likely to receive post-harvest treatments such as irradiation, methyl bromide, or steam sterilization because these treatments would likely kill the plants as well as the pests. In addition, the presence of potting media requires specific testing to ensure the efficacy of any proposed post-harvest treatments (Jarvis, 1992). General transport conditions for potted plants range from 10-18°C and 85-90% relative humidity (McGregor, 1987). *Phytophthora ramorum* has an optimum temperature range of 18-25°C (DEFRA, 2004c; Werres *et al.*, 2001) and survives temperatures as low as -9°C (DEFRA, 2004c).

Although not handled as gently as live plants and cut flowers/foliage, other infested plant products such as logs, lumber, wood chips, and firewood may harbor the pathogen and present a pathway for introduction into new areas. For example, *P. ramorum* has been recovered from inner bark and wood chips (Davidson *et al.*, 2003b), suggesting that when the inner bark is exposed, as in the debarking process, and free water is present, the pathogen can sporulate on the exposed surfaces. Additionally, sporulation was stimulated in baiting trials when inoculated "logs" were kept at 12°C prior to baiting (Garbelotto, 2002) and the pathogen has been recovered from firewood stored for six months (Shelly *et al.*, 2005a). For these reasons, the rating for this Subelement is High.

Subelement 2: Establishment and Spread Potential

Suitable hosts must be available to establish and sustain a pest population, and there must be a mechanism for the pest to reach these hosts. Both natural and human-assisted factors aid in the dispersal of *P. ramorum* (Davidson *et al.*, 2003a; Davidson and Shaw, 2003; Hansen *et al.*, 2002). This pathogen infects 70 genera in 35 plant families (Table 1). Many of these hosts are widely distributed in the United States, and conducive climatic conditions are prevalent along the East and West Coasts (Fig. 3). In woody canker hosts, sporulation is not observed on the surface of cankers (Davidson and Shaw, 2003). However, if the inner bark (cambium) is exposed and free water is present, the pathogen can sporulate on exposed surfaces, *e.g.*, the pathogen has been recovered from inner bark, wood chips (Davidson and Shaw, 2003) and from firewood stored for six months (Shelly *et al.*, 2005a). Sporulation in baiting trials was stimulated when inoculated "logs" were kept at 12°C prior to baiting (Garbelotto, 2002). In several tree species, the xylem has recently been shown to harbor mycelia and chlamydospores of *P. ramorum* (Brown and Brasier, 2007; Parke *et al.*, 2007). For these reasons, the rating for this Subelement is High.

Subelement 3: Detection Potential

Species of *Phytophthora*, such as *P. ramorum*, are difficult to detect at ports-of-entry, where visual inspection is the primary method of detection; *Phytophthora* spp. have only been detected 12 times since 1985 (PPQ, 2007a). In addition, there are recent reports of asymptomatic infection and sporulation (Denman *et al.*, 2008; Vettraino *et al.*, 2007). Other pathogens and environmental conditions can elicit the same symptomology in foliar and dieback hosts. Two newly detected *Phytophthora* species, *P. nemorosa* and *P. kernoviae*, induce similar cankers on trees and were found as a result of field analyses for *P. ramorum*. *P. nemorosa* occupies a similar ecological niche to *P. ramorum* in the United States (Hansen *et al.*, 2004) and *P. kernoviae* a similar niche in the United Kingdom (Brasier *et al.*, 2004a, 2005; DEFRA, 2004a).

Isolation techniques, including direct plating and baiting, are used to detect the pathogen in plant tissues, soil, and water. The efficacy of these techniques varies with season and host (Davidson et al., 2002c). Molecular detection techniques include ELISA (at the genus level), AFLP, and a variety of PCR protocols. Nested and real-time PCR methods are currently used for regulatory purposes in the United States (PPQ, 2007b) and real-time PCR methods are used in the United Kingdom (Lane et al., 2007). The internal transcribed spacer (ITS) DNA analysis does not always distinguish P. ramorum from P. lateralis (Blomquist and Kubisiak, 2003) and P. foliorum (Donahoo et al., 2006); however, multiplex methods can increase sensitivity. The possibility of failure of visual inspection to detect latent infections is a concern.

The disease was first noted in California in 1995 (Garbelotto *et al.*, 2001) and, based on the rate of oak death, researchers suggest the pathogen was introduced at least five years before the first detection (Rizzo and Garbelotto, 2003). Since that time, survey and diagnostic methods have improved, thereby increasing the likelihood of detecting the pathogen. Nevertheless, for the above reasons, the rating for this Subelement is High.

The rating for Likelihood of Introduction is High. Both natural and human-assisted factors aid in the dispersal of *P. ramorum* to areas where suitable hosts and climatic conditions are available and conducive to establishing and sustaining a population. Differences in sporulation ability and susceptibility to infection have been reported for foliar, dieback, and canker hosts. The uncertainty lies with the variability in detecting *P. ramorum* and the ability to predict the levels of resistance and susceptibility among hosts and potential hosts occurring in non-infested regions.

D. Pest Risk Potential

The Consequences of Introduction and the Likelihood of Introduction are rated High; therefore, the Pest Risk Potential is High. The overall risk presented by *P. ramorum* is High due to the number of pathways associated with and the biological uncertainties of the pathogen, *e.g.*, the demonstrated long-distance dispersal in trade, long-term viability of infective propagules, detection of the propagules, lack of definitive host range, the sensitivity of detection of infected plants by visual inspection, and means of natural movement. Research is needed on dormancy in chlamydospores; increased sensitivity and specificity of detection techniques; temperature requirements for survival of propagules in various sources, *e.g.*, soil, wood; risk of moving the pathogen in various species and hybrids; screening for more potential hosts including products and propagative material of vegetable, fruit, and nut crops; and natural dispersal, especially animal and aerial dispersal. The lack of a definite host range and a definitive geographic distribution adds to the pest risk potential.

Table 5. Imports of plant materials from Europe and Canada to the United States (quantity in 1000 units; value in \$1000 U.S.).*

Tuoic 5. Imp	Values in 1000 dollars/Quantities in Thousands, Except Where Indicated													
Origin and Commodity	20	000	2001			2002		003		004	2	2005	200)6
	Quantity	Value	Quantity	Value	Quantity	Value	Quantity	Value	Quantity	Value	Quantity	Value	Quantity	Value
EUROPEAN UN	ION – 25										,			
Unrooted Cuttings/Slips, no soil	19,573.5	\$3,817	19,162.3	\$3,793	20,874.8	\$3,895	27,100.9	\$4,910	23,340.5	\$4,945	48,687	\$64,093,534	36,572	\$70,611,795
Other Plants, with soil	12,941.2	\$3,399	14,486.2	\$2,791	14,934.5	\$3,430	15,463.1	\$4,725	13,531	\$4,155	15,023	\$23,251,032	16,650	\$22,493,099
Trees/Shrubs, with soil	186.6	\$269	545	\$651	370.4	\$553	269.4	\$714	209.7	\$610	441	\$920,064	179	\$828,929
Trees/Shrubs, with soil (metric tons)	1	\$4,559	0	\$0	0	\$0	10	\$13,164	6	\$6,509	0	\$0	1	\$6,793
Roses	286.5	\$463	627	\$744	818.1	\$1,429	415.5	\$778	294.4	\$493	259	\$996,087	302	\$1,082,559
Rhododendrons, Azaleas	90	\$8	96	\$12	31	\$3	0	\$0	0	\$0				
Total (thousands)	\$33,077.8	\$7,956	\$34,916.5	\$7,991	\$37,028.8	\$9,310	\$43,248.9	\$11,127	\$37,375.6	\$10,203	\$64,410	\$89,260,717	\$53,703	\$95,016,382
Total (metric tons)	1	\$4,559	0	\$0	0	\$0	10	\$13,164	6	\$6,509	0	\$0	1	\$6,793
CANADA														
Unrooted Cuttings/Slips, no soil	1,675.00	\$802	2,290.50	\$987	3,860.70	\$1,529	612.2	\$240	536.5	\$219	1,013	\$174,477	429	\$264,768
Other Plants, with soil	258,232.80	\$108,713	263,284.20	\$118,760	292,453.30	\$131,710	289,472.50	\$137,898	272,545.30	\$136,290	241,766	119,406,026	238,679	117,307,181
Trees/Shrubs, with soil	3,162.40	\$3,788	3,033.30	\$3,977	3,142.50	\$4,252	7,581.40	\$3,969	5,635.50	\$3,998	5,439	\$4,410,612	9,622	\$5,009,071
Trees/Shrubs, with soil (metric tons)	22,409	\$25,294,003	43,532	\$29,382,769	29,776	\$25,971,459	33,231	\$27,485,040	32,847	\$30,443,868	29,053	\$27,981,827	29,579	\$26,762,738
Roses	6,609.40	\$11,071	6,166.30	\$12,459	6,566.00	\$12,563	7,429.10	\$14,950	7,496.80	\$15,199	7,006	\$13,549,717	6,516	\$13,766,645
Christmas Trees not Firs	447.7	\$5,083	415.3	\$4,534	377.8	\$4,100	344.5	\$4,105	292.4	\$4,342	247	\$3,485,751	338	\$3,698,761
Christmas Trees, Firs	2,063	\$18,944,036	2,195	\$21,618,957	2,241	\$22,113,441	2,169	\$23,461,596	2,015	\$23,358,746	1,981	\$23,676,722	2,063	\$18,944,036
Rhododendrons, Azaleas	665.3	\$3,352	771.2	\$3,665	2,153.80	\$3,237	510	\$2,580	528.6	\$2,842	533	\$3,078,863	695	\$3,874,205
Total (thousands)	272,855.6	19,076,845	388,461.8	21,763,339	431,291.1	22,270,832	441,911.7	23,625,338	428,902.1	23,521,636	396,292	167,782,168	124,072,916.8	162,864,667
Total (metric tons)	22,409	\$25,294,003	43,532	\$29,382,769	29,776	\$25,971,459	33,231	\$27,485,040	32,847	\$30,443,868	29,053	\$27,981,827	29,579	\$26,762,738
	· ·	. , ,	,	. , ,		\$25,971,459	33,231	\$27,485,040	32,847	\$30,443,868	29,053	\$27,981,827	29,579	\$2

^{*}Data compiled by Lynn Garrett, USDA APHIS CPHST PERAL Economist

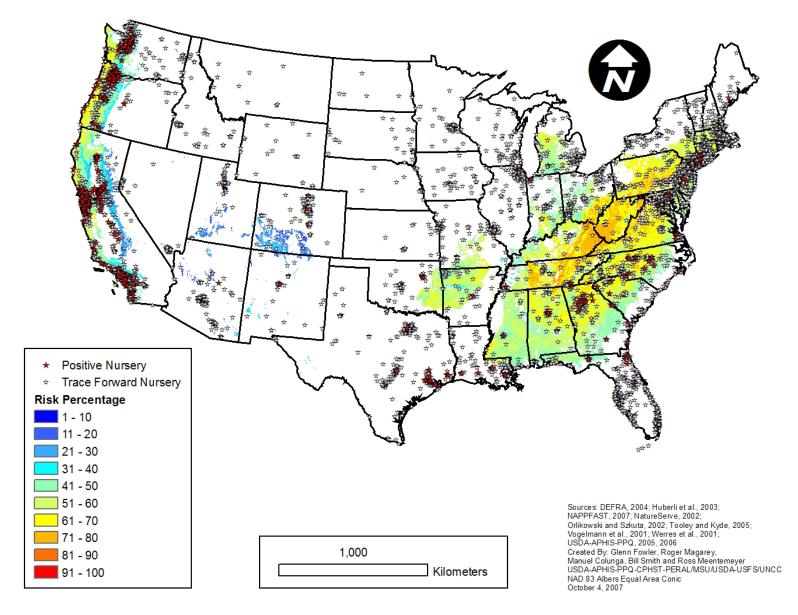


Fig. 3. Locations receiving plants shipped from nurseries testing positive for *Phytophthora ramorum* 2004-2006 overlaid on climate potential.

V. Pathway Assessments

The preceding section contained an overall pest risk assessment for *P. ramorum*. This section takes information from the overall assessment and focuses it on particular pathways. Pathways analyzed are **nursery stock** (**including Christmas trees for planting**), **wood and wood products, cut Christmas trees, cut foliage/flowers, greenwaste, compost, potting media, and soil.** As in the overall assessment, risk levels are categorized as High, Medium, and Low and levels of uncertainty are indicated. The risk ratings for the overall and individual pathway assessments are summarized in a comparative risk matrix (Table 6).

A. Consequences of Introduction

Risk Element 1: Climate-Host Interaction

This risk element considers ecological zonation and the interactions of *P. ramorum* with its hosts in a variety of environments with diverse biotic and abiotic conditions. When introduced into new areas, pests are expected to behave as they do in their native areas if the potential host plants and suitable climate are present. Broad availability of suitable climates and a wide distribution of suitable hosts are assumed to increase the impact of a pest introduction. The ratings for this risk element are based on models, research, and the number of United States Plant Hardiness Zones (USDA, 2003) which contain potential host plants and suitable climate. Because of the large number of hosts and climate range, the analysis of this Element applies to all pathways.

Phytophthora ramorum has a high probability of encountering favorable climatic conditions throughout the ranges of potential hosts that occur in several Plant Hardiness Zones. Modeling of environmental conditions suggests there are many areas in the United States outside the quarantined areas of California and Oregon that have both favorable conditions for disease development and susceptible hosts (Kluza *et al.*, 2007; Magarey *et al.*, 2004, 2007; Smith *et al.*, 2002).

The rating for the Climate-Host Interaction element is High for all pathways assessed. The uncertainty lies in the range of biotic and abiotic factors triggering establishment of *P. ramorum* in new areas.

Risk Element 2: Host Range

The risk posed by a plant pest depends on both its ability to establish a viable reproductive population and its potential to damage plants. This risk element assumes that the consequences of pest introduction are positively correlated with the pest's host range. Aggressiveness, virulence, and pathogenicity may also be factors. The consequences related to host range are rated in accordance with the ability of the pathogen to attack a single species or multiple species within a single genus, a single plant family, or multiple families.

The host range of this pathogen continues to expand though detections in the field. APHIS currently regulates 40 plant species and all species in five genera as proven hosts, with an additional 64 species listed as associated plants (Table 1) (APHIS, 2007a). The potential host range is also increasing, as determined through a variety of screening techniques including detached leaf, whole plant, and log assays (DEFRA, 2004c; Hansen *et al.*, 2005; RAPRA, 2007; Tooley and Kyde, 2007). Experimental evidence indicates that several eastern forest species would be more susceptible than western forest species, such as in affected quarantined areas of

California and Oregon. In addition, differences in host susceptibility are documented for forest and nursery species and may affect disease development in new environments (DEFRA, 2004c; Linderman *et al.*, 2007; Meshriy *et al.*, 2005; Tooley *et al.*, 2004; Tooley and Kyde, 2007).

Nursery Stock (Including Christmas Trees for Planting)

Nursery plants are intended for planting in the landscape. The locations of these plantings include commercial plantings, private residences, arboreta, large parks, and interiorscapes. Christmas trees are often planted in home landscapes. *Pseudotsuga menziesii* var. *menziesii* and *A. grandis* are confirmed hosts used as Christmas trees. Several nursery plants, specifically *Camellia* spp., *Pieris* spp., *Rhododendron* spp., and *Viburnum* spp., have already been implicated in the movement of *P. ramorum*. These four genera have been associated with repeated regulatory actions in North America and Europe and appear to present a greater risk for the pathogen's movement (APHIS, 2005b, 2007b; COMTF, 2005; EC, 2007).

Wood and Wood Products

Wood and wood products can be an important pathway for the movement of *P. ramorum*. Brown and Brasier (2007) isolated *P. ramorum* from the xylem up to 25 mm below the phloem and up to 27 weeks after removal of the phloem of *A. pseudoplatanus*, *F. sylvatica*, *Q. cerris*, *Q. acuta*, and *Q. petraea*. Parke *et al.* (2007) found *P. ramorum* in xylem tissue and chlamydospores in xylem vessels. *Phytophthora ramorum* has been recovered from firewood after six months of storage (Shelly *et al.*, 2005b). Sporulation has not been observed on the outside, intact bark of infected *Quercus* spp. or *L. densiflorus* logs (Davidson *et al.*, 2005). However, the pathogen has been recovered from or observed to sporulate on various wood products, *e.g.*, flooded chips of infected *L. densiflorus* and the flooded, cut edges of *Q. agrifolia* cankers (Davidson and Shaw, 2003). Results from log inoculation tests of *P. menziesii* var. *menziesii* have been inconsistent (Hansen *et al.*, 2004). The main trunks of *P. menziesii* var. *menziesii* and *S. sempervirens*, important timber species, have not been found to be infected by *P. ramorum* (Davidson *et al.*, 2003c)

Cut Christmas Trees

Pseudotsuga menziesii var. menziesii and A. grandis are grown in plantations and farmed for Christmas trees (COMTF, 2005). Pseudostuga menziesii var. glauca, while not a host, has demonstrated susceptibility to P. ramorum in controlled studies; it is native to the intermountain zones (Rocky Mountains) and occurs at higher elevations and has greater cold hardiness than P. menziesii var. menziesii. In mixed forests, P. ramorum has been found infecting understory P. menziesii var menziesii and small branches, needles of sprouts, and twig tips of S. sempervirens. Studies are underway to examine sporulation on these two hosts (Davidson et al., 2003c). Twenty of the conifer species tested, including many important species used as Christmas trees, were susceptible to P. ramorum (Chastagner et al., 2004). Some Abies spp. were highly susceptible. Symptoms included needle blight, a shoot blight resulting from needle infections, and stem lesions resulting from the growth of the pathogen from infected needles into the stem. Growth stage has an apparent significant effect on susceptibility (Chastagner et al., 2004).

Cut Flowers/Foliage

Leaves and branches of hosts such as *U. californica*, *P. menziesii* var. *menziesii*, and *S. sempervirens* are used in wreaths and garlands (Davidson and Shaw, 2003). *Rhododendron* and *U. californica* leaves can be dried for several weeks, and the pathogen is still viable after rehydration (Garbelotto, 2003a). Numerous hosts of *P. ramorum* are popular for cut flower

production, including *Acer*, *Camellia*, *Hamamelis*, *Kalmia*, *Pieris*, *Rhododendron*, *Rosa*, and *Syringa* (Bachmann, 2002). There are multiple areas of uncertainty, including a lack of data on infestation and transmission rates of *P. ramorum* in other host species used for cut foliage and flowers. For example, movement of *P. ramorum* in *Viburnum* and *Rhododendron* nursery stock is documented (APHIS, 2005b, c; COMTF, 2005), but not in cut flowers. The intended uses and disposal of plants for planting and internal ornamental use differ. Cut flowers and foliage are less likely to come into contact with live hosts, since most of this material is used for decorative purposes indoors and then discarded.

Greenwaste and Compost

An estimated 10 million tons of greenwaste infected by *P. ramorum* accumulate in coastal California each year (Garbelotto, 2003a). Greenwaste containing host material from infested areas may serve as a source of inoculum, especially from leaves of foliar hosts. *Rhododendron* and *U. californica* leaves can be dried for several weeks and the pathogen is still infectious after rehydration (Garbelotto, 2003a). Although it has not been demonstrated, it is postulated that spores could be dispersed from foliar hosts via rain-splash if open transit containers are used, or that infected leaves could detach and blow away (Davidson and Shaw, 2003).

When infected wood chips, firewood, and branches are kept in a cool and moist environment, they can harbor viable *P. ramorum* for long periods (Shelly *et al.*, 2005a, b). These substrates are commonly brought into commercial composting facilities (Garbelotto, 2003a).

Municipal composting processes reduce the viability of many plant pathogens, including *P. ramorum*, due to high temperatures and enzymatic activity. Composting has been demonstrated to reduce *P. ramorum* populations below detectable levels; however, preliminary data suggest re-infestation of finished compost by the pathogen is possible (Swain and Garbelotto, 2006).

Potting Media

Potting media are composed of organic and inorganic matter and are intended for various uses both indoors and outside. Experimental evidence indicates that *P. ramorum* may survive and infect plants via potting media. Parke *et al.* (2004a) and Parke and Lewis (2007) found that *P. ramorum* moved through a sterile potting medium and infected *Rhododendron* plants. *P. ramorum* survived in *Camellia* leaves up to 100 days in a potting medium, even after the leaves were decaying (Shishkoff and Tooley, 2004), and up to 11 months on *Camellia* and *Rhododendron* roots buried in potting media (Shishkoff, 2007). In laboratory tests, the form of inoculum influenced the survival of *P. ramorum* in potting media, *e.g.*, six months when introduced as sporangia and 12 months when introduced as chlamydospores (Linderman and Davis, 2006c). *Phytophthora ramorum* has also been recovered from potting media at an infested nursery (OSOS, 2004).

Soil

Phytophthora ramorum has been isolated seasonally from soil in hiking trails and from soil on hikers' boots (Cushman and Meentemeyer, 2006; Davidson *et al.*, 2002c; Tjosvold *et al.*, 2002b; Webber and Rose, 2008). In this same study, a survey of those visitors with infested shoes showed that many people leaving the park were going to other parts of California, the United States, or Europe (Tjosvold *et al.*, 2002b). Recovery rates of *P. ramorum* in areas with host plants was equal from soil samples collected on hiking trails and off the trails (Cushman and

Meentemeyer, 2006). The pathogen was only recovered from samples collected from the trails in two areas without hosts, suggesting human-assisted movement of the pathogen along the trails. In the laboratory survival tests, Linderman and Davis (2006c) inoculated several substrates (coir dust, composted dairy manure, fir bark, peatmoss, potting mix, alluvial sand, sawdust, and garden clay loam soil) with *P. ramorum* sporangia. The pathogen was recovered up to six months in coir dust, composted dairy manure, fir bark, potting mix and sawdust; five months in peat moss; four months in garden clay loam soil; and two months in alluvial sand.

The rating for the Host Range Risk Element is High for the Nursery Stock, Wood and Wood Products, Greenwaste, Compost, Potting Media, and Soil pathways. The Cut Christmas Trees and Cut Flowers/Foliage pathways are rated Medium because of end use and disposal. The uncertainty in the ratings for this Element lies in the unknowns, *e.g.*, the extent of the host range, infestation and transmission rates, and disposal methods.

Risk Element 3: Dispersal Potential

Pests may disperse after introduction into new areas. The dispersal potential indicates how rapidly and widely the pests may spread within the importing country or region and is related to the pest's reproductive potential, inherent mobility, and external dispersal facilitation modes. Factors for rating the dispersal potential include the presence of multiple generations per year or growing season, the relative number of offspring or propagules per generation, any inherent capabilities for rapid movement, the presence of natural barriers or enemies, and dissemination enhanced by wind, water, vectors, or human assistance.

The scattered pattern of sites where *P. ramorum* has become established suggests it has both natural (Hansen *et al.*, 2002; Tjosvold *et al.*, 2002c) and human-assisted movement (Werres *et al.*, 2001). Long-distance spread may occur when strong winds, common during heavy rains along the California coast, move the easily detached sporangia great distances (Hansen *et al.*, 2002). Initial survey results in California and Oregon indicate *P. ramorum* is in streams and rivers adjacent to and far from known infested areas (Hansen *et al.*, 2005; Murphy *et al.*, 2006). Anthropogenic movement includes soil on hikers' boots (Davidson *et al.*, 2002c, 2005; Tjosvold *et al.*, 2002b) and nursery stock (APHIS, 2005b, c).

Nursery Stock (Including Living Christmas Trees)

Phytophthora ramorum is a polycyclic pathogen on many nursery hosts; evidence indicates that inoculum production follows periods of rain and that certain foliar hosts, including *Rhododendron* and *Syringa*, are prolific producers of sporangia or chlamydospores or both (Davidson *et al.*, 2003c). Pathogen transmission has been documented from one nursery to another on nursery stock. Confirmed positive sites from the trace-forward, national, and other survey total 176 in 21 states (APHIS, 2005a, b). While most of these were nursery finds, the total includes three residential finds. As of January 10, 2005, all nursery stock shipped from California, Oregon, and Washington is regulated to prevent movement of this pathogen (APHIS, 2005a). In Europe, *P. ramorum* has been transported into a number of countries via infected nursery stock (Davidson and Shaw, 2003; Lilja *et al.* 2007; RAPRA, 2007).

Wood and Wood Products

Wood and wood products can be an important pathway for the movement of *P. ramorum*. The pathogen has been recovered from or observed to sporulate on various wood products. Sporulation has occurred on flooded chips of infected *L. densiflorus* and the flooded, cut edges of *Q. agrifolia* cankers (Davidson and Shaw, 2003; Davidson *et al.*, 2005). The pathogen has been recovered 3 cm into wood of *Quercus* spp. (D. Rizzo, unpublished data in Davidson and Shaw, 2003), up to 25 mm from *A. pseudoplatanus*, *F. sylvatica*, *Q. acuta*, *Q. cerris*, and *Q. petraea* (Brown and Brasier, 2007); up to 4 cm in *L. densiflorus* (Parke *et al*, 2007); and from firewood after six months of storage (Shelly *et al.*, 2005a), indicating that wood products (mulch, firewood, chips, *etc.*) may be infective. Sporulation has not been observed on the outside of intact bark of infected *Quercus* spp. or *L. densiflorus* logs (Davidson *et al.*, 2005). However, the main trunks of *P. menziesii* var. *menziesii* and *S. sempervirens*, important timber species, have not been found to be infected by *P. ramorum* (Davidson *et al.*, 2003c) and so logs, lumber, and other wood products of these species are not regulated.

There is uncertainty with this pathway. Data on infestation and transmission rates of *P. ramorum* in wood products indicate that the recovery of the pathogen is low. When coupled with the uncertainties about *P. ramorum* survival, especially chlamydospores, these rates may be deceptively low.

Cut Christmas Trees

Pseudotsuga menziesii var. menziesii, a host of P. ramorum, is native to the Sierra Nevada and coastal mountains of California, Oregon, Washington, and British Columbia; it is grown in plantations and farmed for Christmas trees. P. ramorum infects the small branches of P. menziesii var. menziesii and the small branches and needles of S. sempervirens. Studies are underway to examine sporulation on these two hosts (Davidson et al., 2003c). In 2004, 665 Christmas tree plantations in Oregon and 100 in Washington were surveyed and found negative for P. ramorum (COMTF, 2004b). Twenty of the conifer species tested, including many important species used as Christmas trees, were susceptible to P. ramorum (Chastagner et al., 2004, 2006b). Some Abies spp. were highly susceptible in laboratory tests. Symptoms included needle blight, a shoot blight resulting from needle infections, and stem lesions where the pathogen infecting needles grew into the stem. Growth stage had a significant effect on susceptibility.

Cut Flowers/Foliage

Leaves and branches of hosts such as *U. californica, P. menziesii* var. *menziesii*, and *S. sempervirens* are used in wreaths and garlands. Some of these plants are grown within the regulated counties in California and have been sold throughout the United States. Even without sporulation, fir wreaths and Christmas trees could serve as an infection pathway if hyphae were able to grow from infected branch tips and needles (Davidson and Shaw, 2003).

Additional host species are used for cut flowers and foliage, including *Rhododendron, Kalmia, Camellia*, and *Syringa* on which *P. ramorum* can effectively produce spores (Beales *et al.*, 2004b; Davidson *et al.*, 2003c; DEFRA, 2004c; Parke *et al.*, 2002a). Although there are data for the movement of *P. ramorum* in these hosts as nursery stock, there are no data for cut flowers. These products may be capable of disseminating the pathogen, but their intended use and disposal are principally indoors, reducing the likelihood that they will contact hosts in a new environment.

Greenwaste/Compost

Greenwaste containing host material from infested areas may be a source of inoculum, especially from leaves of foliar hosts (Davidson and Shaw, 2003). Even green material dried for several weeks can be problematic because some plant tissue, such as *Rhododendron* leaves, will sporulate upon wetting (Garbelotto, 2003a). Although it has not been demonstrated, it is postulated that spores could be dispersed from foliar hosts via rain-splash during transit in open containers, or that infected leaves could detach and blow away (Davidson and Shaw, 2003).

Composting can reduce available inoculum from *P. ramorum* infected materials (Aveskamp and Wingelaar, 2006; Garbelotto, 2003a; Swain *et al.*, 2005, 2006), but is not equally effective on all materials (Swain *et al.*, 2005). Municipal composting processes reduce the viability of many plant pathogens, including *P. ramorum*, due to high temperatures and enzymatic activity. Composting reduced *P. ramorum* populations below detectable levels; however, preliminary data suggested reinfestation of finished compost is possible (Garbelotto, 2003a; Swain and Garbelotto, 2006; Swain *et al.*, 2006).

Potting Media

The Oregon Department of Agriculture has detected *P. ramorum* in nursery stock, potting media containing compost, and plants in a landscape in Columbia County. This prompted the Oregon Secretary of State to implement an Emergency Quarantine Order in July 1, 2004 to prevent the movement of potting media and compost (OSOS, 2004).

Parke *et al.* (2006b, 2007) demonstrated transmission of *P. ramorum* from infested potting medium to *Rhododendron* plants under greenhouse and laboratory conditions. Linderman and Davis (2005, 2006a, c) compared *P. ramorum* with other *Phytophthora* species in a variety of soil-less potting media (river sand, Douglas-fir bark, coir, sphagnum peat, redwood sawdust, a bark-peat-pumice potting mix), dairy compost, and garden soil. The pathogen was detected for six months from all substrates amended with sporangia or chlamydospores in vermiculite but not with infected leaf inoculum. *P. ramorum* sporangia survived best in peat moss, the potting mix, coir, and Douglas fir bark, and poorest in sand and soil. These results indicate that *P. ramorum* survives very well in potting mix components and soil as culture-produced sporangia or chlamydospores.

Soil

Inoculum has been isolated seasonally from soil in hiking trails and from soil on hikers' boots (Davidson *et al.*, 2002c; Tjosvold *et al.*, 2002b). In this same study, a survey of those visitors with infested shoes showed that many people leaving the park were going to other parts of California, the United States, or Europe (Tjosvold *et al.*, 2002b). The concern for soil and litter movement by equipment has prompted California authorities to request that vehicles and other equipment, including tents and shoes, be washed prior to leaving an established area. *Phytophthora ramorum* has been recovered, albeit at low levels, from a variety of unprocessed and processed wood products (Shelly *et al.*, 2005b). Soil on felled trees or logging equipment from infested forests may also contain spores (Davidson and Shaw, 2003). Recovery rates of *P. ramorum* in areas with host plants was equal from soil samples collected on hiking trails and off the trails. The pathogen was only recovered from samples collected from the trails in two areas without hosts, suggesting human-assisted movement of the pathogen along the trails (Cushman and Meentemeyer, 2006). Fichtner *et al.* (2007a) reported that it is difficult to detect chlamydospores using current baiting methods, and indicated an underestimation of the amount

of inoculum present in the soil. Rhododendron leaf baits were demonstrated to be superior to pear baits for detection of sporangia; neither bait detected chlamydospores. In addition, soil-incubated inoculum exhibited greater than 60% survival at the end of summer and also supported elevated chlamydospore production, which may provide a reservoir of inoculum for the fall disease cycle (Fichtner *et al.*, 2007a).

The rating for the Dispersal Potential Element is High for the Nursery Stock, Wood and Wood Products, Greenwaste/Compost, Potting Media, and Soil pathways. The risk for Cut Foliage/Flowers and Cut Christmas Trees is Medium because of intended use, *i.e.*, indoors. However, the uncertainty is the final disposition, which could be indoors in trash or outside in compost or greenwaste.

Risk Element 4: Economic Impact

Introduced pests cause a variety of direct and indirect economic impacts, such as reduced yield, reduced commodity value, loss of foreign or domestic markets, and non-crop impacts. Factors considered during the ranking process included the following: effect on yield or commodity quality; plant mortality; ability to act as a disease vector; increased costs of production, including pest control costs, lower market prices, effects on market availability, increased research or extension costs, or reduction in recreational land use or aesthetic value; ability of the pest to attack the hosts or products with significant commercial value, to directly cause tree mortality, or to predispose the host to mortality by other organisms; impact of the pest on the value of the affected host, *e.g.*, by lowering its market price, increasing cost of production, maintenance, or mitigation, or reducing value of property where it is located; and lack of effective control measures.

The economic impact of each pathway is addressed below. Losses in real estate value and costs of removal and disease management are estimated to be about \$100 million/year (Stipes and Campana, 1981). In addition, plant pathogens of forest plants cause the loss of approximately 9%, or \$7 billion, of forest products each year (Hall and Moody, 1994; USBC, 1998). The proportion of introduced plant pathogens in forests is similar to that of introduced insects (about 30%); thus, approximately \$2.1 billion in forest products are lost each year to non-indigenous plant pathogens in the United States. In addition, tourism can be affected, as visitors to parks and forests may find that access to selected areas is restricted during certain seasons to prevent the pathogen's movement, or to protect visitors from falling limbs from trees killed by *P. ramorum* (COMTF, 2004a). When visitors are requested or required to take precautions to prevent movement, park and forest staff maybe required to provide educational information, staff cleaning areas, and provide appropriate supplies and equipment to remove soil from shoes and vehicles.

Nursery Stock (Including Living Christmas Trees and All Propagative Material)

Nursery crops are woody perennial plants such as ornamental trees, shrubs, and vines that are primarily used for landscaping. In 2006, the U.S. domestic production of nursery crops was valued about \$12.9 billion. Imports for these crops were \$341 million and exports were \$287 million (Jerado, 2007). Lost in nursery stock from regulatory actions in the state of Washington in 2004 and 2005 was over \$400,000, and this value does not include labor or other costs associated with the destruction of the plants and eradication efforts at the nurseries involved (Dart and Chastagner, 2007).

The presence of *P. ramorum* has resulted in restrictions in the foreign and domestic trade of nursery stock. Australia, Canada, Korea, New Zealand, the European Union, and Switzerland have placed restrictions on the movement of affected plants and plant parts from the United States (EXCERPT, 2007; Rizzo and Garbelotto, 2003). In addition, the United States has placed restrictions on movement of propagative material from Europe (Aley, 2007).

Wood and Wood Products

Thousands of *Quercus* and *L. densiflorus* trees have died following infection by this pathogen, requiring expensive removal in certain settings, more intensive fire management in others, and limited access to parts of parks and forests (COMTF, 2004c). Economic losses from removal of infected *Quercus* trees may be partially offset by utilization of the material for wood products (Shelly *et al.*, 1996). The presence of *P. ramorum* has resulted in restrictions in foreign and domestic trade. Canada, Korea, Australia, New Zealand, and the European Union have placed restrictions on the movement of affected plant and plant parts from the United States (EXCERPT, 2007; Rizzo and Garbelotto, 2003). Should *P. ramorum* become established in other U.S. hardwood forests, the potential economic threat to commercial timber production exceeds \$30 billion (Kliejunas, 2003).

Cut Christmas Trees

The U.S. cut Christmas tree industry had a wholesale value of \$520 million in 2003 (Jerado, 2004). Oregon leads with a total production of \$158 million (Jerado, 2004) and markets throughout the United States, Canada, and Mexico (OASS, 2004). Washington and California follow with values of \$60 and \$10.4 million, respectively (Jerado, 2004). A major Christmas tree species, *P. menziesii* var. *menziesii*, is a host of *P. ramorum*. Chastagner *et al.* (2004) found other important species susceptible in laboratory trials, and other species have been found naturally infected, *e.g.*, *Abies grandis* and *A. magnifica* (Table 1.)

Cut Flowers/Foliage

U.S. production exceeded \$406 and \$542 million respectively for cut flower and foliage sales (Jerado, 2007). Many of the species surveyed and listed by National Agricultural Statistics Service (NASS, 2007) were not hosts of *P. ramorum*, but there is an increase in flower production of woody ornamentals and many of these plants are hosts for the pathogen, including *Acer, Camellia, Hamamelis, Kalmia, Pieris, Rhododendron, Rosa*, and *Syringa* (Bachmann, 2002).

Greenwaste/Compost

A major economic issue for quarantined counties in California is appropriate disposal of *P. ramorum*-infested or contaminated greenwaste. It is estimated that about 10 million tons of infected greenwaste are accumulating in quarantined counties in California per year (Garbelotto, 2003a). This is complicated by the fact that only 50% of this material can go into landfills (COMTF, 2005).

Twenty-nine of 143 nurseries questioned by State officials or industry representatives in the quarantined counties of California indicated they would suffer a financial loss if they could no longer use native soil or local compost (Jordan, 2003).

Potting Media and Soil

The pathogen was detected in potting media at an infested nursery in Oregon (OSOS, 2004). Subsequently, Oregon requires potting media used at certified nurseries to be tested. There is experimental evidence that river sand, Douglas-fir bark, coir, sphagnum peat, redwood sawdust, and a bark-peat-pumice potting mix are capable of harboring *P. ramorum* (Linderman and Davis, 2006a, c).

Twenty-nine of 143 nurseries questioned by State officials or industry representatives in the quarantined counties of California indicated they would be affected financially if they could no longer use native soil or local compost (Jordan, 2003).

The Economic Impact rating for all pathways is rated High. Uncertainty stems from unknowns regarding the extent of the host range, the restricted movement imposed by the quarantines, the length of pathogen survival in various pathways, and the value of these products on the open market.

Risk Element 5: Environmental Impact

The ratings for this risk element are based on three aspects: the potential of the pest to disrupt native ecosystems and habitats within its current geographic range, the need for additional chemical or biological control programs, and the potential of the pest to directly or indirectly impact species listed as Threatened or Endangered (50 CFR §17.11-12). When a pest is known to infest or infect other species within the same genus, and host specificity data do not exist with the listed plant, then the listed plant is assumed to be a potential host.

In forests, more than 20 non-indigenous species of plant pathogens attack woody plants (Liebhold *et al.*, 1995). Two of the most destructive plant pathogens are *Cryphonectria parasitica* and *Ophiostoma ulmi*, the causal agents of chestnut blight and Dutch elm disease, respectively. Before the introduction of chestnut blight, approximately 25% of eastern U.S. deciduous forest consisted of American chestnut trees (*C. dentata*) (Liebhold *et al.*, 1995). These trees have all but disappeared. In urban and forest environments, species and cultivars of *Ulmus* have been destroyed by *O. ulmi*. Environmental costs associated with prevention, eradication, or suppression of this pathogen include indirect ecological consequences (perturbations of hydrological cycles, *e.g.*, flood control and water supply, waste assimilation, nutrient recycling, conservation and regeneration of soils, crop pollination) and must take into account both current-use value and future-use values.

Quercus species are the most important and widespread of the hardwood trees in the North Temperate Zone (Pavlik et al., 1991, as cited in Kliejunas, 2003). These woodlands yield important benefits, e.g., water and watershed protection, grazing, wildlife food and habitat, recreation, and wood products (Monahan and Koenig, 2006; Thomas, 1997); are known for their scenic beauty; and contribute to tourism and high property values. The loss of keystone Quercus species in these forests would be detrimental to forest health. In addition, the effects on rare and endangered plant species in these regions are unknown. Phytophthora ramorum is expected to cause significant direct environmental effects such as extensive ecological disruption or large-scale reduction of biodiversity. This pathogen has already caused environmental damage with the death of thousands of Quercus and Lithocarpus trees. The loss of one particular oak species, Q. agrifolia, has been shown to negatively affect the populations of five California bird species

(Monahan and Koenig, 2006). Barrett *et al.* (2006) indicated that dozens of wildlife species would be negatively affected by the loss of *L. densiflorus*, *Q. kelloggii*, and *Q. agrifolia* and the associated loss of food, nesting, and den sites.

A number of genera on the APHIS List of Proven Hosts and Associated Plants have species on the U.S. Fish and Wildlife Service Threatened and Endangered Species List (USFWS, 2007). These are *Arctostaphylos confertiflora*, *A. glandulosa* ssp. *crassifolia*, *A. hookeri* var. *ravenii*, *A. morroensis*, *A. myrtifolia*, *A. pallida*, *Prunus geniculata*, *Q. hinckleyi*, and *R. chapmanii*.

Although the rate of introduction may vary with each pathway, the impact on the environment is the same. *Phytophthora ramorum* can move in nursery stock (APHIS, 2005b; Bulluck *et al.*, 2006), wood and wood products (Shelly *et al.*, 2005b), cut Christmas trees (Chastagner *et al.*, 2004), cut flowers/foliage (Davidson *et al.*, 2003c; DEFRA, 2004c; Parke *et al.*, 2002a), greenwaste/compost (Garbelotto, 2003a; Swain *et al.*, 2005, 2006), potting media (Linderman and Davis, 2006a, c; Parke *et al.*, 2006b), and soil (Davidson *et al.*, 2002c; Davidson and Shaw, 2003; Tjosvold *et al.*, 2002b). All of the pathways present a potential risk of contaminating the environment with *P. ramorum*.

The Environmental Impact rating for all pathways is High. The uncertainty lies with the difficulty in producing estimates for the costs of *P. ramorum* that address all of the relevant ecological components. These include: (1) the environmental costs of prevention, eradication, or suppression due to herbicide use; (2) the effects on endangered species; and (3) the indirect ecological consequences (changes in locally important ecological processes such as perturbations of hydrological cycles, *e.g.*, flood control and water supply, waste assimilation, nutrient recycling, conservation and regeneration of soils, and crop pollination and habitat destruction).

B. Likelihood of Introduction to New Areas in the United States

Risk Element 6: Pest Opportunity (Survival and Access to Suitable Habitat and Hosts)

Subelement 1: Entry Potential

The rating for this risk element is based on the volume of domestic shipments and imports from Europe and Canada. This rating is also based on the likelihood that the pathogen will survive post-harvest treatments and shipping conditions. Live plants are grown, shipped, and sent to areas conducive to their survival. Handling of plant products may not be detrimental to the survival of *P. ramorum*. Although not handled as gently as live plants and cut flowers/foliage, other infested plant products such as logs, lumber, wood chips, and firewood still harbor the pathogen and present a pathway for introduction into new areas. Living plants are not likely to receive post-harvest treatments such as irradiation, methyl bromide, or steam sterilization because there is no "harvest" of the commodity, and the types of treatments that would kill the pests are also likely to kill the plants. In addition, the presence of potting media or soil requires specific testing to ensure the efficacy of any proposed post-harvest treatments (Jarvis, 1992). General transport conditions for potted plants range from 10-18°C and 85-90% relative humidity (McGregor, 1987).

Nursery Stock (Including Living Christmas Trees)

Phytophthora ramorum is likely to survive in the plant host during transportation. This was demonstrated recently when infected nursery stock in 21 states was traced to infested nurseries in California. In Europe, P. ramorum was introduced to Majorca, Spain via a shipment of infected rhododendrons, and many of the infections found in nurseries in Europe could be traced to plant transport from other nurseries (Davidson and Shaw, 2003; Lilja et al., 2007; RAPRA, 2007; Rytkönen et al., 2007). Chlamydospores are often formed inside host tissue (Parke and Lewis, 2007; Pogoda and Werres, 2004), and are unlikely to be dislodged during standard harvesting, handling and shipping operations. *Phytophthora ramorum* has survived up to six months in greenhouse conditions (Linderman and Davis, 2006a, c), overwintered in the United Kingdom (DEFRA, 2004c), and over-summered in the United States (Fichtner et al., 2006a, b, 2007a). The biology of chlamydospores of *P. ramorum* and their epidemiological role is still under investigation, but chlamydospores of other *Phytophthora* species can survive for up to five years (Erwin and Ribeiro, 1996). Detached *Rhododendron* and *U. californica* leaves still produced sporangia several weeks after drying (Garbelotto, 2003a). In addition to movement with the aerial portions of the host, there is laboratory evidence that the pathogen may move in potting media and evidence of root infection in nursery stock (Linderman and Davis, 2006a, 2006c; Parke et al., 2004a; Parke and Lewis, 2007; Shishkoff, 2007).

Wood and Wood Products, Cut Christmas Trees, Cut Flowers/Foliage

This pathogen has been detected and isolated from bark, cambium, and xylem and is usually limited to a depth of 2.5 cm in *Quercus* (Brown and Brasier, 2007) and as deep as 4 cm in *L. densiflorus* (Parke *et al.*, 2007). Chlamydospores are often formed inside host tissue (Parke *et al.*, 2003; Pogoda and Werres, 2004) and are unlikely to be dislodged during standard harvesting, handling, and shipping operations. *Phytophthora ramorum* has survived up to six months in greenhouse conditions (Linderman and Davis, 2006a, c), overwintered in the United Kingdom (DEFRA, 2004c), and over-summered in the United States (Fichtner *et al.*, 2006b, 2007a). Much of the biology of its chlamydospores is still under investigation, but chlamydospores of other *Phytophthora* species can survive for up to five years (Erwin and Ribeiro, 1996).

Greenwaste/Compost, Soil, Potting Media

Phytophthora ramorum has been detected from greenwaste (Shelly et al., 2005a), compost (Garbelotto, 2003a), soil (Tjosvold et al., 2002b), and potting media (Linderman and Davis, 2006a, c; Parke et al., 2006b; Shishkoff, 2007). Spores of P. ramorum have been detected on hikers' shoes and on the tires of mountain bikes and vehicles used on dirt roads or trails in California (Tjosvold et al., 2002b). Linderman and Davis (2006a, c) compared P. ramorum with other Phytophthora species in a variety of media (river sand, Douglas-fir bark, coir, sphagnum peat, redwood sawdust, a bark-peat-pumice potting mix, a dairy compost, and a garden soil) and found that the pathogen was detected for six months from all substrates.

The risk rating for the Entry Potential is High for all pathways except compost, which is rated Medium. Uncertainty factors include lack of data on infection and pathogen survival rates for most products, especially cut flowers and foliage; long-term survival in greenwaste, compost, potting media, and soil; and propagules present in wood and wood products.

Subelement 2: Establishment and Spread Potential

Phytophthora ramorum is a polycyclic pathogen; evidence indicates that inoculum production follows periods of abundant rainfall, and *P. ramorum* produces large numbers of sporangia, chlamydospores, or both on certain foliar hosts (Davidson *et al.*, 2003a, 2005). Phytophthora ramorum has an optimum temperature range of 18°-25°C (DEFRA, 2004c; Werres *et al.*, 2001) and survives temperatures as low as -9°C (DEFRA, 2004c). Suitable hosts must be available to establish and sustain a pest population, and there must be a mechanism for the pathogen to reach these hosts. Both natural and human-assisted factors aid in the dispersal of *P. ramorum* (Davidson *et al.*, 2003a, b; Hansen *et al.*, 2002). The host range of this pathogen has expanded and currently attacks 70 genera in 35 plant families (Table 1) (Brasier *et al.*, 2002; Hansen *et al.*, 2005; Tooley and Kyde, 2003, 2007; Tooley *et al.*, 2004). Many of these hosts are widely distributed in the United States, and conducive climatic conditions are prevalent along the East and West Coasts. Modeling of environmental conditions suggests there are several areas in the United States outside of quarantined zones that have both favorable conditions for disease development and susceptible hosts (Kluza *et al.*, 2007; Magarey *et al.*, 2004, 2007).

The pathogen is established in forests in fourteen counties in California and one county in Oregon (APHIS, 2007b) and has been detected from limited established plantings of ornamental shrubs and trees in Europe (DEFRA, 2006; EPPO, 2004; RAPRA, 2007). Newly established populations in forests or other natural environs may go undetected for many years owing to their cryptic nature, concealed activity, slow development of damage symptoms, or misdiagnosis (Rizzo *et al.*, 2002b). However, survey and diagnostic methods have improved, thereby increasing the likelihood of detecting the pathogen. Eradication is currently not feasible for certain forest situations, but is being attempted in Curry County, Oregon (Goheen *et al.*, 2002b; Hansen and Sutton, 2006; Kanaskie *et al.*, 2008) and in garden settings in the United Kingdom. Six years after initial eradication efforts, *P. ramorum* is still being found in native soil and streams in Curry County, Oregon (Kanaskie *et al.*, 2008; Prospero *et al.*, 2007). Although eradication was not considered feasible, suppression efforts are underway in Humboldt County, California (COMTF, 2005).

Nursery Stock (Including Living Christmas Trees)

Many of the hosts on the regulated and associated host lists are major nursery and/or forest/understory species. There is contiguous distribution of hosts, potential hosts, and favorable conditions along the East and West Coasts of the United States (Magarey *et al.*, 2004, 2008). *Phytophthora ramorum* has been detected and eradicated in nursery stock shipped from California to 21 other States. This pathogen has also been detected in nursery stock in many European countries, and from a few established plantings on *Rhododendron* and various tree hosts (EPPO, 2004). In addition, in infested nurseries soil or mulch in the pots of rhododendron plants, other host plants, and even non-host plants that appear healthy may contain spores of *P. ramorum* (Davidson and Shaw, 2003). *Phytophthora ramorum* has also been isolated from irrigation water from an infested rhododendron nursery (Tjosvold *et al.*, 2002c). In addition, *P. ramorum* has been detected downstream from nurseries with infested nursery stock (APHIS, 2007d).

Wood and Wood Products

Phytophthora ramorum has been recovered from chips (Davidson et al., 2003b) and the inner bark and xylem (Brown and Brasier, 2007) of hardwood species, suggesting that when the inner bark is exposed, as in the debarking process, and free water is present, the pathogen can

sporulate on the exposed surfaces. Sporulation was stimulated in baiting trials when inoculated "logs" were kept at 12°C prior to baiting (Garbelotto, 2002) and has been recovered from firewood stored for six months (Shelly *et al.*, 2005a). Tubajika *et al.* (2008) demonstrated that heat treating naturally and artificially inoculated wood rounds and boards to 56°C for 30 minutes may not have been adequate to kill the pathogen. APHIS does not regulate the movement of conifer wood or wood products (APHIS, 2007b).

Cut Foliage, Flowers, and Christmas Trees

Phytophthora ramorum readily sporulates on *U. californica* leaves under moist, temperate conditions (Davidson *et al*, 2003a); chlamydospore formation has also been observed. In a laboratory study, Linderman and Davis (2007a) found varying degrees of infection and sporulation by *P. ramorum* on all species, cultivars, and hybrids of *Camellia*. Chastagner *et al*. (2008) found *P. ramorum* on flowers of *U. californica* and flower peduncles of *Phoradendron serotinum* ssp. *macrophyllum*; Tjosvold *et al*. (2006b) reported infection of *Camellia* flower buds. This pathogen infects small branches of *P. menziesii* var. *menziesii* and small branches and needles of *S. sempervirens*. In 2004, 665 Christmas tree plantations in Oregon and 100 in Washington were surveyed and found negative for *P. ramorum* (COMTF, 2004b). Twenty of the conifer species tested, including many important species used as Christmas trees, were susceptible to *P. ramorum* (Chastagner *et al.*, 2004). Cut flowers and foliage are less likely to come into contact with live hosts because much of the discarded material will end up in landfills, whereas discarded Christmas trees are more likely to end up as greenwaste.

Greenwaste/Compost/Potting Media/Soil

Phytophthora ramorum has been detected from greenwaste (Shelly et al., 2005a), compost (Garbelotto, 2003a), and potting media (Parke et al., 2006b). Greenwaste containing host material from infested areas may serve as a source of spores, especially leaves of foliar hosts (Davidson and Shaw, 2003). Green material dried for several weeks, such as rhododendron leaves, will still sporulate upon wetting (Garbelotto, 2003a). Although it has not been demonstrated, it is likely that spores could be dispersed from foliar hosts via rain-splash during transit in open containers, or that infected leaves could detach and blow away (Davidson and Shaw, 2003). Linderman and Davis (2006a, c) compared the survival of P. ramorum with other Phytophthora species in a variety of media (river sand, Douglas-fir bark, coir, sphagnum peat, redwood sawdust, a bark-peat-pumice potting mix, a dairy compost, and a garden soil) and found that the pathogen was recovered from all substrates for six months.

Recovery rates of *P. ramorum* in areas with host plants were equal from soil samples collected on and off hiking trails. The pathogen was only recovered from samples collected from the trails in two areas without hosts, suggesting anthropogenic movement of the pathogen along the trails (Cushman and Meentemeyer, 2006). Fichtner *et al.* (2007a) indicate current baiting techniques can underestimate the amount of inoculum present in the soil.

The risk rating for Establishment and Spread is High for Nursery Stock, Wood and Wood Products, Greenwaste, Compost, Potting Media, and Soil. The risk is Medium for Cut Christmas Trees because the negative detections in nursery surveys show a lack of association with the pathway. Cut Flowers and Foliage are also rated Medium because the intended use does not put them into contact with suitable hosts in suitable environments. There is uncertainty in ratings for Cut Christmas trees and Cut Flowers and Foliage because the species used are susceptible.

Subelement 3: Detection Potential

Species of *Phytophthora*, such as *P. ramorum*, are difficult to detect at the ports–of-entry, where visual inspection is the primary method of detection; *Phytophthora* spp. have only been detected seven times since 1985 (PPQ, 2007a). However, Brown and Brasier (2007) have used ELISA to detect species of *Phytophthora* in the field; this may have applicability at ports-of-entry. Other pathogens and environmental conditions can elicit the same symptomology in foliar and dieback hosts. Two new *Phytophthora* species, *P. nemorosa* and *P. kernoviae*, induce similar cankers on trees and were found as a result of field analyses for *P. ramorum*. *P. nemorosa* occupies a similar ecological niche to *P. ramorum* in the United States (Hansen *et al.*, 2004), and *P. kernoviae* occupies a similar niche in the United Kingdom (Brasier *et al.*, 2005; DEFRA, 2004d). Eradication efforts at nurseries and in forests are not always successful. Soil still harbored *P. ramorum* three years after initial eradication efforts in Curry County, Oregon (Hansen *et al.*, 2005), and the pathogen resurfaced at nurseries in the United States and the United Kingdom even after prescribed control measures had been completed (APHIS, 2005b; DEFRA, 2005b).

Isolation techniques, including direct plating and baiting, are used to detect the pathogen in plant tissues, soil, and water. The efficacy of these techniques varies with season and host (Davidson et al., 2002c; Fichtner et al., 2007a). Molecular detection techniques include ELISA (at the genus level), AFLP, and a variety of PCR protocols. A real-time PCR method is currently being used for regulatory purposes in both the United States and the United Kingdom. The ITS DNA analysis does not always distinguish P. ramorum from P. lateralis; however, multiplex methods can increase sensitivity. The possibility of latent infections is a concern.

Newly established populations may go undetected. The disease was first noted in California in 1995 (Garbelotto *et al.*, 2001); researchers suggest the pathogen was introduced at least five years before the first detection (Rizzo and Garbelotto, 2003).

Nursery Stock (Including Living Christmas Trees), Cut Foliage/Flowers, Cut Christmas Trees

Visual diagnosis is still typically the first step in the detection of *P. ramorum* and can be complicated by other factors. For example, environmental conditions and other pathogens produce similar symptoms (Davidson *et al.*, 2003b), and fungicides commonly used to control other *Phytophthora* species on rhododendron may mask symptoms of *P. ramorum* (Davidson and Shaw, 2003). In addition, in infested nurseries soil or mulch in the pots of *Rhododendron* plants, other host plants, and even non-host plants that appear healthy may contain spores of *P. ramorum* (Davidson and Shaw, 2003; Parke *et al.*, 2007; Shishkoff, 2007) and be a source for reinfestation (DEFRA, 2005b). Chastagner *et al.* (2006a) reported that the application of contact fungicides in laboratory trials did not limit the recovery of *P. ramorum* from inoculated conifer

hosts, although growth was slower. There are also recent reports of asymptomatic infection and sporulation (Denman *et al.*, 2008; Vettraino *et al.*, 2007).

Wood and Wood Products

Detection methods for assessing wood products present unique challenges. Direct isolation on a semi-selective medium or baiting has been used to recover the pathogen from symptomatic wood and bark. The efficacy of these methods depends on the host and time of year. Isolation frequencies from wood tend to be lower than from other plant parts. Recovery was increased by taking plates of the semi-selective medium (PARP) to the field; however, 60% of the samples were negative (Storer *et al.*, 2002). The pathogen could not be isolated from wood chips after air drying for two weeks (Swain *et al.*, 2002), but lack of isolation is not definitive evidence that the pathogen is devitalized. The most sensitive detection method, PCR, detects the presence of the DNA, but does not indicate the viability of the pathogen. Recently, Brown and Brasier (2007) were successful at detecting several *Phytophthora* species, including *P. ramorum*, in wood using direct plating (within 4–24 hours post-collection and storage of 4-10°C) isolation and lateral flow ELISA kits.

Greenwaste/Compost, Soil, and Potting Media

Fichtner *et al.* (2006a, 2007a) found that current soil baiting techniques are adequate to detect sporangia but not chlamydospores in soil and thereby underestimate the amount of inoculum present. The same baiting techniques are used to recover *P. ramorum* from greenwaste, compost, and potting media.

The risk rating for Detection Potential is High for all pathways. The sensitivity and specificity of these methods vary with season, host, host part, and pathogen propagule.

C. Pest Risk Potential

The overall risk presented by *P. ramorum* is High due to the number of pathways moving the pathogen and the associated uncertainties, *e.g.*, long-term viability of infective propagules, difficulty in detection of the propagules, lack of definitive host range, and means of natural movement. Research is needed on: (1) factors inducing germination of chlamydospores; (2) improved detection protocols; (3) improved mitigation measures; (4) temperature requirements for survival of propagules in various sources, *e.g.*, soil, wood; (5) risk associated with various species and hybrids of plants for planting, *e.g.*, Christmas trees (cut and uncut), cut flowers and cut foliage; (6) screening for more potential hosts, including products and propagative material of vegetable, fruit, and nut crops; (7) natural dispersal, especially animal-aided and aerial dispersal; and (8) the origin and geographic distribution of *Phytophthora ramorum*.

Table 6. Comparative risk matrix for *Phytophthora ramorum* assessment and selected unmitigated pathways.

•	Organism Assessment	Pathways							
Risk Element/ Subelement		Nursery stock	Wood/Wood Products	Cut Christmas Trees	Cut Flowers/ Foliage	Greenwaste/ Compost	Potting Media	Soil	
Consequences of Introduction									
Climate/Host Interaction	High	High	High	High	High	High	High	High	
Host Range	High	High	High	Medium	Medium	High	High	High	
Dispersal Potential	High	High	High	Medium	Medium	High	High	High	
Economic	High	High	High	High	High	High	High	High	
Environment	High	High	High	High	High	High	High	High	
Likelihood of Introduction									
Pest Opportunity	High	High	High	High	High	High	High	High	
Entry Potential	High	High	High	Medium	Medium	High	High	High	
Spread/Establishment	High	High	High	High	High	High	High	High	
Detection Potential	High	High	High	High	High	High	High	High	
Risk Potential	High	High	High	High	High	High	High	High	

VI. Mitigation Measures

A. Introduction

The risks associated with the importation and domestic movement of hosts and products from hosts of *P. ramorum* from infested areas without specified growing, inspection, and certification requirements were analyzed to be High. A risk potential of High necessitates the implementation of appropriate mitigation measures.

Traditional regulatory mitigation measures for pests and plant pathogens consist of exclusion, containment, suppression, and eradication (Kahn, 1991). Where specific efficacies were not known for *P. ramorum*, they were deduced from the biology and management of other *Phytophthora* spp. Diagrams for the foliar host (Fig. 4), canker host (Fig. 5), soil (Fig. 6), and dormancy phase (Fig. 7) include points where mitigation measures may be applied. Rizzo *et al.* (2005) reviewed available research and suggested management options.

It is difficult to design management strategies for forests (Rizzo *et al.*, 2005). Disease management requires an understanding of forest ecology and pathogen biology and ecology.

Large scale management efforts have met with varying levels of success (Rizzo et al., 2005). Clear-cutting was attempted to slow the spread of chestnut blight (*Cryphonectria parasitica*) (Anagnostakis, 1987); removal of alternate host material for control of white pine blister rust (*Cronartium ribicola*) (Kinloch, 2003); sanitation programs for Dutch elm disease (*Ophiostoma ulmi*, *O. novo-ulmi*) in North America and Europe, *P. cinnamomi* in Australia, and *P. lateralis* in Oregon and California (Hansen et al, 2000; Hardy et al., 2001); and fungicides to reduce the spread of *P. cinnamomi* in Australia (Hardy et al., 2001).

The divergent goals of various stakeholder groups add to the complexity of managing *P. ramorum* in forests. Where timber production is not a major goal, management has been directed toward watersheds, fuel load, wildlife, and aesthetics (Rizzo *et al.*, 2005). Where timber production is a goal, management plans must also incorporate riparian preservation and

endangered species concerns. Ultimately, any forest management strategy must integrate prevention, treatment, restoration, and conservation (Rizzo *et al.*, 2005).

Prevention of human-mediated spread has been focused, nationally and internationally, on quarantines on nursery plants and plant products (APHIS, 2005). There are reports of disease outbreaks at the urban-wildland interface associated with the planting of ornamental rhododendrons in California (Rizzo *et al.*, 2005). Similar associations have been observed in Europe with *Rhododendron* planted near tree hosts (Brasier *et al.*, 2004). Other pathways for human-mediated transport include soil, green waste, and stream water (Cushman and Meentemeyer, 2006; Davidson *et al.*, 2005; Garbelotto, 2003a; Tjosvold *et al.*, 2002b).

The following portion of the document contains an overview of the mitigation measures in place for *P. ramorum*, as well as mitigation approaches that may be used to address the major pathways identified for its movement. The pathways are nursery stock, Christmas trees, cut flowers and foliage, wood and wood products, greenwaste, compost, soil, and potting media.

B. Regulatory Measures

Exclusion

Exclusion of *P. ramorum* is facilitated by large geographical barriers. The caducous sporangia and zoospores are easily dispersed, locally, in rain events, and have been postulated to move long distances by significant weather events (Hansen *et al.*, 2002). Sporangia of other *Phytophthora* species, *i.e.*, *P. infestans*, do not survive long-distance dispersal because viability is decreased under dry conditions (Ristaino and Gumpertz, 2000).

Exclusion of the A1 mating type from Europe is possible by prohibiting the entry of living plant hosts and untreated plant-derived products, compost, and potting media. Based on the general biology of heterothallic *Phytophthora* species, more virulent strains can result from genetic recombination (Erwin and Ribeiro, 1996), *e.g.*, *P. infestans* (Smart and Fry, 2001). Limited introductions of European-type isolates of *P. ramorum* have already occurred in North America (Grunwald, 2007) and there is evidence that at least one of these isolates is more aggressive than the North American isolates (Garbelotto *et al.*, 2004; Parke *et al.*, 2004a). Exclusion of *P. ramorum* from non-infested areas is possible by prohibiting movement of all hosts (providing all hosts have been identified) from infested areas.

APHIS implemented emergency phytosanitary measures to restrict the movement of nursery stock and other plant materials from all European Union member states due to concerns about *P. ramorum*. In addition to the current measures in 7 CFR 319.37, the following apply to propagative plant materials of all *P. ramorum* host plants: 1) the National Plant Protection Organization (NPPO), or an agency accredited by the NPPO, must conduct an annual survey of nurseries exporting propagative plant materials of host plants to determine that those nurseries are free of *P. ramorum*; 2) the NPPO, or an agency accredited by the NPPO, must inspect all shipments of host materials exported to the United States, and must sample and test plants bearing symptoms of *P. ramorum*; and 3) propagative plant materials of host plants must be accompanied by a phytosanitary certificate stating that the plants originate in a nursery which is inspected and tested annually and found free of *P. ramorum* and that the plants have been inspected prior to export and found free of the pathogen (Aley, 2007).

Containment

Under current Federal domestic regulations (7 CFR §301.92), nurseries in the quarantined areas must be inspected, sampled, and tested annually for symptoms of *P. ramorum*. In addition, preshipment inspections are required prior to interstate movement. The Emergency Federal Order Restricting Movement of Nursery Stock from California, Oregon, and Washington Nurseries (Dec. 21, 2004) and 7 CFR §301.92 also require nurseries in regulated areas of California, Oregon, and Washington to have annual and pre-shipment inspections of host materials prior to interstate shipment. If the pathogen is detected during any inspection process, eradication efforts are initiated.

Suppression

Efforts to prevent the spread of *P. ramorum* from the quarantined counties of California have focused on educational outreach, the seasonal closure of trails (COMTF, 2005), and facilities for soil removal from shoes (Davidson *et al.*, 2005) and bicycle tires (Tjosvold *et al.*, 2006a).

In Humboldt County, California, efforts to reduce inoculum load by removing infected trees have been initiated (COMTF, 2004c). More recently, a cooperative effort among State and Federal agencies and industry has initiated a project to suppress the northward movement of *P. ramorum* by creating a "No Host Zone" (Cannon, 2007). Currently, herbicides are being used to kill *L. densiflorus* in a band seven miles long by two miles wide along the Van Duzen River. This band is 20 miles north of the most northern forest detection in California.

Suppressive mitigation measures used for other *Phytophthora* species include sanitation, disinfectants, fungicides, fumigants, methods of water treatment and distribution, and type and form of bed beneath the pots (Erwin and Ribeiro, 1996; Hartmann *et al.*, 2002). Suppression efforts for nursery stock and live Christmas trees focused on surveys, the development of best management practices, and education of producers (Suslow *et al.*, 2005, 2008) and are currently part of the eradication efforts described below.

Eradication

Removal and destruction of plant material and related articles are the major eradication efforts for *P. ramorum* in forests and nurseries. When infected plants are found in forests, a buffer area of at least 0.25 miles from the outermost infected plants is used to establish the quarantined area (APHIS, 2006); in Oregon a buffer area of 0.5 miles is currently used (OSOS, 2007). The only current effort to eradicate *P. ramorum* from a forest setting is underway in Curry County, Oregon (Goheen *et al.*, 2002a; Goheen *et al.*, 2007; OSOS, 2007). The recent detection in Curry County of *P. ramorum* in soil six years after host eradication (Kanaskie *et al.*, 2008), coupled with research evidence that *P. ramorum* has a soil phase (Fichtner *et al.*, 2007a, b; Parke *et al.*, 2004a, 2006b; Shishkoff, 2006; Shishkoff and Tooley, 2004), suggests that additional eradication measures may be needed.

The APHIS-confirmed nursery protocol requires a delimiting survey of the entire nursery, including inspection of all host and associated plant (HAP) genera, including plants for sale or propagation (APHIS, 2007c). All HAP genera within 2 meters of infected plants will be destroyed by incineration, deep burial, or a combination of steam sterilization and deep burial. All HAP genera within 10 meters of the positive block(s) shall be considered exposed to *Phytophthora ramorum* and shall be held for the 90-day quarantine period (90 days of conducive environmental conditions). In addition to plants, water, media components, and soil will be

sampled during the delimiting survey. Positive samples will be treated according to the protocol (APHIS, 2007c). The USDA has developed a protocol in response to *P. ramorum*-infected plants installed in the landscape (APHIS, 2007e).

Eradication of the pathogen via chemicals is problematic. The pesticides available for control of *Phytophthora* species (Table 7) are fungistatic and not fungicidal. Additionally, metalaxyl resistance has been detected in *P. ramorum* (Rizzo *et al.*, 2005). Herbicides are being used to kill host plants to prevent inoculum survival by the pathogen in forests in Curry County, OR and Humboldt County, CA (Goheen *et al.*, 2007; Cannon, 2007).

Sanitation

Sanitation in all stages of propagation is necessary to maintain pathogen-free material (Pegg, 1978 and Hansen, 1970 *in* Erwin and Ribeiro, 1996; Jones and Benson, 2001). For control of a polycyclic foliar pathogen like *P. ramorum* in the field, sanitation needs to be 99.9% effective (Van der Plank, 1963). Sanitation practices should include removal and testing of symptomatic nursery stock, sterilization of potting media, and disinfection of tools, benches, workers' shoes and gloves, and other equipment. All symptomatic material or diseased plants should be disposed in a sanitary landfill, incinerated, or otherwise treated to prevent the spread of *P. ramorum*.

Garbelotto *et al.* (2003) found that 9-12 hours of leaf wetness at 18-22°C are needed to obtain significant infections on *U. californica* leaves. Contaminated irrigation and contaminated recycled water disperse *Phytophthora* propagules, either directly by delivering contaminated water or indirectly by splashing inoculum from plant and ground surfaces to other plants (Erwin and Ribeiro, 1996; Werres *et al.*, 2007). Methods to disinfest water are available and include chlorine products, filters, and ozonation (Hartmann *et al.*, 2002). Water treatment should be coupled with testing before and after treatment. The recycling of irrigation water has been adopted for environmental reasons, but this process increases the risk of spreading the pathogen.

Chemical control would include fungicides and disinfectants for benches, tools, and equipment. Sodium hypochlorite is a commonly used source of chlorine that is suitable for these surfaces, but can be phytotoxic (Hartmann *et al.*, 2002; Jones and Benson, 2001). Pesticides are available and registered for use with *Phytophthora* species (Table 7), but these products are fungistatic and not fungicidal.

Table 7. Fungicides labeled for control of *Phytophthora* diseases¹.

ACTIVE INGREDIENT	PRODUCT	COMPANY	REGISTERED
Azoxystrobin	Amistar	Syngenta	Vegetable crops
Chlorothalonil	Daconil Ultrex	Syngenta	Ornamentals
Chlorothalonil	Echo 720 T&O	SipCam Agro	Turf, ornamentals
Copper hydroxide	Champ Formula 2 flowable, wp	Nufarm	Ornamentals
Copper hydroxide	Champion WP	Nufarm	Ornamentals
Copper hydroxide	Kocide 2000 T/N/O	Griffin	Turf, ornamentals
Dimethomorph	Acrobat 50WP, MZ	BASF	Potatoes
Etridiazole	Banrot 40WP, 8G	Scotts	Ornamentals
Etridiazole	Terrazole 35WP	Crompton-Uniroyal	Ornamentals
Etridiazole	Truban 25EC, 30WP, 5G	Scotts	Turf, ornamentals
Fluaxinam	Omega 500F	Syngenta	Peanuts and potatoes
Mancozeb	Dithane 75 DF	Dow	Ornamentals
Mancozeb	Fore	Dow	Turf, ornamentals
Mancozeb	Gavel 75 DF	Dow	Vegetable crops
Mancozeb+Cu(OH)2	Mankocide	Griffin	Fruits and vegetables
Mefanoxam	Apron XL LS	Syngenta	Vegetable crops
Mefanoxam	Mefanoxam 2	SipCam Agro	Ornamentals
Mefanoxam	Ridomil Gold	Syngenta	Fruits and vegetables
Phosphonate	Aliette WDG Chipco	Bayer	Turf, ornamentals
Phosphonate	Phostrol	Nufarm	Fruits and vegetables
Phosphonate	Vital	Griffin	Ornamentals
Propamocarb	Banol	Bayer	Turf, ornamentals
Pyraclostrobin	Cabrio EG	BASF	Fruits and vegetables
Pyraclostrobin	Headline	BASF	Vegetable crops
Trifloxystrobin	Flint	Bayer	Fruits and vegetables

¹This list is not comprehensive and does not constitute an endorsement by USDA of any products listed here.

A series of best management practices based on epidemiological factors could include multiple mitigations, such as pathogen-free propagating material, a pathogen-free water source, clean potting media, pots, a strict sanitation protocol including cleaning and testing of benches and beds, and cleaning of tools, equipment, shoes, hands, *etc*. The Nursery Committee of the California Oak Mortality Task Force (COMTF) began developing best management practices in 2004, which were modified in 2005 (Suslow *et al.*, 2005).

The State of Oregon established regulations in 2001 prohibiting the entry of products of susceptible oaks from California unless they have been kiln-dried or heat-treated to 71.1°C for 75 minutes measured at the core (ODA, 2001). Oregon required that soil associated with oak commodities be sterilized by dry heat at 110°C for 16 hours (ODA, 2001). After the quarantine was enacted in Oregon, the pathogen was detected at several sites in Curry County; all infected plants are being burned on site. The eradication efforts in Curry County are cooperative among State and Federal agriculture and forestry agencies.

C. Nursery Stock, Christmas Trees, and Cut Foliage/Flowers

Chemical Treatment

Linderman *et al.* (2006b) evaluated fungicides labeled for use on several *Phytophthora* species (*P. cactorum, P. citricola, P. nicotiana, P. citrophthora*, and *P. ramorum*). Systemic and translaminar fungicides were effective in disease suppression but were not effective as eradicants. Of all fungicides tested, menfenoxam was the most effective on all of the species tested, with the exception of *P. citrophthora*.

Tjosvold *et al.* (2006a) also evaluated registered products on *Rhododendron*, *Camellia*, *Pieris*, and *Viburnum*. Those products most efficacious on *Rhododendron* were selected for timing of application studies. Maximum rates of mefenoxam (metalaxyl-M), dimethomorf, pyraclostrobin, and fenamidone were applied as foliar sprays on wounded and non-wounded leaves. Preventative activity was observed for two weeks but not for four weeks. Post-infection treatments were ineffective. Only dimethomorf significantly reduced the success of isolation recovery from lesions. Metalaxyl –M, azoxystrobin, and fenamidone/mancozeb completely inhibited symptom development on *Rhododendron* spp. (Turner *et al.*, 2006). Heungenis *et al.* (2005) tested the efficacy of metalaxyl, dimethomorf, cyazofanid, fosphetal Al, cymoxanil, and mancozeb to control *P. ramorum* on *Rhododendron*. Metalaxyl, dimethomorf, and cyazofanid were the most effective, fosphetal Al, and cymoxanil were intermediate, and mancozeb was least effective. The best control was achieved when the lower leaf surface was covered with the fungicide. Fungicides were better as protectants and not effective as curatives (Heungenis *et al.*, 2005).

Chastagner *et al.* (2006a) evaluated 20 systemic and contact fungicides on seedlings of *Pseudotsuga menziesii* var. *menziesii*. A drench application of mefenoxam prior to bud break prevented infection, and post-bud break applications of mancozeb, maneb, and metiram provided 100% control. Variable results were obtained with other fungicides. The surfactant Latron CS-7, applied at post-bud break, yielded 60-100% reduction in infection. A concern associated with the potential use of fungicides to control this disease is the possibility that fungicides might suppress symptom development on infected plants. Systemic fungicides might have the potential to suppress symptom development, but this is not likely with the contact types of fungicides found to be effective in protecting seedlings from *P. ramorum* (Chastagner *et al.*, 2006a).

Dimethomorph and phosphate were applied to *V. ovatum, L. densiflorus, R. macrophyllum*, and *U. californica* in the field at one and two times the recommended rates. Detached leaves were taken to the laboratory for wound inoculation assays. No treatment provided complete protection (Goheen *et al.*, 2006a).

Biological Control

Bacillus brevis and Paenibacillus polymixa were tested for antagonistic activity against five *Phytophthora* species, including *P. ramorum*. Both antagonists significantly inhibited *P. ramorum in vitro*, but were ineffective in inoculation assays of leaves dipped in a cell suspension of each *Phytophthora* species (Linderman and Davis, 2006b). Out of 100 fungi bacteria and antinomycetes isolated from soil, leaf surfaces and plant parts, 15 microorganisms inhibited *P. ramorum* in culture; six of these organisms completely inhibited zoospore germination while one enhanced germination. Percentage leaf necrosis was less for all six than the water control, but none completely inhibited necrosis. (Widmer, 2007).

Cultural Control

Sanitation during and after propagation is necessary to maintain and monitor pathogen-free material (Pegg, 1978 and Hansen, 1970 in Erwin and Ribeiro, 1996). For control of a polycyclic foliar pathogen such as *P. ramorum* in the field, sanitation needs to be 99.9% effective (Van der Plank, 1963 *in* Erwin and Ribeiro, 1996). Sanitation practices should include removing and testing symptomatic stock, sterilizing potting media, and disinfecting tools, benches, and workers' shoes, gloves, and equipment (Erwin and Ribeiro, 1996). All symptomatic material or diseased plants should be disposed in a sanitary landfill or otherwise treated to prevent the spread of *P. ramorum*. Micropropagation media have been demonstrated to support the growth of the pathogen, facilitating detection (Linderman and Davis, 2007b).

Irrigation water can be a pathway for dissemination of *P. ramorum* (Werres *et al.*, 2007), especially in water that is re-circulated. A source of pathogen-free water is necessary to prevent infection. A variety of methods to disinfest water exist, including ozonation, chlorination, filtration, and UV irradiation (Jarvis, 1992; von Broembsen, 2005). Kaminski *et al.* (2006) built and tested three different filtration systems for the non-chemical elimination of *P. ramorum*.

Physical Control

The combined use of heat and vacuum prevented the recovery of *P. ramorum* from *U. californica* leaves while maintaining the volatiles needed in the leaves for cooking. However, the lack of recovery of the pathogen does not necessarily mean that the pathogen has been devitalized (Harnik *et al.*, 2004). Linderman and Davis (2005, 2006a, c) demonstrated that *P. ramorum* can readily survive in potting media or soil after deliberate contamination with culture-produced sporangia or chlamydospores. They detected the pathogen for six months by baiting or direct plating from all contaminated substrates. *P. ramorum* sporangia survived best in peat moss, potting mix, coir, and Douglas fir bark, and poorest in sand or soil. They also found that the use of heat via aerated steam mixtures at temperatures of 50°C or higher for 30 minutes was an effective means of eradicating *P. ramorum* from infested media and contaminated containers without destroying the containers (Linderman and Davis, 2006a).

Best Management Practices

The Nursery Committee of the COMTF has formulated best management practices to control or eliminate diseases caused by *P. ramorum* (Suslow *et al.*, 2005). They divided the practices into three categories: exclusion, prevention, and monitoring.

Suslow (2008) reported that there were three factors responsible for the decline in nursery detections: the Federal Order, grower education, and critical nursery research. Pilot programs are being initiated to validate selected best management practices (COMTF, 2007; ODA, 2007; Suslow, 2008).

Breeding for Resistance

Several studies testing host susceptibility suggest resistance to *P. ramorum* is present in several taxa. De Dobbelaere *et al.* (2006) screened 21 species and 42 hybrids of *Rhododendron* for susceptibility to *P. ramorum* using four inoculation methods (wounded or non-wounded detached leaves and wounded or non-wounded branches). Significant differences in disease susceptibility were observed among species as well as among hybrids with all methods used. Inoculation of wounded leaves and stems showed that most species and hybrids were susceptible to some extent. Inoculation of non-wounded leaves and/or stems resulted in a large degree of

variation in susceptibility. The results suggested that if significant resistance is present, it probably occurs at the level of tissue penetration. Shishkoff (2007b) evaluated the relative susceptibility of 25 species and cultivars of *Syringa* to *P. ramorum* using detached leaf assays. The cultivar tested had a significant effect on percent lesion area. Linderman *et al.* (2006) evaluated the effect of different species of *Phytophthora* and isolates of *P. ramorum* (both mating types) on detached leaves of *Rhododendron*, *Syringa*, and *Viburnum* inoculated under controlled conditions. They found significant differences in virulence among *Phytophthora* species and *P. ramorum* isolates.

D. Wood and Wood Products

Woody canker hosts are unique in that sporulation is not observed on the surface of cankers; however, the pathogen can be isolated from bark and wood (Davidson *et al.*, 2003b). The pathogen was also recovered from firewood stored for six months (Shelly *et al.*, 2005a). Sporulation in baiting trials was stimulated when inoculated "logs" were kept at 12°C prior to baiting (Garbelotto, 2002). Studies are needed to determine if chlamydospores or dormant mycelia are produced in phloem and xylem, and if so, whether these forms of inocula are destroyed by natural or kiln drying.

Physical Treatments

There are a few treatments available to mitigate this pathogen in wood: physical removal of infected bark and wood, air drying, and heat treatment. *Phytophthora ramorum* has been detected in phloem and xylem of multiple tree hosts (Brown and Brasier, 2007; Parke *et al.*, 2007). Mycelium was found in multiple cells types and chlamydospores were found in the xylem vessels of *L. densiflorus* (Parke *et al.*, 2007). Chlamydospores are considered a survival stage for *Phytophthora* spp. (Erwin and Ribeiro, 1996), but the specific role of this structure in the disease cycle of *P. ramorum* is incompletely known (Fichtner *et al.*, 2007a).

Debarking is a standard quarantine treatment for the movement of logs and lumber. However, Brown and Brasier (2007) have shown that *P. ramorum* is often active and can remain viable up to 25 mm into the xylem, and Parke *et al.* (2007) have found *P. ramorum* at depths of 4 cm. These data suggest that a more stringent treatment is required to prevent the spread in wood. They recommend removal of at least 3 cm of outer sapwood, and where quarantine issues arise it may be preferable to destroy the infected trees (Brown and Brasier, 2007).

Prescribed periods and conditions for air-drying or heat treatment are possible mitigation procedures for wood products. In the laboratory, *P. ramorum* was not recovered from infected wood chips after two weeks of drying at 55°C (Swain *et al.*, 2005). It is known that the core temperature of piles of bark used for commercial mulch exceeds 55°C (Titko, 2003) indicating a potential mitigation. One preliminary study reports that 30 minutes at 56°C, the IPPC standard treatment for wood drying, may not be sufficient to devitalize the pathogen (Tubajika *et al.*, 2008. However, Swain *et al.* (2005, 2006) found that cultures of *P. ramorum* were killed after exposure to 45°C for 24 hours or 55°C for one hour and that the temperatures in compost piles devitalized the pathogen. Microbial competition or other biological activity or products resulting from digested plant material may play a role in reducing inoculum (Hoitink and Fahy, 1986). Recently, a limited study using a radio frequency treatment on three tree genera indicated some success at controlling certain wood decay and sapstaining fungi (Tubajika *et al.*, 2006); however, this technique has not been tested against *P. ramorum*.

Chemical Control

P. ramorum is sensitive to copper hydroxide, metalaxyl, phosphate, phosphites, and phosphonates (Garbelotto et al., 2002c; Harnik and Garbelotto, 2006; Schmidt et al., 2006). Copper hydroxide was effective up to six weeks post-treatment on U. californica (Harnik and Garbelotto, 2006). On Quercus spp. and L. densiflorus, phosphite injections and topical applications significantly reduced lesion size. However, Kanaskie et al. (2006) found variation in application method and effects on lesion size and location on L. densiflorus. A combination of injection and topical applications of phosphonate on Q. parvifolia var. shrevei was more effective than either treatment alone (Schmidt et al., 2006). All three treatments were more effective than the control for L. densiflorus, and dosage may be as important as the application method. Range in susceptibility of the hosts to the pathogen may affect the outcome of the treatment (Schmidt et al., 2006).

Methyl bromide has been used as a fumigant for wood products, but the data on control of fungi and related organisms in wood are limited. However, methyl bromide has a long history for soil fumigation in the field and greenhouse (Erwin and Ribeiro, 1996). It has commonly been used in combination with chloropicrin for control of *Phytophthora* species and other pests in strawberry beds (Wilhelm and Paulus, 1980), and has been used as a soil treatment for the mitigation of *P. cinnamomi* in citrus groves (Menge and Nemec, 1997).

Magnusson *et al.* (2001) listed methods to mitigate the risk for other pathogens and pests in wood chips: heat treatment; pressure impregnation at temperature and pressures to kill fungi, insects, and nematodes; and in-transit shipboard fumigation. They also noted that economically feasible treatments for wood chips are currently lacking, leaving regulation of trade the sole strategy.

Cultural and Biological Control

A number of cultural methods are used to mitigate root rot and canker diseases of citrus caused by *P. cinnamoni* and *P. citrophthora*, including management of the source and the amount of nitrogen and water (Menge and Nemec, 1997). Elevated levels of calcium, phosphorus, iron, and copper are inhibitory to zoospores of these two species. Most of the measures are to control the root rot phase, but the nitrogen and water levels also affect the amount of succulent growth produced above ground. Menge and Nemec (1997) recommended cultural measures such as pruning low-hanging branches and removal of mulch from the trunk to eliminate moisture on the trunk and thereby prevent canker formation.

Avoiding both overwatering and excess nitrogen application is recommended to reduce the likelihood of infection by *P. ramorum* (Garbelotto, 2003c). Trees with higher water potentials are at a higher risk for infection than trees with less than optimal water potentials (Swiecki and Bernhardt, 2002a, b). Factors that encourage rapid growth of trees create natural openings and thinner cells in the outer bark, and may increase the efficiency of infection by *P. ramorum*.

In vitro laboratory research with biological antagonists indicated that control was possible, but field tests did not indicate control (Garbelotto, 2003c).

Breeding for Resistance

Levels of resistance are being detected both in and between populations of *U. californica* (Meshriy *et al.*, 2006) and *Quercus* (Dodd *et al.*, 2005; Hüberli *et al*, 2002). Work with a variety of hosts and *Phytophthora* species indicates strategies to use natural resistance. Menge and Nemec (1997) found that it was important to consider time of year, cultural factors, and tissue that is susceptible when screening for resistance.

The USDA Forest Service, Pacific Southwest Research Station, the Midpeninsula Open Space District, and University of California Berkeley are assessing levels of *P. ramorum* resistance in *L. densiflorus*. Acorns collected from stands starting at Big Sur into Oregon were used in field and laboratory studies to determine the source of and develop a reliable test for resistance (Frankel, 2007).

E. Greenwaste and Compost

Physical Control

Evidence exists that composting, as specified by the California Integrated Waste Management Board, may be an effective cultural control of *P. ramorum* in yard waste (Swain and Garbelotto, 2006; Swain et al., 2005). The minimum temperature required by the State of California for pathogen control in compost is 55°C for 3 days (CIWMB, 2007). Swain et al. (2005, 2006) found that cultures of P. ramorum were killed after exposure to 55°C for one hour or 45°C for 24 hours. Tests indicate that P. ramorum in greenwaste mulch is killed in compost after being held at 55°C for two weeks (Swain et al., 2006). Phytophthora ramorum could not be recovered by baiting from leaf and twig samples after tunnel composting at a minimum of 60°C for 10 hours (Aveskamp and Wingelaar, 2006). Similar temperatures can be reached in bark piles; therefore, a composting system may be developed (Titko, 2003). Additional information on chlamydospore biology, such as factors affecting germination, is needed before composting methods can be proven as effective control measures. Heat may not be the only factor detrimental to P. ramorum in the composting process. Microbial competition or other biological activity or products resulting from digested plant material may play a role in reducing inoculum (Hoitink and Fahy, 1986). Composting also requires a monitoring program to ensure that P. ramorum is not re-introduced (Garbelotto, 2003a). Research indicates that the source of the material may affect the ability of the composting process to devitalize P. ramorum (Swain and Garbelotto, 2006).

F. Potting Media and Soil

Domestic movement of nursery material allows for movement in plants in potting media; some mitigation measures for potting media are covered in section C.

Physical Control

Asymptomatic roots and infested potting media can harbor *P. ramorum* (Fichtner *et al.*, 2007b; Linderman and Davis, 2006a, c; Parke *et al.*, 2006b; Shishkoff, 2007). Aerated steam mixtures were tested for mitigation potential for *P. ramorum* and other pathogens in potting media in containers. *Phytophthora ramorum* could not be recovered from media subjected to aerated steam mixtures of 60°, 65°, or 75°C for 30 minutes (Linderman and Davis, 2006a).

A recent study of potential mitigation for *P. cinnamomi* in soil indicated prescribed burning did not attain sufficient temperatures for use as a control (McLaughlin *et al.*, 2007).

Chemical Control

Several soil fumigants are listed in APHIS protocols for mitigation in nurseries and landscapes with confirmed detections of *P. ramorum*: chloropicrin, methyl bromide, metam sodium, and Dazomet (APHIS 2004c, 2007c).

Methyl bromide has a long history for soil fumigation in the field and greenhouse (Erwin and Ribeiro, 1996). It has commonly been used in combination with chloropicrin for control of *Phytophthora* species and other pests in strawberry beds (Wilhelm and Paulus, 1980), and has been used for soil treatment for the mitigation of *P. cinnamomi* in citrus groves (Menge and Nemec, 1997).

Agrifos® (phosphonate) and Pentrabark® were approved in 2003 by the California Department of Pesticide Regulation for use together as a treatment for *P. ramorum* on *Quercus* spp. And *L. densiflorus*. The efficacy of this chemical varies with application method and the location of the pathogen in plant tissue (Garbelotto *et al.*, 2003; Kanaskie *et al.*, 2006; Schmidt *et al.*, 2006). Though phosphonate can be applied as a soil drench, Garbelotto *et al.* (2002c, 2003b) and Tjosvold *et al.* (2006a) found that method ineffective.

Cultural Control

Sanitation by removal of plant debris and humus reduced the level of *P. ramorum* recovered by baiting at the soil surface but did not affect recovery at 20 cm (Aveskamp *et al.*, 2006).

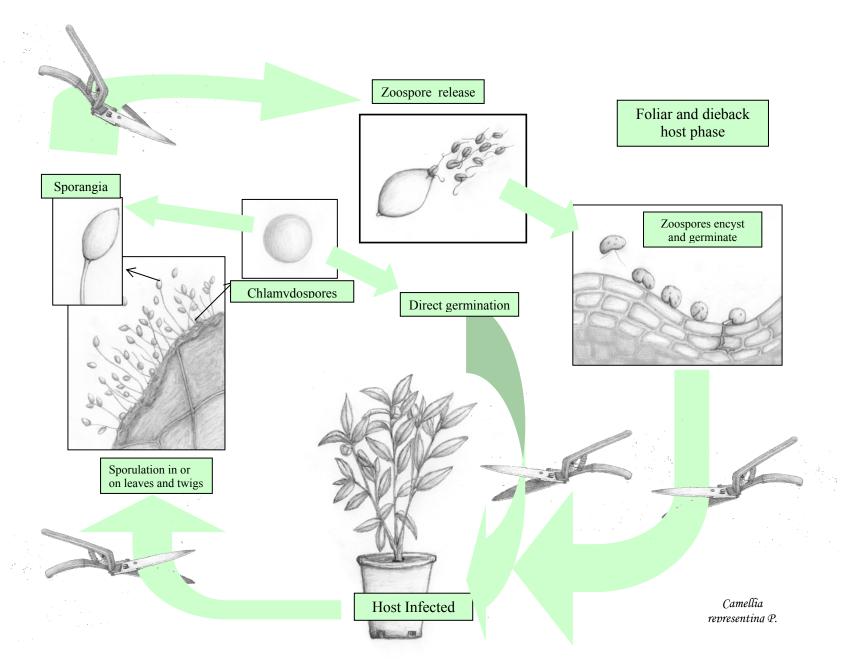


Fig. 4. Potential points for the application of mitigation measures for the foliar host phase are indicated with pruning shears

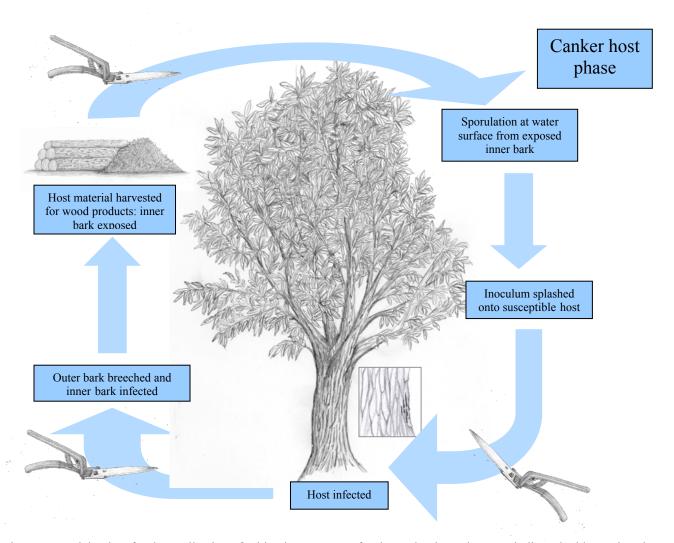


Fig. 5. Potential points for the application of mitigation measures for the canker host phase are indicated with pruning shears.

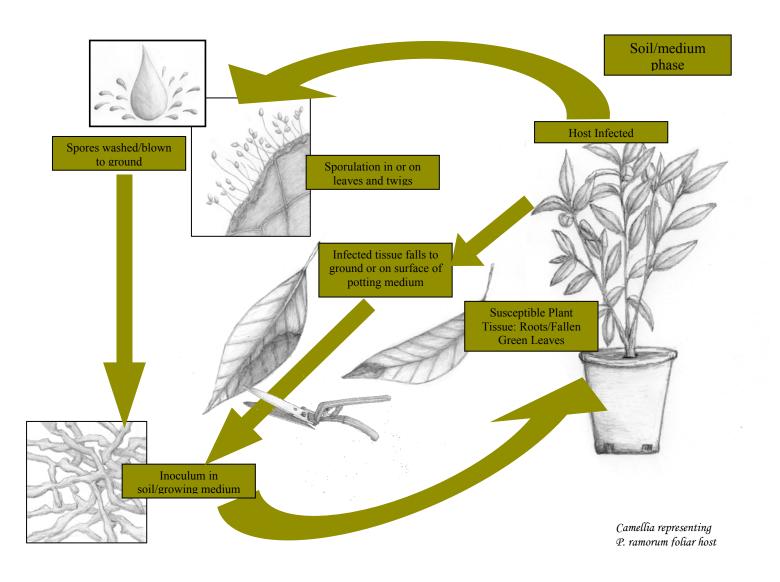


Fig. 6. Potential points for the application of mitigation measures for the soil phase are indicated with pruning shears.

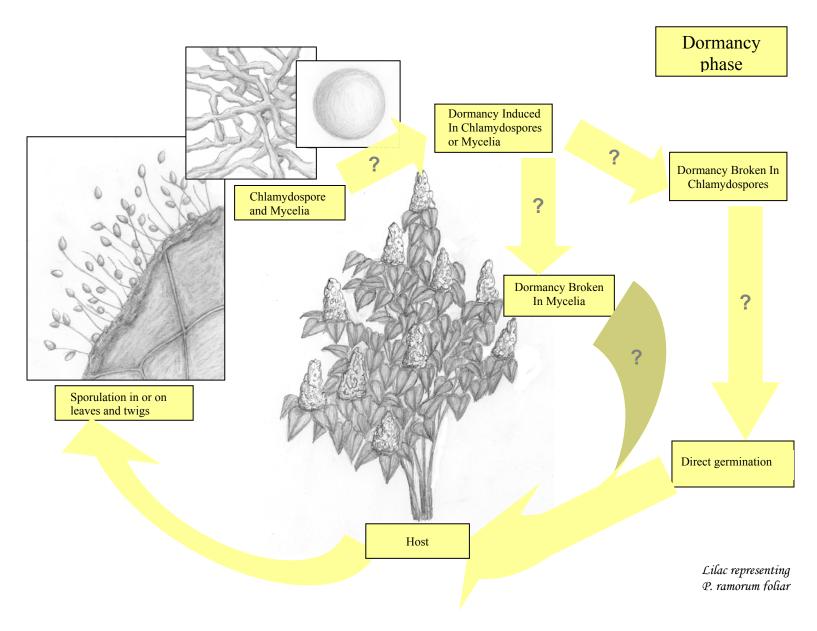


Fig. 7. Potential points for the application of mitigation measures for the dormancy phase are indicated with pruning shears.

VII. Acknowledgements

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