## Phytophthora ramorum Werres, de Cock & Man in't Veld Pest Risk Assessment for Oregon

This risk assessment (PRA) follows the format used by the Exotic Forest Pest Information System for North America. A risk assessment for the United States was prepared by Drs. G.L. Cave, B. Randall-Schadel, and S.C. Redlin, USDA Animal and Plant Health Inspection Service, on November 19, 2007.

# IDENTITY

Name: Phytophthora ramorum Werres, de Cock & Man in't Veld Taxonomic Position: Oomycota: Peronosporales Common Names: Sudden oak death (Phytophthora canker), Ramorum leaf blight, Ramorum dieback

# **RISK RATING SUMMARY**

Numerical Score: 9 Relative Risk Rating: VERY HIGH Uncertainty: Moderately Certain

**Uncertainty in this assessment results from:** The entire host range of this pathogen is unknown as is how the pathogen was initially introduced into Curry County, OR. Pathogen spread in Curry County has been limited by an active eradication and containment program. Thus, the epidemiology of the pathogen in Oregon's natural areas in the absence of disease management is not entirely understood.

### RISK RATING DETAILS Establishment Potential Is HIGH Criteria:

a. Organism has successfully become established in locations outside its native distribution.

c. Organism has demonstrated ability to utilize new hosts.

e. Organism has high inoculum potential or high likelihood of reproducing after entry.

# Justification:

Although the origin of *P. ramorum* is presently unknown, extensive genetic analyses indicate there are three clonal lineages present in the world today; EU1 first reported in Europe and NA1 and NA2 first reported in North America. These clones are genetically distinct from each other, but have little diversity within them. This supports the idea that *P. ramorum* is an introduced pathogen that has spread from its native habitat.

USDA APHIS presently recognizes 127 different plant species and genera as susceptible to *P. ramorum*. Koch's Postulates have been completed on 45 of those plants, while *P. ramorum* has been reported naturally infecting the other 82 species. DEFRA and the ODA and CDFA have recently reported *P. ramorum* infecting an additional four plant species, suggesting this pathogen's host range has yet to be determined.

Studies have been done indicating the high sporulation potential of *P. ramorum* on several plant species including California bay laurel, tanoak, and lilac.

# **Spread Potential Is HIGH**

### Criteria:

a. Organism is capable of dispersing more than several km per year through its own movement or by abiotic factors.

b. Organism has demonstrated ability for redistribution through human-assisted transport.

c. Organism has high reproductive potential.

d. Potential hosts have a contiguous distribution.

e. Newly established populations may go undetected for many years due to cryptic nature, concealed activity, slow development of damage symptoms, or misdiagnosis.

g. Organism has a broad host range.

### Justification:

Epidemiology studies in Oregon indicate that the majority of natural spread within a mixed tanoakconifer forest is within 300-m of a known infected tree. This same research demonstrated that jumps of up to 4-km may also occur.

*P. ramorum* has been reported infecting nursery plants shipped interstate and internationally. In Europe, disease spread from infected nursery stock into landscape trees has been demonstrated. In the United Kingdom, *P. ramorum* was discovered infecting trees in Japanese larch plantations. It has spread rapidly within these plantations, causing extensive mortality.

Studies have been done indicating the high sporulation potential of P. ramorum on several plant species including California bay laurel, tanoak, and lilac.

In Oregon, the natural ranges of the known oak, tan oak, and coast redwood tree hosts are limited mainly to the southwestern corner of the state and the southern Willamette Valley. Populations of wild rhododendrons, Pacific madrone, manzanita, and other hosts can be found scattered throughout the state. The natural range of evergreen huckleberry, big leaf maple, and Douglas-fir extends up the Oregon coastline into Washington and British Columbia. Populations are also found in the Cascade Mountain Range and, in the case of Douglas-fir, elsewhere in Oregon. Other known susceptible species are grown as landscape plants and nursery stock throughout the state. Thus, potential hosts are available throughout Oregon.

Current research indicates tree hosts such as tanoak may be infected for a 1-yr or more before appearing to suddenly die from the infection. Other *Phytophthora* species cause symptoms similar to Ramorum leaf blight and dieback on common nursery hosts, such as rhododendron. DNA testing or culturing is needed to distinguish *P. ramorum* infections from those caused by other Phytophthora species on these hosts.

USDA APHIS presently recognizes 127 different plant species and genera as susceptible to P. ramorum. Koch's Postulates have been completed on 45 of those plants, while P. ramorum has been reported naturally infecting the other 82 species. DEFRA and the ODA and CDFA have recently reported *P. ramorum* infecting an additional four plant species, suggesting this pathogen's host range has yet to be determined.

### Economic Impact Potential Is HIGH Criteria:

a. Organism attacks hosts or products with significant commercial value.

b. Organism directly causes tree mortality or predisposes host to mortality by other organisms.

d. Organism may cause loss of markets due to presence and quarantine-significant status. f. No effective control measures exist.

### Justification:

The nursery industry is Oregon's top agricultural commodity with \$820 million in gross sales in 2008. The timber industry harvested 2.748 billion board feet in 2009; Oregon is the nation's largest producer of softwood lumber. Economic analyses done in 2008 and in 2009 determined the potential economic impact of *P. ramorum* on the nursery and timber industries, respectively, in Oregon. If *P. ramorum* were allowed to spread unchecked from the current infested area in Curry County into the rest of the state, the adverse impact on the nursery industry was estimated to be \$64.93 - 652.30 million over 20-yr. Lost market share, nursery cultural control costs, and quarantine inspection costs were all considered important factors. Under a similar scenario, the adverse impact on the timber industry was estimated to be \$21.34 million to \$1.24 billion over 20-yr depending upon the rate of spread. Quarantine-related costs were considered the primary factor for the timber industry.

This pathogen is capable of killing healthy mature oak and tan oak trees, Japanese larch trees, wild rhododendrons, evergreen huckleberries, and *Viburnum* species. It can reportedly kill seedlings of Pacific madrone and coast redwood, while causing leaf spots and/or blights and tip dieback on mature plants. It also causes tip dieback on Douglas fir, thus affecting seedling establishment and plant growth. It causes disfiguring leaf spots on other host species, including several common nursery species, rendering the plants unmarketable.

Member countries of the European Union have adopted regulations for *P. ramorum*, as have Australia, Brazil, Canada, Chile, India, Israel, Korea, Mexico, New Zealand, Norway, South Africa, and Taiwan. The USDA APHIS has also adopted a federal interim rule for *P. ramorum*. This rule places 14 counties in California and a 419.6 km<sup>2</sup> area in Curry County under quarantine and regulates the movement of nursery stock from the three West Coast states (California, Oregon, and Washington) into the rest of the country. Oregon has adopted state regulations that mirror the federal interim rule.

Extensive research has gone into identifying control measures for *P. ramorum*. In Oregon, eradication has been attempted on known infection centers within the 419.6 km<sup>2</sup> guarantine area in Curry County with mixed success. In California, injection of trees with phosphite has been shown to suppress, but not eliminate infections. Foliar sprays with copper hydroxide may manage growth of *P. ramorum* in bay laurel leaves. Treatment of nursery plants with certain pesticides can provide protection from infection, although this was expressed primarily as reduced lesion growth. Fungicide treatment of already infected nursery plants did not significantly reduce lesion growth. Thus, virtually all pesticides tested on nursery stock have been fungistatic, not fungicidal. The pathogen may be eliminated from soil by fumigation or solarization to specified temperatures. The pathogen can be eliminated from plastic potting containers with aerated steam. Composting to EPA standards can eliminate *P. ramorum* from infected plant materials, although the pathogen can be reintroduced to mature compost. Vacuum plus heat treatment can eliminate infections from bay laurel leaves meant for human consumption. Cedar heartwood contains chemical agents that are sporicidal towards P. ramorum. The USDA APHIS PPQ requires enforcement of Confirmed Nursery Protocols (CNP) within retail and wholesale nurseries in which P. ramorum has been detected. The purpose of these protocols is to eradicate *P. ramorum* from the nursery site. However, several nurseries in multiple states have had subsequent detections of the pathogen

after undergoing the CNP.

# Environmental Impact Potential Is HIGH Criteria:

a. Organism is expected to cause substantial direct environmental effects, such as extensive ecological disruption or large-scale reduction of biodiversity.

c. Organism is expected to have indirect impacts on species listed by Federal, Provincial, or State agencies as endangered, threatened, or candidate. This may include disruption of sensitive or critical habitat.

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d. Organism may attack host with small native range.

e. Introduction of the organism would likely result in control/eradication programs that may have potential adverse environmental effects.

### Justification:

A disease progression study was done in Marin County, CA to determine the affect of *P. ramorum* infection on stands (cohorts) of coast live oaks, California black oaks, and tanoaks. Once the pathogen became established in a cohort, the median survival time decreased from 29-yr to 2.7-yr for coast live oak and from 12.6-yr to 2.9-yr for tanoak. From 2000 to 2003, mortality in affected coast live oaks increased from 5.8% to 17.4%, in California black oaks from 3.8% to 9.4%, and in tanoaks from 8.3% to 22.2%. In the Big Sur ecoregion of California, the loss of 235,678 trees (12,650 m<sup>2</sup> of tree basal area) has been attributed to *P. ramorum*, with 63% of the mortality occurring in redwood-tanoak forests and 37% in mixed oak woodlands. In mixed-evergreen forests along the northern California coast, coast live oak mortality has increased ten-fold with the introduction of *P. ramorum*. Similar losses could be expected throughout the native range of tanoak in Oregon.

Tanoaks, oak species, rhododendrons, Pacific madrone, manzanita, Douglas fir, and evergreen huckleberry play important ecological roles in forests and woodlands as food and habitat for wildlife, and as soil cover in watersheds. The tanoaks, coast redwoods, Douglas fir, and oaks are significant riparian species, particularly in southwestern Oregon. Loss of these species could affect fish habitats such as those of endangered wild salmon populations and affect habitats of the endangered spotted owl and marbled murrelet. The evergreen huckleberry is an important food and habitat source for wildlife. The loss of oak species, tanoaks, and evergreen huckleberry would also reduce biodiversity in these ecosystems.

Within Oregon, the native range of coast redwood is limited to Curry County. Tanoaks have a scattered distribution within Coos, Curry, and Josephine counties, with one isolated population in Douglas County. Tanoaks are the primary host for *P. ramorum* in Oregon forests.

An active eradication and containment effort for *P. ramorum* is ongoing in Curry County. This effort has met with mixed success. Treatments used include cutting, piling, and burning all known infected plants and all healthy-appearing tanoaks, rhododendrons, and huckleberries within 100-m of the known infected plant(s). Tanoaks may also be treated with herbicides prior to cutting and burning, with the goal of this treatment to eliminate sprouting from the stumps. Broadcast burns are recommended on sites after pile burns are completed. Although non-host plants are left on site, landslides can be an issue on treated sites particularly when few non-hosts are present. Also, concerns have been expressed about the use of herbicides and of pile and broadcast burns.

### HOSTS

The following are listed as proven hosts for *Phytophthora ramorum*: Acer macrophyllum (big leaf maple), A. pseudoplatanus (planetree maple), Adiantum aleuticum (western maidenhair fern), Ad. jordanii (California maidenhair fern), Aesculus californica (California buckeye), Ae. hippocastanum (horse chestnut), Arbutus menziesii (Pacific madrone), Arctostaphylos manzanita (manzanita), Calluna vulgaris (Scotch heather), Camellia spp. (all species, cultivars, and hybrids), Castanea sativa (sweet chestnut), Fagus sylvatica (European beech), Frangula californica (= Rhamnus californica, California coffeeberry), F. purshiana (= Rhamnus purshiana, cascara), Fraxinus excelsior (European ash), Griselinia littoralis (griselinia), Hamamelis virginiana (witch hazel), Heteromeles arbutifolia (toyon), Kalmia spp. (mountain laurel, all species, hybrids, and cultivars), Lithocarpus densiflorus (tan oak), Lonicera hispidula (California honeysuckle), Laurus nobilis (bay laurel), Magnolia doltsopa (= Michelia doltsopa, michelia), Maianthemum racemosum (= Smilacina racemosa, false Solomon's seal), Parrotia persica (Persian ironwood), Pieris spp. (Andromeda, pieris – all species, hybrids, and cultivars), *Pseudotsuga menziesii* var. menziesii (Douglas-fir), Quercus agrifolia (coast live oak), Q. cerris (European turkey oak), Q. chrysolepsis (canyon live oak), Q. falcata (southern red oak), Q. ilex (Holm oak), Q. kelloggii (California black oak), Q. parvula var. shrevei (Shreve's oak), Rhododendron spp. (rhododendron including azalea – all species, hybrids, and cultivars), Rosa gymnocarpa (wood rose), Salix caprea (goat willow), Sequoia sempervirens (coast redwood), Syringa vulgaris (lilac), Taxus baccata (European yew), Trientalis latifolia (Western starflower), Umbellularia californica (California bay laurel, pepperwood, Oregon myrtle), Vaccinium ovatum (evergreen huckleberry),

*Viburnum* spp. (viburnum – all species, hybrids, and cultivars). Proven hosts are plants that have been found naturally infected with *P. ramorum* and on which traditional Koch's Postulates has been completed.

The pathogen has also been found naturally infecting the following plant species, although Koch's Postulates has yet to be completed: Abies concolor, A. grandis, A. magnifica, Acer circinatum, Ac. Davidii, Ac. Laevigatum, Arbutus unedo, Arctostaphylos Columbiana, Ar. uva-ursi, Ardisia japonica. Berberis diversifolia (= Mahonia aquifolium), Calycanthus occidentalis, Castanopsis orthacantha, Ceanothus thyrsiflorus, Cercis chinensis, Choisya ternate, Cinnamomum camphora, Clintonia andrewsiana, Cornus kousa, C. kousa x C. capitata, Corylopsis spicata, Corylus cornuta, Daphniphyllum glaucescens, Distylium myricoides, Drimys winteri, Dryopteris arguta, Eucalyptus haemastoma, Euonymous kiautschovicus, Fraxinus latifolia, Garrya elliptica, Gaultheria shallon, Hamamelis x intermedia (H. mollis x H. japonica), H. mollis, Ilex aquifolium, I. purpurea, Leucothoe axillaries, L. fontanesiana, Lithocarpus glaber, Loropetalum chinense, Magnolia cavalieri, M. denudata x salicifolia, M. ernestii (= Michelia wilsonii), M. figo, M. foveolata, M. grandiflora, M. kobus, M., liliiflora (= M guinguepeta), Magnolia x loebneri, M. maudiae (= Michelia maudiae), M. salicifolia (= Magnolia proctoriana), Magnolia x soulangeana, M. stellata, Magnolia x thompsoniana (M. tripetala and M. virginiana), Manglietia insignis, Nerium oleander, Nothofagus oblique, Osmanthus decorus (= Phyllyrea decora = P. vilmoriniana), Osmanthus delavayi, O. fragrans, O. heterophyllus, Osmorhiza berteroi, Parakmeria lotungensis, Physocarpus opulifolius, Pittosporum undulatum, Prunus Iusitanica, P. laurocerasus, Pyracantha koidzumii, Quercus acuta, Q. petraea, Q. rubra, Ribes laurifolium, Rosa (specific cultivars – Royal Bonica, Pink Meidiland, Pink Sevillana), R. rugosa, Rubus spectabilis, Schima wallichii, Taxus brevifolia, Taxus x media, Torreya californica, Toxicodendron diversilobum, Vancouveria planipetala, Vaccinium myrtilis, and V. vitis-idaea.

Recent reports of the pathogen infecting *Betula pendula, Larix kaempferi, Trachelospermum jasminoides,* and *Tsuga heterophylla* show the complete host range of this pathogen remains to be determined.

### **GEOGRAPHICAL DISTRIBUTION**

Canada: British Columbia (nursery stock)

United States: Alabama, California, Connecticut, Florida, Georgia, Louisiana, Maryland, Mississippi, New Jersey, North Carolina, Oregon, Pennsylvania, South Carolina, Tennessee, Virginia, Washington. Predominantly found in nursery stock, although *P. ramorum* has been detected in waterways in several states. Forest infestations exist in 14 California counties (Alameda, Contra Costa, Humboldt, Lake, Marin, Mendocino, Monterey, Napa, San Francisco, San Mateo, Santa Clara, Santa Cruz, Solano, and Sonoma) and in part of Curry County (419.6 km<sup>2</sup> quarantine area), Oregon.

Europe: Belgium Denmark, Finland, France, Germany, Ireland, Italy, Netherlands, Norway, Poland, Slovenia, Spain, Sweden, Switzerland, and United Kingdom.

### BIOLOGY

*Phytophthora ramorum* is genetically similar to *P. lateralis* (Port Orford cedar root rot), *P. hibernalis* (brown rot of citrus), and *P. foliorum* (leaf blight of azalea). It is a cool temperature organism, with optimum growth at 20°C. Numerous deciduous sporangia are produced *in vitro* and *in vivo*. Production of thick-walled chlamydospores is prolific *in vitro* and *in vivo*. These chlamydospores are desiccation-resistant, but have a limited tolerance for very high (>35°C) or very low (<-10°C) temperatures *in vitro*. Temperature tolerance is greater if *P. ramorum* is *in planta*. Moderate temperatures (20.5°C) and the presence of moisture favored infection of rhododendron plants by *P. ramorum*. At 20°C, dew periods are short as 1-hr resulted in a small amount of disease, although period of 24- to 48-hr resulted in the greatest amount of disease. Salt stress also contributes to the onset and severity of disease symptoms in rhododendron and viburnum. Symptom expression and mortality is typically delayed by 1- to 2-wk in non-stressed plants by comparison.

*Phytophthora ramorum* is a heterothallic organism. Although natural formation of sexual oospores has not been observed *in planta*, the formation of oospores has been induced by crossing North American isolates with European isolates. There are presently three recognized clonal lineages of the pathogen; NA1, NA2, and EU1. The lineages reportedly differ in their aggressiveness on certain susceptible plant species, with EU1 and NA2 generally recognized as more aggressive and NA1 as less. All three clonal lineages have been detected in nursery stock in the US, although only EU1 and NA1 have been detected from Oregon nursery stock. Forest isolates from Curry County all belong to the NA1 lineage.

Symptoms caused by the pathogen are host-dependent. On oaks and tanoak, the pathogen typically attacks phloem tissues, commonly extending into the xylem. Infected phloem tissue is discolored while infected xylem tissues typically exhibit a dark, often streaky discoloration. A dark brown to black line is usually evident at the margins of the infected areas in both xylem and phloem. The pathogen has been recovered from infected xylem tissue of beech and oaks 24- to

27-mo after debarking and phloem removal. On infected tanoak leaves, typically the petiole is darkly discolored; the pathogen can be recovered from this tissue. On some hosts, such as evergreen huckleberry and rhododendron, the pathogen may infect and cause dark purple to black stem cankers on small branches and twigs leading to dieback of branch leaders. On rhododendron, histological studies show the pathogen's growth extending from necrotic stem cankers into healthy-looking green tissue. Chlamydospores were formed only within the necrotic tissue. On other hosts, such as big leaf maple, the pathogen causes dark purple to black leaf spots. These host species are generally infected through natural openings in the leaves (e.g., stomata). These foliar hosts, in particular California bay laurel, are believed to play a key role in the spread of *P. ramorum* in California. In Oregon, tanoak appears to be the species most critical to natural spread. Root infection of rhododendron and lilac from infested potting media has been demonstrated in the laboratory, although this has rarely been observed in naturally infected plants. Infected roots may appear asymptomatic, even while sporangial cysts are being formed. Zoospores appear to be attracted to wounds and root primordia, and then colonize the cortex and vascular tissue of roots and stems.

### PEST SIGNIFICANCE

**Economic Impact:** *Phytophthora ramorum* infects and kills or injures at least 45 plant species (see Hosts). It has been recovered from an additional 82 "associated" host species. Recently, *P. ramorum* has been reported infecting four additional plant species not currently recognized by USDA APHIS as hosts or associated hosts. The severity of the infection is host dependent, with some hosts killed outright. Once infected, these hosts are more susceptible to secondary invaders such as wood-rotting fungi that often render the dead wood unusable. On other hosts (e.g., coast redwood), *P. ramorum* is known to kill only the immature seedlings while causing tip dieback on mature plants. On still other hosts (e.g., California bay laurel), the pathogen causes unsightly leaf spots that can serve as a source of inoculum for other, more susceptible host species.

Economic analyses for the impact of *P. ramorum* on the nursery and timber industries have been conducted for Oregon. Both analyses focused on the impact of potential natural spread from Curry County into the rest of Oregon. Thus, the disease management options (no treatment, maintaining the current treatment program, and attempting eradication) considered in the analyses were the same. To account for costs and benefits over time, both analyses covered a 20-yr time span. For the nursery industry, analysts determined if no treatments were done in Curry County, the subsequent spread of *P. ramorum* would result in approximately 777,000 km<sup>2</sup> under guarantine by year 2028. At the lowest estimated rate of disease spread (19 km/yr), the potential economic impact was estimated to be \$64.93 million over 20-yr. At the highest estimated rate of disease spread (75 km/yr), the impact would be \$652.30 million. Increases in guarantine inspection costs and nursery cultural control costs accounted for about 40% of the economic impact, while losses in market share/consumer confidence accounted for the rest. For the timber industry, analysts determined if no treatments were done in Curry County, the potential economic impact would range from \$21.3 million (3.7 km/yr spread rate) to \$1.24 billion (75 km/yr spread rate) over 20-yr. The final figure depends greatly upon the increased costs associated with harvesting trees within a guarantined area. Costs for monitoring for the pathogen were also considered important. In Washington, economic losses due to regulatory action at retail nurseries found infested with P. ramorum were estimated. Estimated economic loss was based on the number of plants destroyed and the retail value of those plants. From 2004 to 2005, 17,266 containerized nursery plants were destroyed at 32 nurseries, with the value of these plants estimated at \$423,043. Estimations have

not been made for economic losses associated with the enforcement of the USDA Confirmed Nursery Protocol within Oregon nurseries in which *P. ramorum* has been detected.

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Although not considered in this assessment, the presence of P. ramorum in Oregon also has the potential to impact the following industries primarily through trade restrictions: Christmas tree, specialty forest products, and blueberry. Oregon is the #1 producer of Christmas trees in the United States and ranks #3 nationally for blueberry production. In addition, plant species such as evergreen huckleberries are of particular importance to our Native American peoples. The wholesale price for one pound of huckleberries varied from \$2.98 to \$2.19 in 1995 and 1996. respectively, while the retail price varied from \$7.61 to \$12.83. Several host species are also popular landscape plants in Oregon and can increase a property's value by several thousand dollars. For example, in southwestern Oregon, mature black oak trees may increase a property's value by \$5,000 - 30,000 (survey of local realtors and property appraisers). The cost for removal of a single dead tree from a homeowner's property could run as high as \$3,000 (survey of tree care professionals). In California, researchers have determined that foliar moisture content in P. ramorum-infected trees is reduced by 4.5%, while the foliar moisture content in P. ramorum-killed trees is reduced by 70%; this puts these trees at high risk for crown ignition. Therefore, the presence of dead tanoaks in southwestern Oregon would also be expected to increase the costs of fire protection and prevention.

**Environmental Impact:** The potential environmental impact of *P. ramorum* on Oregon forests and other ecosystems is described below for the known host species most severely affected by this pathogen. In Oregon, oak savanna covers over 80,937 hectares in the Willamette Valley alone. The predominant species is the Garry oak (*Q. garryana*). California black oak, coastal live oak, canyon live oak, and interior live oak (*Q. wislizeni*) are also frequently encountered native species. California black oak and tan oak are important components of the mixed hardwood-conifer forests of southwestern Oregon.

The USDA Forest Service estimates the volume of California black oak in Oregon forests at 6 million cubic meters and the volume of tanoak at 14.5 million cubic meters (USDA Forest Service, PNW Research Station, Resource Monitoring and Assessment Program / FIA, Portland, OR, personal communication). Since 2001 there have been 97 hectares of primarily tanoak infected with P. ramorum in Curry County. An additional 1214 hectares have been treated for the disease as described below. Tanoak in particular is a critical under- and over-story species. Numerous oaks are also planted as ornamentals throughout the state, particularly along streets in urban landscapes. Oak savanna and oaks found in mixed forests yield important benefits, including water and watershed protection, grazing, wildlife food and habitat, fish habitat, recreation, and wood products. Several of the remaining host species may be found growing throughout Oregon with the largest populations concentrated near the Coast and Cascades Mountain Ranges. Hosts like wild rhododendron, Pacific madrone, manzanita, and evergreen huckleberry are valuable understory shrubs or minor tree species that provide water and watershed protection, wildlife food and habitat, and recreational benefits. They are the dominant under-story shrubs in several regions. Losing the wild Vaccinium species may have an even greater environmental impact than losing the other host species in terms of harm to wildlife (B. Newhouse, Native Plant Society of Oregon, 2001, pers. comm.). Heavy losses of oaks, tanoaks, and other P. ramorum host species could result in significant ecological effects, including changes in forest composition, loss of wildlife food and habitat, loss of fish habitat, increased soil erosion, and a significant increase in fuel loads in

populated urban-forest interfaces.

Control: In Curry County, Oregon, eradication and containment treatments have included herbicide treatment of tanoaks (to prevent sprouting), followed by cutting, piling, and burning of infected and nearby trees and shrubs. Sites are then broadcast burned when possible. Timing of these treatments after initial detection is considered critical to success of the treatment. In California, injection of trees with phosphite has been shown to suppress, but not eliminate infections. Spraying California bay laurel foliage with copper hydroxide limits pathogen growth. Treatment of nursery plants with certain fungicides can provide protection from infection for 14- to 28-d, although this protection was expressed primarily as reduced lesion growth. Fungicide treatment of already infected nursery plants did not significantly reduce lesion growth and only one fungicide tested (dimethomorph) consistently affected recovery of the pathogen from diseased rhododendron leaves. The pathogen may be eliminated from soil by fumigation with metam sodium, chloropicrin, Vapam, Basamid (if fully incorporated into the soil), and iodomethane. Solarization to >40°C for three consecutive days or to 35-40°C for 42-d effectively eliminated detectable propagules of *P. ramorum* from soil. Aerated steam treatments to  $\geq$ 50°C for 30-min eliminated P. ramorum propagules from plastic potting containers. Composting according to US EPA 40 CFR Part 503 requirements successfully eliminated P. ramorum from infected foliage and woody debris. However, researchers cautioned the pathogen could survive if reintroduced to mature compost. Heat treatment to 55°C for 2-wk can eliminate infections from bay laurel leaves meant for human consumption. However, this ruins the leaves as a spice. An alternative treatment that preserves the spiciness of the bay leaves requires the leaves be placed in a vacuum (0.133 kPa), gradually heated from 15 to 60°C over a 5-hr period, and then gradually cooled to 15°C for a total treatment time of 22-hr. Extracts from incense and western red cedar heartwood have exhibited strong antimicrobial activity against P. ramorum. Extracts from yellow-cedar, western juniper, and Port Orford cedar were moderately effective in comparison. The USDA APHIS PPQ has developed protocols to eliminate P. ramorum from wholesale and retail nurseries and from commercial landscape settings in which it has been detected. All three protocols contain the same basic elements: 1) delimitation survey to determine the extent of the infestation, 2) monitoring of soil substrate and water for the pathogen, 3) removal and destruction of infected plants, 4) treatment of soil substrate and/or water if *P. ramorum* is detected, 5) guarantine period to ensure all infected plants were found. Monitoring of the sites for another 2-yr after treatment is also required. Despite these efforts, so-called recurrent nurseries have been detected. Recurrent nurseries are those that have had multiple detections of P. ramorum over time, either because the pathogen survived the initial eradicative treatment or because it was re-introduced to the site.

#### DETECTION AND IDENTIFICATION

**Symptoms:** In general, symptoms on host plants fall into the following categories: stem cankers, foliar blight, and dieback. The symptoms caused on specific hosts are described in detail below.

Diseased tan oaks and oaks exhibit symptoms that include crown dieback or wilting, stem bark lesions or cankers, and tarry, reddish to dark brown exudations characteristic of a *Phytophthora* root and collar rot. The exudations do not always develop on tan oak, especially on smaller diameter branches. The cankers are typically found on the lower trunk, within 1- to 5 m of the soil line, sometimes extending all the way to the soil line. Branch cankers, especially on tan oak, also occur. These cankers have been observed as high as 18.3 m up in the canopy. Infected hosts

often appear to die suddenly, with crowns turning from apparently healthy green to yellow-green to brown within a few weeks. Tanoak leaves infected with *P. ramorum* develop necrotic petioles, with the necrosis sometimes extending the entire length of the leaf's mid vein.

The symptoms observed on *Rhododendron* species are typical of a *Phytophthora* foliar blight. Leaf spots are irregular and necrotic and may occur on the leaf tip or the petiole. Spots could be easily confused with leaf scorch, chemical damage, or other abiotic injuries. Leaf petioles may also turn a black color as infection spreads. Branch cankers are typically black, but not necessarily sunken. If left unchecked, the stem canker can spread into the main stem of the plant eventually killing it.

*Phytophthora ramorum* causes a dieback symptom on infected evergreen huckleberries similar to the symptoms produced on rhododendrons. Stem infections can kill the canes all the way back to the soil line. This pathogen also causes a rarely-seen leaf spot symptom on evergreen huckleberry. Leaf margins turn a dark purple to black color with the discoloration eventually spreading to cover the whole leaf. In general, however, the leaves fall off shortly after becoming infected.

**Morphology:** Colony characteristics and gametangial morphology have been described by Werres et al. Morphologically, *P. ramorum* is characterized by the abundant production of chlamydospores ( $20-91 \mu m$ ) and elongate, ellipsoid, semi-papillate, deciduous sporangia with a short pedicel. *P. ramorum* is heterothallic. To date, oogonia with amphigynous antheridia and plerotic oospores have only been observed *in vitro*.

Testing Methods for Identification: The USDA APHIS PPQ CPHST has established a national diagnostic protocol for *P. ramorum* in the United States. Plant samples may be pre-screened with an approved DAS-ELISA test kit available commercially from AgDia Inc. (Elkhart, IN). Samples that test ELISA-negative are considered free of *P.ramorum*. Samples that test ELISA-positive must be tested with a molecular diagnostic method approved by USDA APHIS PPQ CPHST. These samples may also be isolated from onto an approved selective medium per federal protocol. Samples from which isolates are obtained that appear to be *P. ramorum* morphologically must be sent to the USDA APHIS PPQ MDL for official confirmation of the culture identification. A sample that tests ELISA-positive and culture-negative must be tested with a molecular diagnostic method approved by USDA APHIS PPQ CPHST. Presently, USDA APHIS PPQ CPHST recognizes three molecular methods as valid tests for *P. ramorum*: a quantitative multiplex real time PCR assay targeting the ITS region of the nrDNA, a quantitative multiplex real time PCR assay targeting the elicitin genomic loci, and conventional multiplex and nested PCR assays that target the ITS region of the nrDNA. An ELISA-positive sample that tests positive for *P. ramorum* using the ITS and elicitin real time PCR assays or the conventional multiplex and nested PCR assays must be submitted to the USDA APHIS PPQ MDL for official confirmation of the test result. Samples officially confirmed by USDA APHIS PPQ MDL are considered positive and trigger a regulatory response.

Additional molecular testing methods have been developed, but are not considered approved by USDA APHIS PPQ CPHST. However, the use of multiple diagnostic methods, such as culturing and molecular diagnosis or two molecular assays, to confirm the presence of *P. ramorum* has been verified.

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The pathogen can be isolated from bark lesions, leaf spots, or stem cankers by plating onto a selective medium. *P. ramorum* has also been isolated from soil substrate, potting media, rain-splash, irrigation water, and stream water by using a baiting technique. Typically, rhododendron and/or camellia leaves are recommended as baits, although other susceptible plants' leaves and pears may also be used successfully. Stream monitoring has been an effective tool for early detection of *P. ramorum* in Curry County, OR. When the pathogen was detected in stream baits, intensive ground surveys of the surrounding area located an infected tanoak or other host plant an average of 306 m upstream from the bait station.

### MEANS OF MOVEMENT AND DISPERSAL

*Phytophthora ramorum* has been isolated from infected nursery stock and other plant materials, from soil, from potting media, from rain-splash, and from stream water. Long-distance spread of this pathogen on infected nursery stock has been demonstrated in Europe, the United States, and in Canada. In mainland Europe, the pathogen has been considered predominantly a nursery problem, although the pathogen has reportedly spread from infected nursery plants into urban landscapes and gardens. In the United Kingdom, *P. ramorum* was recently reported causing extensive mortality in Japanese larch plantations. Most forest infestations in this country are reportedly associated with infected *R. ponticum*.

In U.S. nurseries, *P. ramorum* has been detected in plants moving through the trade, in potting media associated with infected plants, in leaf litter, in soil substrate below infected plants, and in irrigation water. Irrigating with water contaminated by *P. ramorum* can result in plant infections, whether the source is a contaminated stream or nursery holding pond. Sprinkler irrigation seems to promote foliar infections when the inoculum source is contaminated water or potting media. When *P. ramorum* is present in the soil substrate, it is usually found in the organic layer or at a depth of 0 - 5 cm; it is not found below 10 cm. Chlamydospores of the pathogen may persist for at least 12-mo in potting media and soil substrate. For plants like camellia, once their foliage becomes infected the leaves readily dehisce. This leaf litter then serves as an inoculum source of other foliar infections and for contamination of the soil substrate and potting media.

In Oregon, epidemiology research in Curry County has shown that half of all new infection centers are located within 122 m of a previous infection and 79% within 300 m. Dispersal up to 4 km has been recorded, although only occasionally. These dispersal events are believed to be associated with local rain splash and long-distance turbulent air dispersal of inoculum. By comparison, in California epidemiological studies suggest dispersal 1.5 - 10 km, with researchers assuming this spread is due to wind events. The pathogen can be recovered from soil and leaf litter beneath known infected trees, even years after the site has been treated (E. Goheen, USDA Forest Service, unpublished). P. ramorum has been shown to move from green leaf litter in the soil into healthy, above-ground bay laurel leaves. In rainwater, inoculum was shown to move 5-10 m from the inoculum source by splashing. In the natural environment, researchers have correlated periods of peak spore production with rain or high moisture events. In Oregon, it has been demonstrated that spores can be produced year-round in Curry County, including from underneath infected trees with dead foliage (E. Hansen, Oregon State University, unpublished). In California, P. ramorum has been baited out of soil collected from hiking trails and from hikers' shoes. Phytophthora ramorum has also been recovered from stream water flowing out of infested areas in California and Oregon, although the role of stream water in disease spread is still under investigation.

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