



## CALIFORNIA OAK MORTALITY TASK FORCE REPORT NOVEMBER 2007

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### MONITORING

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***Phytophthora ramorum* inoculum was baited from Norton Creek (a small coastal stream in northern Humboldt County in the town of McKinleyville) early in the summer of 2006, with a repeat detection in the spring of 2007. The recovered isolates were genotyped in October 2007, which revealed that two lineages had been detected: the Northern American (NA1) and European (EU1). This is the first find in North America of the EU1 lineage in a wildland environment. Efforts are being made to determine the spore source location, including exhaustive streamside surveys, but none has yet revealed the source. A small retail nursery in downtown McKinleyville has been found to have *P. ramorum*-positive plants, including three plants confirmed with the EU1 strain, but the nursery is located in a different watershed than the infested stream. No definitive linkages between the nursery and stream have been established. In response to the recent findings, the nursery has implemented best management practices and is cooperating with regulatory agencies on conducting a treatment.**

The riparian areas along this coastal stream are dominated by common willows, alders, and some Sitka spruce, all of which are not known to be hosts to *P. ramorum*. Typical wildland hosts such as California bay laurel and tanoak are absent from the stream corridor. This stream positive is 47 miles north of the northern most known wildland infestation in California. For more information, contact Jeff Dolf, Humboldt County Department County, at [JDolf@co.humboldt.ca.us](mailto:JDolf@co.humboldt.ca.us) or Amber Morris, California Department of Food and Agriculture, at [amorris@cdfa.ca.gov](mailto:amorris@cdfa.ca.gov).

### REGULATIONS

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**The Oregon Department of Agriculture (ODA) has published a notice to amend the State quarantine for *Phytophthora ramorum* (OAR 603-052-1230) and the regulated area for nursery stock (603-052-1250). The proposed amendments would harmonize the state's *P. ramorum* rules with current federal regulations and protocols. The area under quarantine for *P. ramorum* in Curry County would increase to 166 square miles (for a map of the expanded quarantine area, go to the [Oregon Map](#), and include five newly detected sites outside of the existing quarantine area (603-052-1230). The complete announcement can be found in the 11/1/07 issue of the Oregon Bulletin ([http://arcweb.sos.state.or.us/rules/1107\\_Bulletin/1107\\_rulemaking\\_bulletin.html](http://arcweb.sos.state.or.us/rules/1107_Bulletin/1107_rulemaking_bulletin.html)). ODA will accept written comments on the proposed amendments until 11/26/07. Comments may be submitted to: Sue Gooch; ODA; 635 Capitol St. NE; Salem, OR 97301-2532. Please reference *P. ramorum* in your comments.**

### RESEARCH

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**Dart, N.L., Chastagner, G.A., Rugarber, E.F., and Riley, K.L. 2007. Recovery frequency of *Phytophthora ramorum* and other *Phytophthora* spp. in the soil profile of ornamental retail nurseries. Plant Disease 91:1419-1422. DOI: 10.1094/PDIS-91-11-1419**



**Abstract:** We tested the hypothesis that inoculum of the aboveground exotic plant pathogen *Phytophthora ramorum* would be limited to the organic layer (top layer of plant debris) of soils at infested retail nurseries located outside of the area where the pathogen has become established in the landscape. To test this hypothesis and compare inoculum levels of *P. ramorum* with levels of other *Phytophthora* spp. in the soil profile, soil cores were collected and sampled from three Washington State retail nurseries at which the soil had previously tested positive for *P. ramorum*. *Phytophthora* was isolated from soil using rhododendron leaves as bait, and pure cultures were obtained and stored on V8 juice agar. Isolates were identified to species using a combination of DNA sequencing of the internal transcribed spacer (ITS) region of rDNA, real-time polymerase chain reaction (PCR) diagnostic testing, and culture morphology. Recovery frequencies were tabulated and compared by species at the organic layer, 0 to 5 cm, 5 to 10 cm, and 10 to 15 cm depth classes. The three most common *Phytophthora* spp. recovered from the soil cores were *P. citricola* (32%), *P. drechsleri* (32%), and *P. ramorum* (27%). *P. citricola* and *P. drechsleri* were more evenly distributed throughout the soil profile, whereas *P. ramorum* was primarily recovered from the organic and 0 to 5 cm depth class (86% of recoveries). *P. ramorum* was not detected below 10 cm.

#### **RELATED RESEARCH**

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##### **A New Species of *Phytophthora* Associated with Dying Pine Needles in Chile:**

During the course of the past three years, a new needle disease has appeared in *Pinus radiata* plantations, chiefly in the Arauco province of Chile. The disease, locally referred to as Daño Foliar del Pino (DFP), is typified by the relatively rapid death of needles and subsequent defoliation of trees. Infections usually begin to appear in late autumn and coincide with the onset of rain. Infected needles typically display distinct resinous bands on their lamina's. Where infections reach the needle bases, copious amounts of resin exude from the points of attachment with the stems and dead needles falling from the trees. New needle growth in the following season is typically not affected and trees appear to recover unless a new season of infection occurs the following year. Newly planted seedlings and naturally regenerated plants die in the first year of growth. Isolations from infected *P. radiata* needles using selective media have consistently yielded a *Phytophthora* sp., the identification of which has been confirmed based on morphological characteristics including non-papillate and caducous sporangia. DNA sequence analyses for the ITS region of the rDNA and COX II region of isolates have confirmed the identification of the fungus from infected *P. radiata* needles as a *Phytophthora* sp. Furthermore, they have provided robust evidence to suggest that it represents a new species, *Phytophthora pinifolia* nom. prov. A team of scientists linked to FABI (<http://www.fabinet.up.ac.za>) are currently considering the origin of the pathogen, its possible role as the cause of DFP, and improved management options for the disease. Symptomatic pictures can be found at <http://www.fabinet.up.ac.za/tpcp/pinifolia>.

**Mrazkova, M.; Cerny, K.; Gabrielova, S.; and Tomsovsky, M. Nov. 2007. First Report of Leaf Spot, Shoot Blight, and Stem and Collar Canker of *Rhododendron* spp.**



Caused by *Phytophthora citricola* in the Czech Republic. Disease Notes Vol. 91, No. 11: 1515. DOI: 10.1094/PDIS-91-11-1515B.

**Abstract:** During the summer and autumn of 2006, a disease of rhododendron plants (*Ericaceae*) was found in nurseries and public gardens in several areas of the Czech Republic. Leaves of damaged plants showed dark brown-to-black lesions extending along the mid-rib and commonly spreading to petioles and shoots. The infected shoots turned black and died. The cankers on branches, stems, and collars were characterized by reddish, brownish, or blackish discoloration. The disease was identified on *Rhododendron catawbiense*, *R. repens*, and other *Rhododendron* spp. After plating pieces of symptomatic tissue on PARPNH medium (2), several isolates of a homothallic *Phytophthora* sp. were acquired. Ten representative isolates of the pathogen were cultivated on V8A plates and examined for cultural and morphological characteristics. Colonies had a stellate pattern of growth with sparse aerial mycelium at 20°C; optimum temperature for growth was 25 - 28°C, minimum was 4°C, and maximum was 33°C. Radial growth was 14 mm per day at 20°C on V8A. The isolates produced terminal, spherical, smooth-walled oogonia, which were 19 - 37 µm in diameter. Oospores were plerotic (17 to 32 µm) with walls 2 - 4 µm thick; antheridia were paragynous. Single, terminal, noncaducous, semipapillate sporangia were formed on simple (occasionally sympodial) sporangiophores in nonsterile soil filtrate. The sporangia (28 to 61 × 24 to 35 µm, L:B ratio 1.5) were mostly obpyriform, rarely obovoid, or ovoid-ellipsoid. Morphological and cultural characters resembled those described for *Phytophthora citricola* Sawada. The ITS sequences of the rDNA of the two representative isolates (GenBank Accession Nos. EF194772 and EF194773) showed 100% homology to *P. citricola* sequences obtained from GenBank, thus the identity was confirmed as *P. citricola*. Both specimens were deposited in CCF (Culture Collection of Fungi, Charles University, Prague, Czech Republic). To confirm the pathogenicity of isolates, Koch's postulates were tested using 40 3-year-old potted rhododendron (*R. catawbiense* and *R. repens*) plants and the two *P. citricola* strains deposited in CCF. Surfaces of attached healthy leaves were disinfected with 95% ethanol and gently abraded with a sterile scalpel near the mid-rib. Agar plugs from the margin of a 5-day-old colony grown on carrot agar were placed on leaf surfaces and also inserted under flaps of stem tissues made with a sterile scalpel. The leaves and stems were then sealed with Parafilm. Control plants were treated in the same manner with sterile agar plugs. All plants were watered with deionized water, covered with a plastic bag, and maintained in a greenhouse at 21°C for 6 weeks. All inoculated plants exhibited necrotic lesions on leaves and stems around the points of inoculation after 4 days, whereas the control plants remained healthy. The pathogen was consistently reisolated from symptomatic plants. *P. citricola* is well known as a pathogen of rhododendron, but to our knowledge, this is the first report of *P. citricola* on *Rhododendron* sp. in the Czech Republic. *P. citricola* has been found at five different locations and in the most frequently isolated *Phytophthora* spp. from rhododendron in the Czech Republic.



**Varela, C. Pintos; Vázquez, J.P. Mansilla; Casal, O. Agúin; and Martínez, C. Rial.** November 2007. First Report of *Phytophthora pseudosyringae* on Chestnut Nursery Stock in Spain. Disease Notes Vol. 91, No. 11: 1517. DOI: 10.1094/PDIS-91-11-1517A.

**Abstract:** *Phytophthora pseudosyringae* causes stem necrosis and collar rot of deciduous tree species (*Quercus* spp., *Fagus sylvatica*, and *Alnus glutinosa*) in several European countries. In November 2006, we received diseased *Castanea sativa* seedlings from a nursery in Galicia (northwest Spain). These plants had tongue-shaped necroses of the inner bark and cambium. Reddish, sunken lesions occurred on the surface of the bark, either in the stem base or higher on the stem. Tissue from the leading edge of the lesions was transferred to a selective V8 agar medium and incubated for 7 days at 20°C in the dark. A *Phytophthora* sp. was isolated, transferred to cornmeal agar (CMA) and V8 agar, and incubated in the dark. Colonies were appressed with stellate to rosaceous growth patterns on CMA and stellate, limited aerial mycelium on V8 agar. Growth on V8 occurred from 2 to 25°C with an optimum at 20°C and a radial growth rate of 4.5 mm per day at 20°C. Chains of inflated spherical to deltoid hyphal swellings with radiating hyphae were abundantly produced in water. Chlamydospores were not observed on agar media. The deciduous, sympodial, semipapillate, rarely bipapillate sporangia with pedicels had a length/breadth average ratio of 1.55. Oogonia, antheridia, and oospores were produced within a single culture. Oogonia were spherical and smooth walled, antheridia were predominantly paragingous, but some were amphigynous, and oospores were plerotic that turned golden yellow with age. Internal transcribed spacer (ITS)-rDNA and mitochondrial DNA (mtDNA) regions were amplified by nested-PCR and sequenced with DNA extracted from mycelium. The amplicon sizes obtained were similar to those reported for *P. pseudosyringae*. DNA sequences showed 99 to 100% homology with those previously identified as *P. pseudosyringae* and deposited in GenBank. Pathogenicity of the isolate was confirmed by inoculating 10 *C. sativa* seedlings, as well as three detached leaves from each of another 10 young plants growing in containers. For the seedlings, one shallow cut was made into the bark on the main stem. A colonized agar plug was inserted beneath the flap that was sealed with Parafilm. Unwounded and wounded detached leaves of *C. sativa* were dipped into a zoospore aqueous suspension ( $1 \times 10^5$  zoospores ml<sup>-1</sup>) for 10 s., seedlings and leaves were incubated at 20°C and 95% humidity for 60 and 7 days, respectively. After 7 days, foliar lesions that developed exceeded 25 mm, and the pathogen was consistently reisolated. Leaves inoculated with sterile water did not develop symptoms. On inoculated seedlings, the external surface of the bark was reddish and sunken. Stem lesions progressed bidirectionally from the wound. *P. pseudosyringae* was recovered from inoculated seedlings but not from controls. On the basis of its unique combination of morphological and physiological characters, pathogenicity, and ITS and mtDNA sequences, the *Phytophthora* isolated from chestnut was identified as *P. pseudosyringae*. To our knowledge, this is the first report of *P. pseudosyringae* on *C. sativa* in Spain.



## COUNTY UPDATE

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**In Humboldt County, 2007 spore trapping and soil sampling in known infested areas** detected very little *P. ramorum* inoculum. Nor was the same suite of foliar symptoms (blackened petioles and shepherd's crooking of tanoak, and large-scale branch-tip dieback in huckleberry, manzanita, and blueblossom) apparent after the dry winter of 2006-2007 as was apparent after the previous, very wet winter. Existing *P. ramorum* infestations, however, continued to fill in with new tanoak mortality. This mortality is speculated to have mostly originated from infections initiated in 2005-2006.

Although dramatic expansions of the infested area were not evident from the ground, the USDA Forest Service, Pacific Southwest Region, Forest Health Protection aerial survey mapped several new large polygons of mortality along the Salmon Creek watershed north of the Redway area, roughly parallel to the South Fork Eel River. Additionally, the aerial survey mapped a polygon of tanoak mortality at Eel Rock on the main stem Eel River to the east of the known infested area, which was subsequently confirmed to be caused by *P. ramorum*. The survey also identified two polygons of mortality in the East Branch South Fork Eel to the southeast of Garberville, a watershed in which *P. ramorum* inoculum had previously been detected but where no infected trees had so far been identified. This infestation is approximately three miles from previously known infested areas near the Garberville airport. For an updated map of the infested area, go to [Southern Humboldt Tanoak Mortality 2007 Map](#).

In March, California State Parks and UCCE Humboldt-Del Norte supervised removal of all California bay laurel trees along a 3.5-mile stretch between the Avenue of the Giants and the South Fork Eel and between Myers Flat and Burlington. UCCE periodically samples bay trees along the Avenue of the Giants and the South Fork Eel to determine *P. ramorum* movement north along the river corridor. The removal included both known infected and unsampled trees. This stretch represented the farthest known extent of the *P. ramorum* infestation to the north in the county and has not yet experienced any *P. ramorum*-caused tanoak mortality. Periodic sampling along the Avenue will continue, along with similar roadside sampling currently in progress on all sides of the county's known infestation.

## NURSERIES

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**Mock Nursery Update:** After a year of negotiations to secure a mock nursery site for *P. ramorum* research at an Alameda County facility, it appears that the site is likely no longer an option. While the County decision is not yet final, bids are being accepted from others looking to develop the site. Therefore, alternate locations for the mock nursery are being sought after for consideration. Location criteria include: five to 10 acres in a quarantined county, a secure site with accessible power and water, no oak forests or nurseries in close proximity, preferably no native hosts such as tanoak and bay nearby, and the ability to collect drainage from the projects. Researchers will be traveling great distances, so proximity to airports and lodging would be optimal. For more information on the mock nursery program or to suggest a possible suitable location, please call Kathy Kosta at (916) 654-1440.



### **CORRECTION**

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**The California Department of Food and Agriculture (CDFA) has submitted the following amendment/clarification to the August COMTF Newsletter regarding *P. ramorum* nursery inspections.**

In alignment with the National Plant Board High Risk Proposal, nursery inspectors are now conducting two inspections per year on “medium-risk” plants and three inspections per year on “high-risk” plants. High-risk (HR) plants (*Camellia* and *Rhododendron*) have been defined as such because of the higher frequency with which these genera are found *P. ramorum* positive compared to other genera. Medium-risk plants include *Viburnum*, *Pieris*, and *Kalmia*. *Viburnum* and *Pieris* are the next most frequently found genera after the HR plants. *Kalmia* is considered medium risk because it is a reasonably good *P. ramorum* host and it is widespread in eastern US forests. With an ever-expanding host list and decreasing resources, inspections are increasingly focused on those plants at greatest risk for *P. ramorum*.

### **RESOURCES**

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**Presentations and posters from the 4<sup>th</sup> IUFRO *Phytophthoras* in Forests and Natural Ecosystems Conference at the Asilomar in Pacific Grove, CA are now available at <http://nature.berkeley.edu/IUFRO2007/phytophthora/programme.html>.**

### **EDUCATION**

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***P. ramorum* nursery training sessions are being offered throughout California this fall for industry professionals.** The sessions are designed to provide updates on the revised USDA *P. ramorum* regulations as well as new scientific and management information. Sessions are free of charge, though online pre-registration is required. For more information on registration, dates, and locations, see the Calendar of Events below.

A comprehensive cd of the nursery training sessions has been produced, and will be posted online at the COMTF website ([www.suddenoakdeath.org](http://www.suddenoakdeath.org)) in the near future. The training sessions and the cd are sponsored by the California Oak Mortality Task Force, the California Department of Food and Agriculture, the California Association of Nurseries and Garden Centers, the USDA Animal and Plant Health Inspection Service (APHIS), and the USDA FS Pacific Southwest Research Station.

### **PERSONNEL**

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**A Sudden Oak Death Outreach Assistant half-time position is available with the UC Cooperative Extension (UCCE) office in Marin County.** Under the supervision of the California Oak Mortality Task Force Sudden Oak Death Outreach Coordinator and the UCCE Marin Environmental Horticultural Advisor, the assistant will help with the preparation and delivery of the California Oak Mortality Task Force’s education and outreach activities. Specific duties include, but are not limited to, assisting in the coordination of professional and community-based training sessions and conferences, public speaking, and designing and developing educational outreach materials. The



salary range for this position is \$15.60 - \$25.68 per hour, and will be set based on experience. Interested applicants must submit their resume to Janice Alexander ([jalexander@ucdavis.edu](mailto:jalexander@ucdavis.edu)) via email by November 9, 2007. For more information on the position, contact UCCE Marin at (415) 499-4204 or email Janice Alexander at [jalexander@ucdavis.edu](mailto:jalexander@ucdavis.edu).

#### CALENDAR OF EVENTS

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- 11/06 – Free Ventura County *P. ramorum* Nursery Industry Training for Professionals;** Government Center, Ventura; 12:00 - 5:00 p.m.; Pre-registration is required and can be done online at [Nursery Online Registration](#). For more information, contact Katie Palmieri at (916) 435-3230 or [palmieri@nature.berkeley.edu](mailto:palmieri@nature.berkeley.edu).
- 11/13 – Free San Diego County *P. ramorum* Nursery Industry Training for Professionals;** Escondido Farm Bureau, Escondido; 12:00 - 5:00 p.m.; Pre-registration is required and can be done online at [Nursery Online Registration](#). For more information, contact Katie Palmieri at (916) 435-3230 or [palmieri@nature.berkeley.edu](mailto:palmieri@nature.berkeley.edu).
- 11/14 – Free Orange County *P. ramorum* Nursery Industry Training for Professionals;** South Coast Research and Extension Center, Irvine; 7:30 a.m. – 12:30 p.m.; Pre-registration is required and can be done online at [Nursery Online Registration](#). For more information, contact Katie Palmieri at (916) 435-3230 or [palmieri@nature.berkeley.edu](mailto:palmieri@nature.berkeley.edu).
- 11/15 – Free Sacramento County *P. ramorum* Nursery Industry Training for Professionals;** Ag Extension Auditorium, Sacramento; 12:00 - 5:00 p.m.; Pre-registration is required and can be done online at [Nursery Online Registration](#). For more information, contact Katie Palmieri at (916) 435-3230 or [palmieri@nature.berkeley.edu](mailto:palmieri@nature.berkeley.edu).
- 11/15 – Green Valley Sudden Oak Death Community Informational Meeting;** Falls School Multipurpose Room; 1634 Rockville Rd.; Fairfield  
7:00 – 8:00 p.m.; For more information, contact Mimi Fleige at [mimi@avita.us](mailto:mimi@avita.us) or (707) 864-2022.