CALIFORNIA OAK MORTALITY TASK FORCE REPORT
JUNE 2007

MONITORING

Oregon identified a new *P. ramorum* outbreak that is approximately 1.5 miles north of the quarantine area. The federal land, administered by the USDI Bureau of Land Management, is forested with large old-growth Douglas-fir in the overstory and predominantly tanoak, Oregon myrtlewood, bigleaf maple, and evergreen huckleberry in the mid- and understory layers.

The site was detected in early March during ground-based surveys that were being conducted as a follow-up to a *P. ramorum*-positive water baiting sample. A one-inch diameter live tanoak with a bleeding canker located approximately 500 feet upstream from the water find was determined to be the source of the stream water positive. An eradication treatment area boundary of 300 feet was immediately established around the known affected tanoak, totaling approximately 4.9 acres. A quarantine area that extends approximately one-half mile in all directions from the affected tree was also established. As of late April, all tanoak and evergreen huckleberry within the treatment boundary had been cut and piled. Piles will be burned as soon as conditions are appropriate. Myrtlewood plants left on site, soil, and resprouting vegetation will be carefully monitored. For more information, contact Ellen Goheen at egoheen@fs.fed.us.

The Mississippi Forestry Commission, MS Department of Agriculture, USDA Forest Service, and USDA Animal and Plant Health Inspection Service (APHIS) have developed a follow-up survey plan in response to the water baiting confirmation made last month from a ditch draining a *P. ramorum*-positive nursery. The plan includes immediately implementing stream baiting, soil baiting around the edge of the waterway where the positives were obtained, and vegetation surveys. As environmental conditions are not ideal this time of year for *P. ramorum* recovery, these same survey activities will be repeated in the fall when weather conditions are more conducive to pathogen detection. For more information, contact Steve Oak at soak@fs.fed.us.

REGULATIONS

APHIS has updated the "Official Regulatory Protocol for Wholesale and Production Nurseries Containing Plants Infected with *Phytophthora ramorum.*" Effective immediately, the revised Confirmed Nursery Protocol (CNP), Version 8.0 is to be used by any nursery found positive for *P. ramorum*. This revised protocol differs most markedly in that it contains additional measures such as biosecurity procedures for nurseries which have been confirmed positive within a year of being found previously positive and then released from emergency measures. To access the new CNP, go to http://www.aphis.usda.gov/plant_health/plant_pest_info/pram/downloads/pdf_files/CNPv8.pdf

NURSERIES

The first US findings of *P. ramorum*-positive Loropetalum have been found at a Sacramento County, CA nursery that has previously been identified with the pathogen.

The California Department of Food and Agriculture (CDFA) confirmed the sample through culturing. Symptoms, unlike other foliar hosts, can include large and small pin-prick size lesions on the underside of leaves, surrounded by red rings.

RESEARCH

Proposed terminology for *P. ramorum* lineages: In the Ivors *et al*. publication, "Microsatellite markers identify three lineages of *Phytophthora ramorum* in US nurseries, yet single lineages in US forest and European nursery populations," (Molecular Ecology [2006] 15, 1493–1505) numerous *P. ramorum* genotypes were identified, representing three total lineages. In light of these results, *P. ramorum* population biologists agreed that a consistent way to identify the lineages would be useful, and therefore labeled each lineage based on the continent where it was first found. Hence, the common European lineage becomes the 'EU1 lineage,' the common US/North American wildland lineage is the 'NA1 lineage,' and the recently identified new, yet rare, US/North American lineage would be known as the 'NA2 lineage.' (To date all the NA2 isolates have been detected in nurseries.) General adoption of this terminology in *P. ramorum* publications will provide more consistency for future research involving the population genetics of *P. ramorum.* For more information, contact Kelly Ivors at Kelly_Ivors@ncsu.edu.

Bilodeau, G.J.; Lévesque, C.A.; de Cock, A.W.A.M.; Duchaine, C.; Brière, S.; Uribe, P.; Martin, F.N.; and Hamelin, R.C. 2007. Molecular detection of *Phytophthora ramorum* by real-time polymerase chain reaction using TaqMan, SYBR Green, and molecular beacons. Phytopathology 97:632-642.

Abstract: Sudden oak death, caused by *Phytophthora ramorum*, is a severe disease that affects many species of trees and shrubs. This pathogen is spreading rapidly and quarantine measures are currently in place to prevent dissemination to areas that were previously free of the pathogen. Molecular assays that rapidly detect and identify *P. ramorum* frequently fail to reliably distinguish between *P. ramorum* and closely related species. To overcome this problem and to provide additional assays to increase confidence, internal transcribed spacer (ITS), β-tubulin, and elicitin gene regions were sequenced and searched for polymorphisms in a collection of *Phytophthora* spp. Three different reporter technologies were compared: molecular beacons, TaqMan, and SYBR Green. The assays differentiated *P. ramorum* from the 65 species of *Phytophthora* tested. The assays developed were also used with DNA extracts from 48 infected and uninfected plant samples. All environmental samples from which *P. ramorum* was isolated by PARP-V8 were detected using all three real-time PCR assays. However, 24% of the samples yielded positive real-time PCR assays but no *P. ramorum* cultures, but sequence analysis of the *cox*I and II spacer region confirmed the presence of the pathogen in most samples. The assays based on detection of the ITS and elicitin regions using TaqMan tended to have lower cycle threshold values than those using β-tubulin and seemed to be more sensitive.

Dart, N.L. and Chastagner, G.A. 2007. Estimated economic losses associated with the destruction of plants due to *Phytophthora ramorum* quarantine efforts in Washington State. Online. Plant Health Progress doi:10.1094/PHP-2007-0508-02-RS.

Abstract: The number and retail value of plants destroyed in Washington State nurseries due to *Phytophthora ramorum* quarantine efforts was estimated using Emergency Action Notification forms (EANs) issued by the United States Department of Agriculture Animal and Plant Health Inspection Service between 2004 and 2005. Data collected from EANs indicate that during this period 17,266 containerized nursery plants were destroyed at 32 nurseries, worth an estimated $423,043. The mean loss per nursery was estimated at $11,188 in 2004, $11,798 in 2005, and at $13,220 per nursery over the 2-year period.

Kluza, D.A.; Vieglais, D.A.; Andreasen, J.K.; and Peterson, A.T. 2007. Sudden oak death: geographic risk estimates and predictions of origins. Plant Pathology. DOI: 10.1111/j.1365-3059.2007.01602.x.

Summary: Ecological niche modeling techniques were applied to address the questions of the origins and potential geographic extent of sudden oak death, caused by the pathogen *Phytophthora ramorum*. Based on an ecological niche model derived from the phytopathogen's California distribution and distributions of potential host species, it was determined that the disease has high potential to colonize the southeastern United States, and that its likely source area is eastern Asia.

Lane, C.R.; Hobden, E.; Walker, L.; Barton, V.C.; Inman, A.J.; Hughes, K.J.D.; Swan, H.; Colyer, A.; and Barker, I. 2007. Evaluation of a rapid diagnostic field test kit for identification of *Phytophthora* species, including *P. ramorum* and *P. kernoviae* at the point of inspection. Plant Pathology. DOI: 10.1111/j.1365-3059.2007.01615.x.

Abstract: Plant health regulations to prevent the introduction and spread of *Phytophthora ramorum* and *P. kernoviae* require rapid, cost effective diagnostic methods for screening large numbers of plant samples at the time of inspection. Current on-site techniques require expensive equipment, considerable expertise and are not suited for plant health inspectors. Therefore, an extensive evaluation of a commercially available lateral flow device (LFD) for *Phytophthora* species was performed involving four separate trials and 634 samples. The assay proved simple to use, provided results in a few minutes and on every occasion a control line reacted positively confirming the validity of the test. LFD results were compared with those from testing a parallel sample, using laboratory methods (isolation and real-time PCR). The diagnostic sensitivity of the LFD (87·6%) compared favourably with the standard laboratory methods although the diagnostic specificity was not as stringent (82·9%). There were a small number (n=28) of false negatives, but for statutory purposes where all positive samples must be identified to species level by laboratory testing, overall efficiency was 95·6% as compared with visual assessment of symptoms of between 20-30% for *P. ramorum* and *P. kernoviae*. This work demonstrates the value of the LFD for diagnosing *Phytophthora* species at the time

of inspection and as a useful primary screen for selecting samples for laboratory testing to determine the species identification.

Swiecki, T.J. and Bernhardt, E.A. 2007. Influence of local California bay distribution on the risk of *Phytophthora ramorum* canker (Sudden Oak Death) in coast live oak. Available online at http://www.phytosphere.com/publications/influence_bay_dist_SOD.htm.

This study addressed the question of whether there is a "safe" distance between California bay laurel and coast live oak beyond which the risk of disease is acceptably low. Bay cover and other factors were quantitatively evaluated in the areas surrounding 247 coast live oaks located in mixed hardwood forest long-term research plots in where *P. ramorum* has been prevalent since 2000.

Study findings indicate that the risk of developing Sudden Oak Death in coast live oak appears to be minimal at bay foliage-oak trunk clearances of 10 m or more. The risk of *P. ramorum* infection and the severity of disease can be greatly reduced, but not completely eliminated by (1) removing bay from within 2.5 m of the trunk of a susceptible oak; (2) extending bay foliage-oak trunk clearance to 5 m where possible, especially in the direction(s) from which storm winds blow; (3) pruning low branches to obtain up to 5 m of clearance in the lower canopy even if upper canopy bay branches are present at closer horizontal distances; and (4) eliminating poison oak climbing at canopy level within an oak or an adjacent tree within 2.5 m of the oak trunk.

Tomlinson, J.A.; Barker, I.; and Boonham, N. 2007. Faster, simpler, more specific methods for improved molecular detection of *Phytophthora ramorum* in the field. Appl. Environ. Microbiol. DOI: 10.1128/AEM.00161-07.

Abstract: *Phytophthora ramorum* is the causal agent of sudden oak death. The pathogen also affects a wide range of tree, shrub, and herbaceous species in natural and landscaped environments, as well as plants in the nursery industry. A TaqMan real-time PCR method for the detection of this pathogen in the field has been described previously; this paper describes the development of a number of assays based on this method which have various advantages for improved use in the field. A Scorpion real-time PCR assay was developed which is twice as fast as TaqMan, allowing detection of *P. ramorum* in less than 30 minutes. A loop-mediated isothermal amplification (LAMP) assay also was designed, which allowed sensitive and specific detection of *P. ramorum* in 45 minutes using only a heated block. A positive reaction was identified by detection of the LAMP product by colour change visible to the naked eye.

OTHER RESEARCH OF INTEREST

Schwingle, B.W.; Juzwik, J.; Eggers, J.; and Moltzan, B. 2007. *Phytophthora* Species in Soils Associated with Declining and Nondeclining Oaks in Missouri Forests. Plant Disease 91:633. Published online as DOI: 10.1094/PDIS-91-5-0633A.

RESOURCES

The American Phytopathological Society's May APSnet Feature, "Why are *Phytophthora* and other Oomycota not true Fungi?" can be found at http://www.apsnet.org/online/feature/oomycetes/. The article covers the characteristics of true Fungi and why Oomycota are grouped with some algae as part of either the Chromista or Straminipila kingdoms.

Western Australia's regional branch of the Australasian Plant Pathology Society is featuring *P. ramorum* online in June as the "Pathogen of the Month." Recent inoculation trials of Australian plants in California and Europe, as well as current climatic risk models, indicate that *P. ramorum* poses a significant biosecurity threat to the Australasian region. The susceptibility and sporulation potential of native Australian plant species is currently being investigated, as well as modeling the climatic suitability of Australian regions for *P. ramorum*. The 'Pathogen of the Month' initiative commenced in February 2006 to disseminate information on new pathogens and pests among local and national researchers. The June 'Pathogen of the Month' article can be found at http://www.australasianplantpathologysociety.org.au/.

CALENDAR OF EVENTS

10/15 – 10/18 - XVI International Plant Protection Congress 2007, Glasgow, UK; Full details on the recently announced call for papers can be found at: http://www.bcpc.org/IPPC2007/Call%5Ffor%5FPapers/. For more information, contact Dr. Slawson, PHSI DEFRA, at: david.slawson@defra.gsi.gov.uk.