

CALIFORNIA OAK MORTALITY TASK FORCE REPORT OCTOBER 2009

MONITORING

The Oregon Department of Forestry (ODF) has found 41 new P. ramorum

infestations in Curry County to date in 2009, for a total of 85 dead trees within 14 acres, resulting in 31 treatment areas over 230 acres. Another 10 trees still remain to be checked on the ground following a July aerial survey. The new sites are within the known infested area, are small in size, and have very few infected trees; however, several of the sites are along the west and northwest edge of the infestation. Many of the confirmed sites have already been treated, yet cutting and burning efforts on many of the private land sites have not been completed due to funding issues, as is the case with a few of the 2008 sites.

One of the more high-profile sites identified is on US Forest Service (Gold Beach Ranger District, Rogue River-Siskiyou National Forest) land near the Chetco River, confirmed positive on 8/21/09. Following the Sudden Oak Death aerial survey, the ODF ground survey crew identified a symptomatic 7-inch diameter tree that was confirmed positive by the Hanson lab, Oregon State University. The tree is located in the understory of a mature coast redwood/Douglas-fir stand adjacent to the very popular Redwood Nature Trail that loops through the area. The trail is used throughout the year and is connected to Loeb State Park on the Chetco River via a riverfront trail. As a result of the find, the trail was officially closed to the public on 8/28 to reduce the risk of pathogen spread along the river trail and into Loeb State Park.

Additional sampling of host vegetation near the confirmed tanoak was conducted on 9/2; no other infected plants were found. A treatment boundary (approximately 300' radius from the infected tree and encompassing approximately 7 acres) was then flagged, and the site evaluated. The area is Late Successional Reserve, marbled murrelet habitat, northern spotted owl habitat, and incorporates a stream (non-fish-bearing) that flows into the Chetco River. A Decision Memo to treat the site using a combination of herbicide (tanoak), and cutting/piling/burning of tanoak, Pacific rhododendron, and evergreen huckleberry was signed by the District Ranger on 9/15. Treatment is expected to begin in early October. The trail will remain closed throughout the time treatment is occurring and until information from post-treatment monitoring of soil and vegetation indicates low risk of pathogen spread. Mitigation efforts on the trail using western red cedar chips are also under consideration. For additional information, contact Ellen Goheen at egoheen@fs.fed.us.

MANAGEMENT

The Washington State Department of Agriculture is working with the USDA

Animal and Plant Health Inspection Service and Forest Service, as well as the State Department of Natural Resources and Pierce County, to mitigate the impacts of the previously identified *P. ramorum*-positive salal plants in a native landscape adjacent to a Pierce County retail nursery. The response will include remounting drainage ditches where the landscape finds were made and lining them with rock. This will prevent any



future nursery runoff from coming in contact with susceptible *P. ramorum* hosts. The infected salal plants have been killed with herbicide, and the debris cleaned out of the ditches. For more information, contact Brad White at <u>bwhite@agr.wa.gov</u>.

NURSERIES

As of 9/18/09, the Oregon Department of Agriculture (ODA) has tested 24,189 samples from 583 grower sites in 2009, and identified six *P. ramorum*-positive nurseries. The USDA Confirmed Nursery Protocol (CNP) has been enacted at all nurseries, and four have completed the process. Of the two nurseries remaining, one entered the CNP process in early September while the other is a recurrent nursery in the midst of an extensive clean up effort. High-risk surveys are also being conducted at nurseries that grow and ship *Rhododendron* and *Camellia*. As of 9/18, the Department has tested 2,853 samples from 143 grower sites. All samples have been negative for *P. ramorum*.

The ODA will also complete data collection for the Grower Assisted Inspection Program (GAIP) pilot study in October. GAIP requires participating nurseries to adopt a systems approach to manage *Phytophthora* disease problems within their growing areas. ODA then audits the nurseries to ensure compliance with the program. Participating nurseries have now been in the program with systems approaches in place for a full year. However, because some nurseries may have residual *Phytophthora* populations present, ODA anticipates it will take more than 1 year to accurately assess the efficacy of the systems approach. For more information, contact Nancy Osterbauer at <u>nosterba@oda.state.or.us</u>.

RESEARCH

Assessing the potential of Silv-Ex[®] and Silv-Ex Plus[®] to eradicate *Phytophthora ramorum* spores in water - Based on two studies at UC Davis, using Silv-Ex[®] or Silv-Ex Plus[®] (foaming agent used in fire control) to treat water that is potentially infested with *P. ramorum*, greatly reduces, but does not eliminate, the concern for inadvertently spreading the pathogen through fire suppression efforts. For more information, contact Kamyar Aram at kamaram@ucdavis.edu.

Tanoak is renamed: Manos, P.S.; Cannon, C.H.; and Oh, Sang-Hun. 2008. Phylogenetic Relationships and Taxonomic Status of the Paleoendemic Fagaceae of Western North America: Recognition of a New Genus, Notholithocarpus. Madroño, Vol. 55, No. 3:181–190.

Abstract: We investigated the phylogenetic relationships and taxonomic status of the castaneoid component (*Lithocarpus* and *Chrysolepis*) of the family Fagaceae that is endemic to the California Floristic Province (CA-FP). Over 7800 basepairs of nuclear and chloroplast DNA were analyzed in 17 taxa representing the breadth of phylogenetic diversity in the family. The genus *Lithocarpus*, as currently defined, is clearly polyphyletic due to the inclusion of *L. densiflorus*. Here, we designate this taxon as a new genus, *Notholithocarpus*, which can be recognized morphologically by its relatively small, subprolate pollen. *Notholithocarpus* is more closely related to *Quercus*, *Castanea*, and *Castanopsis*; *Chrysolepis* was resolved as the sister group to *Lithocarpus* sensu



stricto. These results indicate that *Notholithocarpus* does not possess true 'flower cupules,' which define *Lithocarpus* sensu stricto, but like the oaks, the single flower per cupule is derived through the abortion of lateral flowers within each cupule. Further study is required to confirm this characteristic. A formal taxonomic treatment is presented with new combinations.

Bilodeau, G.; Pelletier, G.; Pelletier, F.; Lévesque, C.A.; and Hamelin, R.C. 2009. Multiplex real-time polymerase chain reaction (PCR) for detection of *Phytophthora ramorum*, the causal agent of sudden oak death. Canadian Journal of Plant Pathology 31:195-210.

Abstract: Since 1995, *Phytophthora ramorum* has been causing sudden oak death in California and Oregon, affecting tens of thousands of oak trees over large areas and infecting more than 100 other plant species. Quarantine measures are in effect, and regulatory agencies need to detect this organism in a timely and efficient manner. Various molecular assays have been developed over the past few years, and redundancy using multiple gene regions of *P. ramorum* was shown to increase the reliability in detecting the pathogen. However, such multi-gene assays require different polymerase chain reactions (PCRs) to test a single sample. To improve P. ramorum detection, three different TaqMan assays were multiplexed with a fourth TaqMan specific to the Phytophthora genus in a single reaction. A second multiplex TaqMan PCR assay to detect oomycetes and give a positive PCR reaction in the presence of plant DNA was also designed and tested in conjunction with the *P. ramorum* internal transcribed spacer (ITS) and Phytophthora genus TaqMan assays. These assays were tested on different Phytophthora species and were verified on two different sets of field samples previously assayed by other laboratories. These were obtained from multiple field hosts infected by various Phytophthora species, and the DNA from one set was extracted from ELISA lysates. All known P. ramorum samples from pure cultures or field samples were detected using these multiplex real-time PCR assays. In general, TaqMan multiplex assays showed lower detection sensitivity than single separated reactions. However, the multiplex assays still detected P. ramorum accurately while decreasing the cost and increasing throughput.

Collins, B.R.; Parke, J.L.; Lachenbruch, B.; and Hansen, E.M. 2009. The effects of *Phytophthora ramorum* infection on hydraulic conductivity and tylosis formation in tanoak sapwood. Canadian Journal of Forest Research 39(9): 1766–1776. DOI: 10.1139/X09-097

Abstract: Tanoak (*Lithocarpus densiflorus* (Hook. and Arn.) Rehder) is highly susceptible to sudden oak death, a disease caused by the oomycete *Phytophthora ramorum* Werres, De Cock & Man in't Veld. Symptoms include a dying crown, bleeding cankers, and, eventually, death of infected trees. The cause of mortality is not well understood, but recent research indicates that water transport is reduced in infected trees. One possible mechanism causing the reduction in hydraulic conductivity is the presence of tyloses in xylem vessels. The development of tyloses was studied in relation to



hydraulic conductivity in *P. ramorum*-infected sapwood. Inoculated logs showed a greater abundance of tyloses than noninoculated logs after 4 weeks. Inoculated trees with xylem infections had significantly more tyloses than noninoculated trees. In addition, the increase in number of tyloses was associated with a decrease in specific conductivity, suggesting that tyloses induced by infection with *P. ramorum* may interfere with stem sap flow. Over time, tylosis development increased in tissues farther from the inoculation site, in advance of the vertical spread of infection. The results suggest that infected sapwood contains numerous tyloses, which could significantly impede stem water transport.

Goss, E.M., Larsen, M., Chastagner, G.A., Givens, D.R., and Grünwald, N.J. 2009. Population genetic analysis infers migration pathways of *Phytophthora ramorum* in US nurseries. PLoS Pathogens 5(9): e1000583.

Abstract: Recently introduced, exotic plant pathogens may exhibit low genetic diversity and be limited to clonal reproduction. However, rapidly mutating molecular markers such as microsatellites can reveal genetic variation within these populations and be used to model putative migration patterns. *Phytophthora ramorum* is the exotic pathogen, discovered in the late 1990s, that is responsible for sudden oak death in California forests and ramorum blight of common ornamentals. The nursery trade has moved this pathogen from source populations on the West Coast to locations across the United States, thus risking introduction to other native forests. We examined the genetic diversity of P. ramorum in United States nurseries by microsatellite genotyping 279 isolates collected from 19 states between 2004 and 2007. Of the three known P. ramorum clonal lineages, the most common and genetically diverse lineage in the sample was NA1. Two eastward migration pathways were revealed in the clustering of NA1 isolates into two groups, one containing isolates from Connecticut, Oregon, and Washington and the other isolates from California and the remaining states. This finding is consistent with trace forward analyses conducted by the US Department of Agriculture's Animal and Plant Health Inspection Service. At the same time, genetic diversities in several states equaled those observed in California, Oregon, and Washington and two-thirds of multilocus genotypes exhibited limited geographic distributions, indicating that mutation was common during or subsequent to migration. Together, these data suggest that migration, rapid mutation, and genetic drift all play a role in structuring the genetic diversity of *P. ramorum* in US nurseries. This work demonstrates that fast-evolving genetic markers can be used to examine the evolutionary processes acting on recently introduced pathogens and to infer their putative migration patterns, thus showing promise for the application of forensics to plant pathogens.

Shishkoff, N. 2009. Evaluating ornamentals as physical barriers between blocks of plants susceptible to *Phytophthora ramorum*, 2007-2008. <u>Plant Disease Management</u> Reports 3:OT029.



MEETINGS

The Continental Dialogue on Non-Native Forest Insects and Diseases Fifth Meeting will be held this month from 10/27 - 10/28 at the Fort Mason Center in San Francisco. The meeting will include two concurrent field trips, one on SOD nursery and forest issues and the other on Port of Oakland inspections and handling of wood packaging. Panel sessions will also be offered on the risks of firewood movement, interstate movement of *P. ramorum*, preventing and slowing the spread of pests of non-quarantine significance, solid wood packaging as a pathway for invasives, and outreach and education challenges. The meeting is open to anyone interested in getting involved. Registration is required. For more information, see the Calendar of Events below or go to www.continentalforestdialogue.org.

REGULATIONS

USDA APHIS PPQ will conduct a comprehensive review of the *P. ramorum* federal program during the first quarter of the 2010 fiscal year. The purpose of the review is to openly exchange ideas and perspectives on federal and state efforts to regulate *P. ramorum.* Some of the goals of the review are to identify and clarify stakeholder concerns and program expectations, determine what's working well and what isn't, and develop recommendations for program improvement. At this time the review is tentatively scheduled for early December 2009. For more information, contact Scott Pfister at <u>Scott.E.Pfister@aphis.usda.gov</u>.

The USDA Animal and Plant Health Inspection Service (APHIS) Official

Regulatory Protocol for *Phytophthora ramorum* Detections in Residential or Landscaped Commercial Settings has been updated. The new "Confirmed Residential Protocol: Version 2.0," revised 9/1/09, has been posted to the APHIS website at: <u>http://www.aphis.usda.gov/plant_health/plant_pest_info/pram/downloads/pdf_files/reside_ntial-protocol-final%20.pdf</u>

RESOURCES

Chapter 6 of the "Sudden Oak Death and Phytophthora ramorum: A Summary of the Literature" is now available on the Task Force website at http://www.suddenoakdeath.org/html/sod_literature_summary.html. The chapter, titled "Economic and Environmental Impacts of Phytophthora ramorum – Literature Review Completed Through August, 2009" is the final chapter in the series.

Related Resource – A goldspotted oak borer website, hosted by the University of California's Agriculture and Natural Resources Division, is newly available at http://groups.ucanr.org/GSOB/index.cfm. Topics addressed include identification of the insect, host trees, impacts, known infestation sites, firewood issues, treatment options, how to report a find, and additional resources.

PERSONNEL

Scott Pfister was hired in June 2009 as the new USDA APHIS PPQ Director of Forest Pest Programs. APHIS created the position in response to the expanding nature of



forest health issues in the US. In this role, Scott oversees forest pest programs, such as those for Asian longhorned beetle, *Phytophthora ramorum*, and emerald ash borer, and he works to coordinate the development of forest pest mitigation measures at the national level. Prior to working in this capacity, Scott had 19 years of service working with plant protection organizations. He most recently worked for the Vermont Department of Forests, Parks, and Recreation, where he held the position of Forest Resource Protection Chief. In that capacity, he managed Vermont's forest health programs, including long-term and fixed plot monitoring, exotic forest pest surveys, aerial survey and suppression programs, and forest regulatory pest programs. He also served as the State Plant Regulatory Official and Plant Pathologist for the Vermont Agency of Agriculture. Scott can be reached at Scott.E.Pfister@aphis.usda.gov.

Erin Lovig has been hired as an Interior Pest Exclusion Agricultural Biologist with

the California Department of Food and Agriculture (CDFA). In her new position she will be responsible for assisting with SOD-related regulatory inquires and statewide management of the regulatory program for *P. ramorum*. Prior to coming to the Pest Exclusion Branch, Erin worked at the CDFA Plant Pest Diagnostic Center in the Plant Pathology lab. Her duties included running molecular diagnostic assays for *P. ramorum* and Asian Citrus Greening disease, as well as many other pathogens. Erin can be reached at <u>elovig@cdfa.ca.gov</u> or (916) 654-0312.

FEATURE STORY

Haas, B. J., et al. 2009. Genome sequence and analysis of the Irish potato famine pathogen *Phytophthora infestans*. Nature: online 09 September 2009. DOI: 10.1038/nature08358.

Over 90 authors from around the globe contributed to the genetic sequencing of *Phytophthora infestans* and its comparison to two other sequenced *Phytophthora* species, *P. sojae* (cause of soybean root rot) and *P. ramorum*. While the Haas et al. (2009) paper published in Nature is primarily focused on the *P. infestans* genome, this summary focuses on what can be learned about *P. ramorum* by genomic comparison.

Amongst all three *Phytophthora* species, there is an unusual genome organization where blocks of conserved gene order (with high gene density and low repeat content) are mixed with blocks where gene order is not conserved (with low gene density and high repeat content). This pattern of intergenic spacing and repeat content seen in *P. infestans* and the other *Phytophthoras* has been suggested for large, unsequenced genomes in the grass family (Poaceae) but has so far not been seen in the genomes of other sequenced eukaryotes.

Within blocks of conserved genes, the genes are tightly spaced in all three genomes (median intergenic distance of 633 base pairs for *P. ramorum*). These conserved blocks represent ~90% of the size of the three genomes, with a core set of approximately 8,500 clusters of shared genes. These core genes code for cellular processes including DNA replication, transcription, and protein translation. In regions between conserved blocks,



intergenic distances are greater and increase with increasing genome size (median 1.5 kb for *P. ramorum* versus 3.7 kb for *P. infestans*). Despite the high degree of similarity between the species, the *P. infestans* genome is much larger than the other two. Within *P. infestans*' genome, there is an abundance of repetitive DNA in the blocks of non-conserved genetic material which accounts for 74% of its size. By contrast, only 24% of *P. ramorum*'s genome is made up of these DNA repeats.

Like many pathogens, *Phytophthora* species secrete effector proteins that alter host physiology and facilitate colonization. Two notable effector protein genes are the RXLR and Crinkler (CRN) cytoplasmic effectors. RXLR genes are more numerous in *P. infestans*, with ~60% more than *P. sojae* or *P. ramorum*. CRN cytoplasmic effectors were also numerous in the *P. infestans* genome (196 CRN genes for *P. infestans* versus 19 CRN genes for *P. ramorum*), as well as being unexpectedly complex and diverse.

Both of these CRN and RXLR genes occur in the sparse, repeat heavy areas of the *P*. *infestans* genome rather than the core, high-density gene areas where the three species are most similar. Since these RXLR and CRN families lie in the most dynamic areas of the genome there is a great ability for them to quickly evolve. Interestingly, another group of genes called NPP1-like proteins that induce necrosis and also are implicated in pathogenesis have 59 genes in *P. ramorum* versus 27 in *P. sojae*. All of these genes are thought to play an important role in the pathogen's ability to adapt to its host plants. Having the blueprint for the genetic arsenal in *Phytophthora* that makes this pathogen group so hard to control is a major step towards development of smart (i.e., informed) breeding and fungicide development programs. We expect that knowing which genes are evolving rapidly will allow breeders and plant pathologists to develop more targeted disease control measures.

CALENDAR OF EVENTS

10/7 - SOD Treatment Workshop; meet at oak outside of Tolman Hall, UC Berkeley Campus; 1 – 3 p.m.; Pre-registration is required. This class is free and will be held rain or shine. To register, email kpalmieri@berkeley.edu, and provide your name, phone number, affiliation and license number (if applicable), and the date for which you are registering. For more information, go to <u>http://nature.berkeley.edu/garbelotto/english/sodtreatmenttraining.php</u> or contact Katie Palmieri at (510) 847-5482 or kpalmieri@berkeley.edu.

10/27 – 10/28 –Continental Dialogue on Non-Native Forest Insects and Diseases Fifth Meeting; Fort Mason Center, San Francisco; For more information on the meeting, or to register, go to <u>http://www.continentalforestdialogue.org/events/dialogue/2009-10-27/index.html</u> or contact Dana Goodson at <u>dgoodson@resolv.org</u> or (202) 965-6209. For more information about the Dialogue, go to: <u>www.continentalforestdialogue.org</u>.

11/4 - SOD Treatment Workshop; meet at oak outside of Tolman Hall, UC



Berkeley Campus; 1 - 3 p.m.; Pre-registration is required. For more information, see the 10/7 listing above.

- 11/17 11/18 58th Annual Meeting of the California Forest Pest Council, What's Ailing California's Forests?; Heidrick Ag History Center; 1962 Hays Lane, Woodland, CA 95776; For more information, contact Kim Camilli at 805-550-8583 or kim.camilli@fire.ca.gov or go to the CA Forest Pest Council website at http://caforestpestcouncil.org/.
- 12/9 SOD Treatment Workshop; meet at oak outside of Tolman Hall, UC Berkeley Campus; 1 – 3 p.m.; Pre-registration is required. For more information, see the 10/7 listing above.
- 3/7/10 3/12/10 5th IUFRO Phytophthora in Forest Trees and Natural Ecosystems Conference; Rotorua, New Zealand; For more information or to register, go to <u>http://www.phyto2010.com/registration.html</u>. Early registration discounts are available until 10/31/09. Abstracts are due by 10/31/09.

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