



## CALIFORNIA OAK MORTALITY TASK FORCE REPORT AUGUST 2008

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### MONITORING

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***P. ramorum* has been confirmed in the Little River at Van Damme State Park in Mendocino County near the town of Mendocino.** Taken from March water baiting samples, the positive cultures represent the northern most detection of the pathogen in the County. Plans are under way to conduct ground surveys of the watershed for terrestrial symptoms. For more information, contact Kamyar Aram at [kamaram@ucdavis.edu](mailto:kamaram@ucdavis.edu).

### NURSERIES

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**A San Mateo County retail nursery was found to have several *P. ramorum*-positive species of plants during an annual inspection.** The nursery is not under compliance for *P. ramorum* and does not ship out of the quarantined counties; therefore, it falls under the California Department of Food and Agriculture's (CDFA) nursery stock standards of cleanliness for cleanup of infection. Upon notification of the positive plants, the nursery owner destroyed the required plants per CDFA's nursery stock standards of cleanliness and voluntarily destroyed the remainder of the plants in the lot even though they were not infected. Trace-back investigations are underway.

All three *P. ramorum* lineages (NA1, EU1, and NA2) were identified during isolate analysis. The shade house where the EU1 and NA1 lineages were detected was inspected, but no symptomatic plants were identified. No plants are currently in the areas that held infected plants. This nursery is not an interstate shipper and is not under the *P. ramorum* Nursery Compliance Program.

Numerous suggestions on best management practices to eliminate or reduce the pathogen levels in the nursery were made to the nursery owner. Continued surveillance and sampling will be carried out as per regulations. Further discussions are scheduled with the nursery owner to assist in development of plans to reduce *P. ramorum* in the nursery.

**A small Oregon retail nursery in Tillamook County was found to have *P. ramorum*-positive *Pieris* sp. during a June 20<sup>th</sup> compliance inspection.** The nursery does not ship out of state, and the retail Confirmed Nursery Protocol (rCNP) is underway.

**A Kings County, Washington wholesale nursery was found to have *P. ramorum*-positive *Viburnum tinus* (Spring Bouquet) on June 9<sup>th</sup> during a nursery stock cleanliness inspection.** The nursery had also been found positive for *P. ramorum* in 2007. CNP is underway.

**A North Carolina retail nursery was found to have *P. ramorum*-infected *Rhododendron* sp., *R. catawbiense* (Slam Dunk), and *Kalmia latifolia* (Bulls eye) on June 6, 2008 as part of a Cooperative Agricultural Pest Survey (CAPS).** The rCNP is underway at the facility.



***P. ramorum*-infected *Rhododendron* sp. and *Kalmia* sp. were found at a South Carolina retail nursery on July 14, 2008 as a result of a trace forward investigation from the NC detection on June 6<sup>th</sup>. The rCNP is underway at the facility. Neither this nursery nor the NC 6/6 nursery have previously been found positive for the pathogen.**

**Two Alameda County sites are under consideration for a mock nursery location.** At a meeting with the Alameda County Board of Supervisors, Public Works Department, County Agricultural Commissioner, and California Department of Food and Agriculture, it was announced that the County is very eager to cooperate on the development of the mock nursery. The abandoned equestrian site previously under consideration may still be available if a plan to share the site with horses and riders can be arranged. An alternate location near the equestrian site has also been proposed by the Public Works Division. The next meeting with the County Agencies will include more of the affected parties (Fish and Game, Flood Control, etc.) and will include a visit to the newly proposed site. Hopes are to move this process along as quickly as possible, but some zoning and environmental impact issues must be addressed.

If implemented, the mock nursery would facilitate research necessary to more fully understand the epidemiology of *P. ramorum* in nurseries. For more information, contact Kathy Kosta at [KKosta@cdfa.ca.gov](mailto:KKosta@cdfa.ca.gov).

#### **FEATURED RESEARCH**

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**Viability of *Phytophthora ramorum* after passage through slugs; Jennifer Parke<sup>1</sup>; Elizabeth Stamm<sup>1</sup>; Akiko Oguchi<sup>2</sup>; Elizabeth Fichtner<sup>2</sup>; and Dave Rizzo<sup>2</sup>; <sup>1</sup>Oregon State University and <sup>2</sup>University of California Davis**

In western US forests where *P. ramorum* is found, chlamydospores are produced in abundance; however, their role in the disease cycle is uncertain because triggers for their germination are not known, and under lab conditions they germinate at a low or unpredictable frequency. In some *Phytophthora* species, the germination of oospores has been increased by passage through the alimentary canal of snails or treatment with snail enzymes. Consequently, we conducted experiments that demonstrated the viability of *P. ramorum* colonies after passage through the digestive tracts of two species of slugs: [banana slugs \(\*Ariolimax columbianus\*\)](#) and [the gray garden slug \(\*Deroceras reticulatum\*\)](#).

Slugs that ingested pure cultures of *P. ramorum*, and those that ate strawberries to which chlamydospores were applied, excreted feces with an abundance of [chlamydospores](#). After passage through the slugs, many chlamydospores either germinated directly, often with multiple germ tubes, or indirectly to form sporangiophores. Feces plated on *Phytophthora*-selective medium yielded *P. ramorum* colonies, and when placed on tanoak or rhododendron leaves, banana slug fecal samples caused infection.

Experiments were also conducted to determine if banana slugs can acquire *P. ramorum* from infected plant sources. Pacific banana slugs were placed in lidded tubs containing [artificially infested "litter"](#) (a mixture of inoculated bay, tanoak, and rhododendron leaves



with *P. ramorum* lesions) as well as in control tubs with non-infested “litter.” Slugs in both containers did not eat the leaves, but rather they appeared to graze the leaf surfaces. After transferring slugs to clean, empty tubs, feces excreted within the first 24 - 48 hours were collected and examined microscopically. Numerous chlamydospores were found present in slug feces from *P. ramorum*-infested “litter,” but not from non-infested “litter.” Banana slugs allowed to feed on *P. ramorum* cultures and subsequently placed in containers with [tanoak logs](#) transmitted disease to 2 of 9 logs. Wounding of the bark was not necessary for lesion development.

These observations have led us to consider the possible role of banana slugs in [chlamydospore germination](#) and disease transmission. Funding is currently being pursued for more thorough investigations using *Ariolimax columbianus* (Pacific banana slug) and *Ariolimax californicus* (California banana slug). Banana slugs are chosen for the study because of their preferred habitat, geographic range, pattern of seasonal activity, and demonstrated role as vectors of fungal spores. They primarily eat herbaceous plants, fungi, and lichens, consuming up to their body weight in food daily. They live on the forest floor and in trees up to 20 m above the ground, and can move 10 m in a 24-hr period. Requiring high moisture for producing slime used for locomotion, navigation, mating, and self-defense, they are most active during the rainy season when temperatures are mild, burrowing into soil during harsher summer and winter months. A 5-year study of *A. dolichophallus* (slender banana slug) in Big Basin State Park (CA) showed that they are most active during April-June, with a second peak of activity in November.

Banana slugs have an important role in decomposition in our forests. Slug findings to date have only been performed under lab conditions; therefore, results may not reflect their food preferences and feeding behavior in nature. There is currently no evidence that slugs actually transmit disease under field conditions. Any potential role of slugs in transmission of *P. ramorum* is likely to be very small compared to transmission by aerial propagules dispersed by wind or rain, which have been demonstrated to occur in infested forests.

## RESEARCH

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**Davidson, J.M.; Patterson, H.A.; and Rizzo, D.M. 2008. Sources of inoculum for *Phytophthora ramorum* in a redwood forest. *Phytopathology* 98:860-866.**

Abstract: Sources of inoculum were investigated for dominant hosts of *Phytophthora ramorum* in a redwood forest. Infected trunks, twigs, and/or leaves of bay laurel (*Umbellularia californica*), tanoak (*Lithocarpus densiflorus*), and redwood (*Sequoia sempervirens*) were tested in the laboratory for sporangia production. Sporangia occurred on all plant tissues with the highest percentage on bay laurel leaves and tanoak twigs. To further compare these two species, field measurements of inoculum production and infection were conducted during the rainy seasons of 2003-04 and 2004-05. Inoculum levels in throughfall rainwater and from individual infections were significantly higher for bay laurel as opposed to tanoak for both seasons. Both measurements of inoculum



production from bay laurel were significantly greater during 2004-05 when rainfall extended longer into the spring, while inoculum quantities for tanoak were not significantly different between the 2 years. Tanoak twigs were more likely to be infected than bay laurel leaves in 2003-04, and equally likely to be infected in 2004-05. These results indicate that the majority of *P. ramorum* inoculum in redwood forest is produced from infections on bay laurel leaves. Years with extended rains pose an elevated risk for tanoak because inoculum levels are higher and infectious periods continue into late spring.

**Hong, C.; Richardson, P.A.; and Kong, P. 2008. Pathogenicity to ornamental plants of some existing species and new taxa of *Phytophthora* from irrigation water. Plant Disease 92:1201-1207.**

Abstract: Eighteen isolates from 12 species of *Phytophthora*, including several new taxa, were tested for pathogenicity to six ornamental and four vegetable species. The following three inoculation methods were used depending on infection court targeted: vermiculite culture inoculation for roots, agar block inoculation for fruit, and zoospore inoculation for foliage. All six new taxa (*P. irrigata*, *P. hydropathica*, Dre III, Cil I, Cip-like, and Gon I) are pathogenic to one or more test plants. Specifically, taxon Cil I was identified as a growing threat to horticultural crops, particularly ornamental crops in container production nurseries. The potential host list of *P. tropicalis* was expanded to four new families (Apocynaceae, Asteraceae, Begoniaceae, and Fabaceae) and one additional genus within each of three existing families (Ericaceae, Cucurbitaceae, and Solanaceae). New potential hosts were also identified for other existing species of *Phytophthora*. The practical implications of these results in crop health management programs for both ornamental and vegetable crops locally, and for development and implementation of agricultural biosecurity programs globally, are discussed.

**Mascheretti, S.; Croucher, P.J.P.; Vettraino, A.; Prospero, S.; and Garbelotto, M. 2008. Reconstruction of the Sudden Oak Death epidemic in California through microsatellite analysis of the pathogen *Phytophthora ramorum*. Molecular Ecology 17, 2755–2768.**

Abstract: The genetic structure of the clonally reproducing Sudden Oak Death (SOD) pathogen in California was investigated using seven variable microsatellites. A total of 35 multilocus genotypes were identified among 292 samples representative of populations from 14 forest sites and of the nursery trade. AMOVA indicated significant genetic variability both within (44.34%) and among populations (55.66%). Spatial autocorrelation analyses indicated that Moran's index of similarity reached a minimum of 0.1 at 350 m, increased to 0.4 at 1500 m and then decreased to zero at 10 km. These results suggest a bimodal pattern of spread, with medium range dispersal (1500–10 000 m) putatively attributed to the presence of strong winds. Lack of genetic structure was identified for three groups of populations. One group notably included the nurseries' population and two forest populations, both linked to early reports of the pathogen. A neighbor-joining analysis based on pairwise  $\Phi_{ST}$  values indicated that the clade inclusive



of the nurseries' populations is basal to all California populations. A network analysis identified three common genotypes as the likely founders of the California infestation and proposes a stepwise model for local evolution of novel genotypes. This was supported by the identification in the same locations of novel genotypes and of their 1- or 2-step parents. We hypothesize that the few undifferentiated population groups indicate historical human spread of the pathogen, while the general presence of genetically structured populations indicates that new infestations are currently generated by rare medium or long-range natural movement of the pathogen, followed by local generation of new genotypes.

**Meentemeyer, Ross K.; Anacker, Brian L.; Mark, Walter; and Rizzo, David M.** 2008. Early Detection of Emerging Forest Disease using Dispersal Estimation and Ecological Niche Modeling. *Ecological Applications*, 18(2). pp. 377–390.

**Abstract:** Distinguishing the manner in which dispersal limitation and niche requirements control the spread of invasive pathogens is important for prediction and early detection of disease outbreaks. Here, we use niche modeling augmented by dispersal estimation to examine the degree to which local habitat conditions vs. force of infection predict invasion of *Phytophthora ramorum*, the causal agent of the emerging infectious tree disease sudden oak death. We sampled 890 field plots for the presence of *P. ramorum* over a three-year period (2003–2005) across a range of host and abiotic conditions with variable proximities to known infections in California, USA. We developed and validated generalized linear models of invasion probability to analyze the relative predictive power of 12 niche variables and a negative exponential dispersal kernel estimated by likelihood profiling. Models were developed incrementally each year (2003, 2003–2004, 2003–2005) to examine annual variability in model parameters and to create realistic scenarios for using models to predict future infections and to guide early-detection sampling. Overall, 78 new infections were observed up to 33.5 km from the nearest known site of infection, with slightly increasing rates of prevalence across time windows (2003, 6.5%; 2003–2004, 7.1%; 2003–2005, 9.6%). The pathogen was not detected in many field plots that contained susceptible host vegetation. The generalized linear modeling indicated that the probability of invasion is limited by both dispersal and niche constraints. Probability of invasion was positively related to precipitation and temperature in the wet season and the presence of the inoculum-producing foliar host *Umbellularia californica* and decreased exponentially with distance to inoculum sources. Models that incorporated niche and dispersal parameters best predicted the locations of new infections, with accuracies ranging from 0.86 to 0.90, suggesting that the modeling approach can be used to forecast locations of disease spread. Application of the combined niche plus dispersal models in a geographic information system predicted the presence of *P. ramorum* across ~8228 km<sup>2</sup> of California's 84 785 km<sup>2</sup> (9.7%) of land area with susceptible host species. This research illustrates how probabilistic modeling can be used to analyze the relative roles of niche and dispersal limitation in controlling the distribution of invasive pathogens.



**Reeser, P.W.; Sutton, W.; and Hansen, E.M. 2008. *Phytophthora* Species Causing Tanoak Stem Cankers in Southwestern Oregon.** Plant Disease. Disease Notes Vol. 92 No. 8 pg. 1252. DOI: 10.1094/PDIS-92-8-1252B

**Vincelli, Paul and Tisserat, Ned. 2008. Nucleic Acid–Based Pathogen Detection in Applied Plant Pathology.** Plant Disease Vol. 92 No. 5. pp. 660 – 669. DOI: 10.1094/PDIS-92-5-0660.

This review article about using molecular tools for plant pathogen detection highlights *Phytophthora ramorum* and how PCR is central to regulatory efforts to detect and eradicate this pathogen. The review discusses the limitations and pitfalls of molecular methods to guide readers toward using various detection tools effectively: false negatives and positives resulting from incomplete specificity, sample collection, viability of positives, contamination risks, and other topics are discussed.

#### **RELATED TOPICS**

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**Some oaks are losing leaves early this year as a result of the drought. In areas such as the Sierra Nevada foothills, blue oaks have begun changing color and dropping leaves.** When faced with low soil moisture, trees either keep their foliage and continue losing water through the leaf pores, or drop their leaves and conserve moisture.

Drought does stress trees, causing slower growth rates and greater susceptibility to insects and diseases. While long-term consequences of repeated droughts may be harmful, it is anticipated that most of the trees affected this year will likely recover during the winter and leaf out normally next spring. Early leaf loss may actually be more harmful to the ecosystem than to individual trees. Loss of leaves can hamper acorn development and maturity, reducing the number of acorns that will germinate and develop into seedlings. Additionally, reduced acorn production adversely affects the many wildlife species that rely heavily them for food. For more information, contact Doug McCreary at [mccreary@nature.berkeley.edu](mailto:mccreary@nature.berkeley.edu).

#### **RESOURCES**

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**USDA APHIS has posted Version 2.1 of the “Trace Forward Protocol for Nurseries that Received Plant Material Shipped from a Confirmed *Phytophthora ramorum* Infested Nursery”** to their website. To access the June 6, 2008 document, go to: [http://www.aphis.usda.gov/plant\\_health/plant\\_pest\\_info/pram/downloads/pdf\\_files/traceforwardprotocol.pdf](http://www.aphis.usda.gov/plant_health/plant_pest_info/pram/downloads/pdf_files/traceforwardprotocol.pdf). This version updates the definition and provides much more detailed and clearly written procedures to follow in the event that a nursery determined to be infested has shipped nursery stock to another nursery.

**USDA APHIS updated the Confirmed Nursery Protocol (CNP) Version 8 on June 26, 2008.** The updated document can be found on the APHIS website at: [http://www.aphis.usda.gov/plant\\_health/plant\\_pest\\_info/pram/downloads/pdf\\_files/CNPv8.0-7-20-07.pdf](http://www.aphis.usda.gov/plant_health/plant_pest_info/pram/downloads/pdf_files/CNPv8.0-7-20-07.pdf). The updates are limited to the Appendices 1, 3, 6, and 7. This provides



updated host information, contact information, soil and growing media detection procedures, and water sampling procedures.

#### **PERSONNEL**

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**In July, Russ Bulluck officially assumed the duties of the Center for Plant Health Science and Technology (CPHST) National Science Program Leader for Response, Recovery and Systems Technology (NSPL-RRST).** In his new role, he will be providing scientific information to APHIS Plant Protection and Quarantine (PPQ) regulatory officials to insure that actions taken have a sound scientific basis. This can include developing technical working groups for specific subjects and responding to new and ongoing emergencies, including *P. ramorum*.

Most recently, Russ served as the Acting Team Leader for Emergency Planning and Preparedness in Emergency and Domestic Programs, where he prioritized, prepared, edited and revised New Pest Response Guidelines and provided technical support for a variety of plant health emergencies. Over the last few years, Russ has been actively involved in PPQs response to significant plant health emergencies, including Sudden Oak Death. Russ can be reached at (919) 855-7646 or [Russ.Bulluck@aphis.usda.gov](mailto:Russ.Bulluck@aphis.usda.gov).

**Norm Dart has left his position with Gary Chastagner at the Washington State University Research and Extension Center for a position as the West Virginia Agricultural Plant Pathologist.** Norm can be reached at his new location via email: [ndart@ag.state.wv.us](mailto:ndart@ag.state.wv.us) or phone: (304) 558-2212, ext. 3720.

**As of July 21, 2008, Greg Parra is the National Program Staff Scientist for the Response and Recovery Systems Technologies (RRST) program with CPHST.** In his new role, one of his areas of responsibility will be assisting the RRST National Science Program Leader along with working in cooperation with other USDA and state personnel on programs related to *P. ramorum*. Greg can be reached at (919) 855-7548 or [Greg.R.Parra@aphis.usda.gov](mailto:Greg.R.Parra@aphis.usda.gov).

#### **CALENDAR OF EVENTS**

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**9/10 - Sudden Oak Death (SOD) Treatment Workshop; 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

[SODtreatment@nature.berkeley.edu](mailto:SODtreatment@nature.berkeley.edu), and provide your name, phone number, affiliation (if applicable), and the date for which you are registering. For more information, go to <http://nature.berkeley.edu/sodtreatment> or contact Katie Palmieri at (510) 847-5482 or [palmieri@nature.berkeley.edu](mailto:palmieri@nature.berkeley.edu).

**10/1 - Sudden Oak Death (SOD) Treatment Workshop; oak outside of Tolman Hall,**

UC Berkeley Campus; 1 – 3 p.m.; Pre-registration is required. This class is free. For more information, see the 9/10 listing above.

**11/12 - Sudden Oak Death (SOD) Treatment Workshop; oak outside of Tolman**

Hall, UC Berkeley Campus; 1 – 3 p.m.; Pre-registration is required. This class is free. For more information, see the 9/10 listing above.



**12/10 - Sudden Oak Death (SOD) Treatment Workshop; oak outside of Tolman Hall, UC Berkeley Campus; 1 – 3 p.m.; Pre-registration is required. This class is free. For more information, see the 9/10 listing above.**