CALIFORNIA OAK MORTALITY TASK FORCE REPORT
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RESEARCH

Sudden Larch Death? – Larch Susceptibility to *Phytophthora ramorum* in Oregon Forests. (summary of a Western International Forest Disease Work Conference 2011 poster)

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Recently an epidemic of 'Sudden Larch Death' caused by *P. ramorum* was reported from Great Britain. The disease was first observed in 2009 in mature Japanese larch (*Larix kaempferi)* plantations in southwest England (Webber et al. 2010). In 2010 it was estimated that roughly half a million larch trees were showing symptoms of *P. ramorum* infection from SW England to Wales and Northern Ireland (Brasier & Webber, 2010).

To test the susceptibility of Japanese larch as well as native North American Western larch (*Larix occidentalis)* to the strains of *P. ramorum* found in Oregon forests, potted seedlings were exposed to natural inoculation under infected tanoak trees in Curry County, Oregon. Douglas-fir *(Pseudotsuga menziesii*) was included for comparison.

Three hundred and six dormant, 2-yr-old seedlings, representing the three different species, were transplanted into gallon pots (two seedlings per pot) and exposed to *P. ramorum*. Presence of inoculum during the test was monitored with two baited rain traps at each site. Seedlings were examined periodically for mortality and symptoms and finally dissected to isolate the pathogen and confirm infection with PCR.

P. ramorum was isolated from symptomatic tissues of most of the Douglas-fir and western larch seedlings at the end of the study. It was not recovered from any control seedlings. PCR results were generally negative for samples that did not yield *P. ramorum* in culture. Overall, 17% of samples that were culture negative were PCR positive.

Results for Japanese larch were complicated by the low vigor of the seedlings. About half of the seedlings (including controls) died without bursting bud, and another 30% died shortly after bud burst. Recovery of *P. ramorum* from Japanese larch was low (25-35 %) across the symptom classes, although PCR results increased the *P. ramorum* positives for dead (class 4) seedlings to 53%. Both Douglas-fir and western larch seedlings had similar percentages of *P.ramorum* recovery and both were infected at all 10 of the field sites. *P. ramorum* was regularly isolated from advancing margins of lesions on twigs at the base of new growth. The main symptom observed was wilting or collapse of the new shoot. Often a girdling lesion low on the stem led to death of all distal tissues. Symptoms consistent with *P. ramorum* infection developed on most exposed seedlings of Douglas-fir and western larch (88% of Douglas-fir and 98% of western larch). 52% of exposed Douglas-fir seedlings were nearly dead or dead. Seedlings rated nearly dead or

dead totaled 59% for western larch. Twelve of the 102 Douglas-fir seedlings exposed in Curry County remained asymptomatic, but *P. ramorum* was recovered from small lesions on 4 of these. Only two western larch seedlings remained symptom free.

In conclusion:

- Exposure of potted seedlings beneath infected tanoak during periods of sporulation provides an effective and realistic susceptibility test.
- Symptoms and intensity of infection on Douglas-fir seedlings were comparable to artificial inoculation and natural infection observed in the forest.
- Western larch is susceptible to the Oregon forest lineage (NA1) of *P. ramorum* under environmental conditions prevalent in the epidemic area of SW Oregon. This confirms previous stem wound inoculation results.
- Japanese larch is also susceptible, although the degree of susceptibility cannot be determined from this experiment.

Japanese larch and various hybrids, as well as the native European larch, are grown as plantation species and are used commercially for timber throughout Europe. In western North America, Japanese larch and some hybrids have been planted in the Coast Ranges on a trial basis. These plantations must be considered at great risk if exposed to *P. ramorum*. The susceptibility of western larch seedlings in this experiment was comparable to Douglas-fir. Western larch is not native to the Coast Ranges, and has not been planted widely there. It remains an open question whether the environment in the western larch native range will support *P. ramorum*.

Western larch is native to the interior northwest, and according to a risk of *P. ramorum* spread map (Kelly et al. 2007), much of western larch's native range falls in a medium-low or medium risk zone. Such risk rating systems are weighted heavily by the presence of host plants, however, and larch was not a known host when this risk rating was done.

Bottle O' Bait Update - In situ baiting of water for *P. ramorum* with host foliage has been used in the field for at least 9 years in the western US and for more than 5 years by the USDA Forest Service for the National *P. ramorum* Early Detection Survey. During this time, the pathogen has been recovered from water in eight states. Leaving baits exposed in water for extended periods allows sampling of large volumes of water over time. However, it also leaves baits vulnerable to loss from vandalism and storm surges, requires sustained water flow during the exposure period, and requires two site visits for each sample period—once to deploy and once to retrieve baits. To address some of these limitations, an in vitro baiting assay using both intact leaves and leaf pieces of rhododendron was examined to recover *P. ramorum* from waterways draining known infested nurseries and forests. A water filtration assay also was used to provide a quantitative estimate of the density of propagules of *P. ramorum* in a water sample. Over a 3-year period, samples were collected from 18 waterways in five states where *P. ramorum* was known to be present based on results from in situ baiting. Each water sample (800 ml) was placed in a 1-liter Nalgene bottle with 20 rhododendron leaf pieces and one non-wounded leaf. Bottles were laid horizontally and held for 3 days at 18-22°C

in the dark. Leaves were then removed, rinsed in distilled water, and blotted dry. Pathogen detection was initiated immediately for leaf pieces, but whole leaves were placed in a moist chamber for up to 7 days to allow lesions to develop. Detection of P. *ramorum* was attempted for all in situ and in vitro baits by isolation on selective medium and by PCR.

The pathogen was detected at 16 of 18 sites by in situ baiting; in vitro (bottle method) baiting detected the pathogen at 14 of the 16 positive sites plus one site where in situ baiting was negative. Ten of the positive water samples detected by the in vitro assay had very low propagule densities: 0 or 1 cfu/liter as determined by filtration. The in vitro assay was nearly as effective as in situ baiting for recovering *P. ramorum* from water even though a very small volume of water was collected at one point in time. However, the lack of sampling over time may challenge pathogen recovery at some sites because only propagules present at the time of sampling can be collected and short-lived extremes in water conditions such as high water temperature and storm flows may reduce inoculum density below detectable levels. The in vitro assay, dubbed bottle o' bait (BOB), is being tested in a comparative pilot survey under field conditions to determine if it is as reliable as the current in situ baiting assay as a survey protocol. For more information, email Steve Oak, USDA Forest Service, Forest Health Protection.

Hüberli, D.; Hayden, K.J.; Calver, M.; and Garbelotto, M. 2011. Intraspecific variation in host susceptibility and climatic factors mediate epidemics of sudden oak death in western US forests. Plant Pathology. DOI: 10.1111/j.1365-3059.2011.02535.x.

Abstract: *Umbellularia californica* is one of the key infectious hosts of the exotic *Phytophthora ramorum*, which causes sudden oak death (SOD) in California and Oregon forests. This study provides a comprehensive analysis of the epidemiologically relevant parameters for SOD in California and southern Oregon, including potential differences between the two states. Experimental infection of *U. californica* leaves was optimal when leaves were wet for 6–12 h, temperature was approximately 19°C and pathogen concentration was approximately $2{\cdot}7 \times 10^4$ zoospores mL^{-1}. Seasonal variation in host susceptibility and disease incidence was examined for two populations by inoculating detached leaves at 12 dates and by monitoring naturally infected leaves, respectively. Susceptibility of *U. californica* and disease incidence varied significantly in time and the variation was highest for both in spring. Susceptibility of trees from 17 natural populations from California and southern Oregon was assessed in detached leaf inoculations. One California and three southern Oregon populations had significantly and repeatable lower average susceptibility in artificial inoculations, but differences among three selected California and Oregon populations were not significant in inoculations of seedlings grown from seed in a common garden. This study concludes that *U. californica* susceptibility has a large environmental component, yet still predicts potential disease severity in different sites especially where infestations are young or the pathogen has not yet arrived. The accuracy and utility of predictive risk models for *P. ramorum* will be enhanced by the inclusion of both the environmental and host susceptibility components.

Pérez-Sierra, A.; Álvarez, L.A.; Vercauteren, A.; Heungens, K.; and Abad-Campos, P. 2011. Genetic diversity, sensitivity to phenylamide fungicides and aggressiveness of *Phytophthora ramorum* on *Camellia*, *Rhododendron*, and *Viburnum* plants in Spain. Plant Pathology 60, 1069–1076. DOI: 10.1111/j.1365-3059.2011.02485.x.

Abstract: *Phytophthora ramorum* has been detected in official plant health surveys on *Rhododendron*, *Viburnum*, and *Camellia* in ornamental nurseries in northern Spain since 2003. A collection of 94 isolates of *P. ramorum* was obtained from 2003 to 2008 from plants with symptoms at different geographical locations. Isolates were identified based on morphology and sequence of the rDNA ITS region. Mating type, genetic variation, sensitivity to phenylamide fungicides and aggressiveness of these isolates were determined. All isolates belonged to the A1 mating type, ruling out the possibility of genetic recombination. Seven microsatellite markers were used to study genetic diversity; three out of the seven microsatellite markers were polymorphic within the Spanish population of *P. ramorum*. This study confirms that all Spanish isolates of *P. ramorum* belonged to the EU1 lineage. Twelve intralineage genotypes were detected, five that are unique to Spain (EU1MG38, EU1MG41, EU1MG37, EU1MG39, and EU1MG40) and seven that are also present in at least one other European country (EU1MG1, EU1MG29, EU1MG22, EU1MG13, EU1MG2, EU1MG18 and EU1MG26). Genotypes EU1MG37, EU1MG39, and EU1MG40 were isolated from *Rhododendron* from one region; EU1MG38 and EU1MG41 were isolated from *Camellia* from two different regions. Isolates of genotype EU1MG38 were resistant to metalaxyl and mefenoxam. The level of genetic diversity within the Spanish population of *P. ramorum* is limited and indicates a relatively recent clonal expansion.

The Proceedings of the 5th meeting of IUFRO unit 7.02.09 *Phytophthora* Diseases on Forest Trees, held in Rotorua, New Zealand from March 7-12, 2010, are being released by the New Zealand Journal of Forestry Science as individual papers following the review process. To access the papers, go to http://www.scionresearch.com/general/science-publications/science-publications/nzjfs/iufro-working-party-S07-02-09,-phytophthora-diseases-in-forests-and-natural-ecosystems-supplement-to-volume-41. If you wish to be notified when new papers are added to the website, sign up for the New Zealand Journal of Forestry Science alert service at http://www.scionresearch.com/general/science-publications/science-publications/nzjfs/sign-up-here-forjournal-alerts. As not all presentations from the meeting have been submitted for publication, abstracts from the meeting are available at http://www.scionresearch.com/__data/assets/pdf_file/0014/33341/Abstracts_Phyto2010.pdf.

MONITORING

An outbreak of *P. ramorum*-infected larch trees was confirmed in October in private woodlands north of Newton Stewart, Galloway following an aerial survey of western Scotland. Approximately 43 acres are affected. At this point it is unclear how the disease arrived there; site investigations are underway. Interim measures will be put in place as a precautionary measure pending further surveys this fall and next spring and

summer. These are likely to include restrictions on larch planting in the area and controls on the movement of larch timber from sites that may have become infected. The outbreak is the third on Japanese larch in Scotland, the first was a small area of Craignish Peninsula larch trees in late 2010, and the second was on the island of Mull in summer, 2011.

FUNDING

The UK has launched a Tree Health and Plant Biosecurity Action Plan that dedicates more than $11 million in funding over the next three years to combat exotic pests and diseases in an effort to protect the country's plants, trees, and habitats. Four key areas of activity addressed in the Plan include: import controls; practical actions; research; and communications and public engagement. Developed by the Forestry Commission; Department for Environment, Food, and Rural Affairs; Food, Environment, and Research Agency; the devolved administrations in Scotland and Wales; forestry and horticultural sectors; and non-government organizations, the Action Plan takes an integrated approach to tackling tree pests and diseases across the country. View the Plan in its entirety.

MANAGEMENT

The British Forestry Commission has stopped issuing new felling licenses for larch in high-risk areas over the winter to avoid infected material inadvertently being spread. After winter needle drop, it is impossible to identify disease symptoms and confirm if trees are infected. Once the trees have flushed with new needles next spring, any infected larch can be identified before felling so that biosecurity measures can be taken to avoid spreading the disease. The embargo will remain in effect until May 31, 2012. This is the second year in a row such an embargo has been put in place.

RELATED RESEARCH

Grünwald, N.J.; Werres, S.; Goss, E.M.; Taylor, C.R.; and Fieland, V.J. 2011. *Phytophthora obscura* sp. nov., a new species of the novel *Phytophthora* subclade 8d. Plant Pathology. DOI: 10.1111/j.1365-3059.2011.02538.x.

RELATED ISSUES

***Phytophthora lateralis* has been found in Devon (a large county in southwest** England) killing Lawson Cypress (*Chamaecyparis lawsoniana*) trees for the first time. The several infected trees identified were in a shelter hedge on an industrial estate. The trees will be felled and disposed of safely. The site is subject to biosecurity measures to prevent spreading the disease via contaminated soil, felling equipment, and other tools. Previous cases in the UK have only been identified in Scotland and Northern Ireland.

Until recently, *P. lateralis* was mostly known in the western states of the US and Canada, but outbreaks have also been recorded recently in Scotland, Northern Ireland, France, and The Netherlands. The pathogen has also been reported recently from Taiwan on Taiwanese yellow cedar (*Chamaecyparis obtuse* var. *formosana*). While not a significant forestry tree, Lawson Cypress is very popular in parks and gardens. If the disease

becomes established, it could be serious for the ornamental plant industry as Lawson cypress is an important ornamental conifer in the plant trade.

KUDOS

Congratulations to Susan Frankel and Ellen Goheen for being chosen to receive the 2011 Outstanding Achievement Award from the Western International Forest Disease Work Conference "for leadership in the science and practice of Forest Pathology and for critical contributions to the management of Sudden Oak Death." Only one award is granted each year, with the intent of recognizing outstanding achievement in the field of forest pathology in Western North America. This is the first time women have been granted the award.

CALENDAR OF EVENTS

11/8 – 11/11 - 2011 IUFRO Forest Protection Joint Meeting, Research Groups 7.02 – 7.03; Colonia del Sacramento, Uruguay; More information will be forthcoming. For questions, contact Alina Greslebin at agreslebin@ciefap.org.ar.

11/9 - 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, or for questions, 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 http://nature.berkeley.edu/garbelotto/english/sodtreatmenttraining.php.

11/13 – Sudden Oak Death workshop for Marin County area residents; Tam Valley Elementary School; 350 Bell Ln, Mill Valley; (backup indoor *rain location* is Tam Valley Community Center; 305 Bell Lane, Mill Valley; 11 a.m. – 1 p.m.

11/16 - 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 11/9 listing above.

6/18 – 6/22/12 – Sudden Oak Death Fifth Science Symposium; More information will be forthcoming.

9/9 – 9/14/12 – Sixth Meeting of the International Union of Forest Research Organizations IUFRO Working Party 7-02-09 "*Phytophthora* in Forests and Natural Ecosystems;" Colegio Mayor Universitario Nuestra Señora de la Asunción, Avd. Menéndez Pidal s/n, 14004 Córdoba, Spain; For more information, contact Mª Pérez Sierra at aperesi@eaf.upv.es.