Diagnosis and Management of *Phytophthora ramorum* Canker in Canyon Live Oak, an Atypical Bole Canker Host

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Abstract

Diagnosis of sudden oak death (SOD) in tanoak (*Notholithocarpus densiflorus* (Hook. & Arn.) Manos, Cannon & S.H. Oh) and susceptible red/black oak species (coast live oak, *Quercus agrifolia* Née; Shreve oak, *Q. parvula* Greene var. *shrevei* (C.H. Mull.) Nixon; California black oak, *Q. kelloggii* Newb.) is facilitated by the fact that infected trees commonly develop obvious bleeding bark cankers. In contrast, canyon live oak (*Q. chrysolepis* Liebm.) exhibits a different pattern of symptom expression, which has made it difficult to identify and document *Phytophthora ramorum* bole cankers in this species.

In 2002, *P. ramorum* was shown to cause dieback in small (<2 cm diameter) branches of canyon live oak, but no bole cankers in mature canyon live oaks were observed (Murphy and Rizzo 2003). In 2006, we observed a cluster of recently-killed mature canyon live oaks on Bolinas Ridge in Marin County. The affected trees were amid *P. ramorum*-infected California bay laurel (*Umbellularia californica* (Hook. & Arn.) Nutt. The overall pattern of disease was consistent with SOD, but affected trees had been dead too long to attempt pathogen isolation.

In 2008, mortality and large trunk cankers were seen in large, mature canyon live oaks in the Los Trancos Open Space Preserve in Santa Clara County. Symptomatic canyon live oaks had bole cankers that were extensively colonized by flat-headed borers and ambrosia beetles and had *Annulohypoxylon thouarsianum* sporulation. *Phytophthora ramorum* symptoms were common on foliage of California bay laurel, which was abundant in the vicinity of affected trees. However, we were unable to recover *P. ramorum* from symptomatic canyon live oak in several rounds of sampling of multiple trees. After ruling out other possible causes of tree decline, we surveyed the affected stand to determine the spatial pattern of disease in the stand. We found that the likelihood of canker symptoms and mortality in canyon live oak were highly associated with proximity to California bay laurel (fig. 1) and the amount of California bay laurel cover within 2.5 to 5 m. This relationship was nearly identical to that seen in SOD-affected coast live oak stands (Swiecki and Bernhardt 2008). We conducted another intensive round of sampling in the stand and obtained positive PCR detections of *P. ramorum* from trunk cankers on two symptomatic canyon live oaks, but no positive cultures.

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We subsequently tested pathogenicity of *P. ramorum* on detached large-diameter canyon live oak logs under lab conditions using established protocols (Kirk and Brasier, n.d.). Large, dark-brown cankers developed in the phloem of inoculated canyon live oak logs within 2 weeks after inoculation, and *P. ramorum* was readily re-isolated from canker margins (Aram et al. 2011). This provided the first clear indication about the internal appearance of early stage *P. ramorum* cankers on this species. However, the detached log assay does not provide reliable information about the appearance of external symptoms that develop in the field.

In July 2010, we initiated a field test by inoculating 18 canyon live oaks (25 cm average DBH) and two Shreve oaks, which served as positive controls, at a site in San Mateo County. Trees were inoculated with agar plugs taken from the margins of actively growing *P. ramorum* cultures, following procedures previously used for coast live oak (Rizzo et al 2002). Each tree was inoculated with two different local *P. ramorum* isolates and a control mock inoculation (sterile agar only). Nine of the inoculated canyon live oaks were located in a natural closed canopy stand on a northeast-facing slope that included some intermixed tanoak and California bay laurel. The canyon live oaks in this lower plot were relatively tall with small crowns. Some of the trees were partially overtopped and many had evident thinning. The other nine inoculated canyon live oaks and two Shreve oaks were in an adjacent hilltop stand (upper plot) that had been planted about 25 years earlier. The seed source(s) are unknown, but likely included trees from well beyond the lower plot area. Although the upper plot area also had a closed canopy, trees were much shorter and had wider crowns than in the lower plot. Most canopies were relatively dense, which has resulted in extensive dieback of the lower branches due to shading. The planting included a few tanoaks along with Shreve and canyon live oaks.

We evaluated symptoms periodically through June 2012, nearly 2 years after inoculation. Bleeding was seen in six of the 18 inoculated canyon live oaks. However, periods of active bleeding were short, so relatively few trees had visible bleeding at any given time (fig 2.). Only 17 percent (6/36) of the *P. ramorum* inoculation points developed any bleeding prior to sampling. The amount of bleeding was usually limited to a few drops up to about 2 mm in diameter. Only one tree, which developed a large canker within 4 months, had easily visible amounts of bleeding. Another tree that never showed bleeding developed large cankers which were detected by the presence of extensive ambrosia beetle boring dust about 1 year after inoculation and *A. thouarsianum* sporulation by 70 weeks after inoculation. No other canyon live oaks were colonized by these secondary organisms by June 2012. Both inoculated Shreve oaks developed large girdling cankers that overgrew the mock inoculation site. Bleeding was seen in one of these trees 19 weeks after inoculation.
We sampled at least two canyon live oaks from both the upper and lower plots at 20 and 40 weeks after inoculation. One Shreve oak, which did not show bleeding symptoms, was sampled at 20 weeks. Cankers were exposed by removing outer bark around the inoculation point with a draw knife. Cankers developed at all sampled P. ramorum inoculation points, but varied widely in size, extending from a few to about 30 cm from the inoculation point (fig. 3). Trees in the lower plot were more likely to have small cankers. We re-isolated P. ramorum from all sampled cankers at both sampling times, with no clear trends relative to plot or sampling depth in the cankers. However, the pathogen was recovered at a higher efficiency from Shreve oak than from canyon live oak.

Using symptom information gleaned from inoculation studies, we have successfully isolated P. ramorum from several naturally infected canyon live oaks, but isolation efficiency has remained very low. Successful isolations came from relatively recent cankers that were identified by small amounts of bleeding. However, our results suggest that many infected trees do not bleed and cankers in these cryptically infected trees cannot be seen until they are colonized by secondary organisms. This has complicated ongoing management studies to prevent P. ramorum infection in canyon live oak. Our results also show that bole cankers caused by P. ramorum or other Phytophthora species in hosts that develop little or no bleeding may be very difficult to detect and could remain unrecognized or attributed to other causes.

Figure 2—Percent of inoculated canyon live oak with recent bleeding symptoms at various times after inoculation with Phytophthora ramorum.

Figure 3—Percent positive Phytophthora ramorum isolations from cankers of inoculated trees 20 and 40 weeks after inoculation (Qc = canyon live oak; Qps = Shreve oak).
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Literature Cited


