Phytophthora ramorum canker (sudden oak death) in coast live oak and tanoak, 2000-2005: factors affecting disease risk, disease progression, and failure potential

2005-2006 Contract Year Annual Report

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SUMMARY

Key words: disease progression, survival, tree failure, regeneration

This report discusses findings after six years of observations in a case-control study examining the role of tree and site factors on the development of *Phytophthora ramorum* stem canker (sudden oak death) in coast live oak (*Quercus agrifolia*) and tanoak (*Lithocarpus densiflorus*). In September of each year from 2000 through 2005, we collected data on *P. ramorum* symptoms, tree condition, midday stem water potential (SWP), and various other factors in 150 circular plots (8 m radius). Each plot was centered around a case (symptomatic) or control (asymptomatic) plot center tree. Plots were located at 12 locations in the California counties of Marin, Sonoma, and Napa in areas where *P. ramorum* canker was prevalent in 2000.

Between September 2000 and September 2005, the percentage of coast live oaks with *P. ramorum* canker symptoms increased slightly, from 23% to 26%. Most of this increase occurred between September 2004 and September 2005 and was due to a large increase in newly-symptomatic coast live oaks at one location. Between 2000 and 2005, tanoaks showed a significantly larger increase in disease incidence, from 31% to 45%.

Most of the *P. ramorum*-related mortality that has occurred since 2000 was seen in trees that had *P. ramorum* canker symptoms in 2000; 58% of the tanoaks and 90% of the coast live oaks killed by *P. ramorum* between 2001 and 2005 were symptomatic in 2000. Overall, symptomatic tanoaks tend to survive for a shorter time after the onset of symptoms than do coast live oaks. Among live trees that had *P. ramorum* canker symptoms in 2000, 73% of coast live oaks but only 50% of tanoaks were still alive in 2005.

Mortality of tanoak and coast live oak has increased in a linear manner between September 2000 and September 2005. Assuming that this linear trend existed prior to 2000, initial mortality was estimated to occur in tanoak plots around 1995 and in coast live oak plots around 1996.

Most recent coast live oak failures in oak woodlands affected by *P. ramorum* have occurred in trees infected by *P. ramorum.* The number of initial failures has been declining over several years following a peak that occurred in the 2001-2002 observation interval. Since about 2003, the cumulative failure rate in *P. ramorum*-infected coast live oaks has been approximately equal to the cumulative mortality rate due to *P. ramorum*. In 2005, the cumulative rates of both mortality and failure were about 10%.

SWP of coast live oak trees continued to show a pattern of change from year to year which is closely tied to annual rainfall. Average SWP at all locations in September 2005 was at or near minimum levels (indicating relatively low water stress) observed over the study; the 2004-2005 rainfall total was also the highest recorded over this period.

Data for the period from September 2000 to September 2005 suggest *P. ramorum* has not impacted seedling populations of either coast live oak or tanoak. Seedling populations in coast live oak plots have varied widely between and within locations over time. Despite fluctuations during the intervening years, seedling densities in 2005 do not differ significantly from those observed in 2000. Tanoak seedling densities have not fluctuated as widely as coast live oak seedling numbers. Counts of tanoak seedlings in plots also showed no significant change between 2000 and 2005. In 2005, all tanoak plots that had *P. ramorum*-related mortality had tanoak seedlings present. In contrast, 8% of all coast live oak plots had *P. ramorum*-related mortality but lacked coast live oak seedlings.

INTRODUCTION

Phytophthora ramorum, the causal agent of sudden oak death (SOD), causes bleeding bark cankers on the main stems of tanoak (*Lithocarpus densiflorus*), coast live oak (*Quercus agrifolia*), and California black oak (*Q. kelloggii*) (Garbelotto and others 2001, Rizzo and others 2002). The bark cankers can expand over time and eventually girdle susceptible trees (Rizzo and others 2002). The sapwood-decaying fungus *Hypoxylon thouarsianum*, ambrosia beetles (*Monarthrum* spp.), and oak bark beetles (*Pseudopityophthorus* spp.) are commonly associated with *P. ramorum*-infected trees in later stages of decline (Garbelotto and others 2001).

We initiated a long-term study to follow disease progress and evaluate disease risk factors in the summer of 2000, shortly after *P. ramorum* (then unnamed) was identified as the cause of SOD. Most of the trees in the study are coast live oaks, but we collected parallel data on tanoaks at two locations for comparative purposes. We used a case-control study design to test whether various tree factors and plot/stand factors were related to the development of *P. ramorum* bole cankers in coast live oaks in areas where the disease was common.

Models based on results from the first three years of this project (Swiecki and Bernhardt 2001, 2002ab) were the first to document that California bay (*Umbellularia californica*) cover and density near coast live oak are significantly correlated with disease risk. Other variables that are positive predictors of disease risk in coast live oak include high canopy dominance (tree canopy sky exposure), low levels of water stress (as assessed by measuring stem water potential [SWP]), larger stem diameter, multiple main stems, and the absence of tree decline associated with other disease agents. Based on the effects of these variables in disease risk models, we inferred that trees with faster growth rates (associated with larger diameter, higher SWP, greater sky exposure, lack of decline from other agents) had an elevated risk of developing *P. ramorum* canker. More recently (Swiecki and Bernhardt 2005b), we showed that disease risk in coast live oak also increases with bark thickness and the amount of bark fissures that show evidence of recent expansion. These risk factors add further support to the concept that fast-growing trees are more likely to develop *P. ramorum* canker.

In this report, we have updated our observations on disease progress, mortality, and failure in trees affected by *P. ramorum* canker. Long-term observations of these trees have allowed us to obtain a clearer picture of the length of time that elapses between the onset of symptoms and tree mortality. We have also for the first time summarized and analyzed data on regeneration of tanoak and coast live oak that we began collecting in 2000.

METHODS

Study site selection

During September 2000, we established plots at 12 study locations (Table 1, Figure 1). Study sites were selected on the basis of appropriate vegetation type (adequate representation of coast live oak or tanoak), the presence of cases (trees with symptoms of *P. ramorum* canker) and controls (asymptomatic trees) in the study area, and absence of recent disturbances that might affect tree health (e.g., root-damaging construction). Plots were established in areas where *P. ramorum* had been shown to be prevalent. Coast live oak was the subject host species at 10 of the 12 locations; tanoak was the subject species at the remaining two locations.

Table 1. Locations of plots and host species studied.

Location number	Location	County	Approximate latitude and longitude	Number of plots	Subject tree species
1	Marin Municipal Water District (MMWD) watershed - Azalea Hill area	Marin	37.9723 N 122.6274 W	12	coast live oak
2	MMWD-Pumpkin Ridge south	Marin	37.9527 N 122.5949 W	16	coast live oak
3	MMWD-Pumpkin Ridge north	Marin	37.9599 N 122.5989 W	11	coast live oak
4	MMWD-Phoenix Lake area	Marin	37.9590 N 122.5770 W	11	coast live oak
5	China Camp SP - Miwok Meadows area	Marin	38.0044 N 122.4848 W	16	coast live oak
6	China Camp SP - SE Buckeye Point area	Marin	38.0044 N 122.4768W	12	coast live oak
7	Woodacre (Private land)	Marin	38.0175 N 122.6472 W	12	coast live oak
8	Lucas Valley (Private land)	Marin	38.0432 N 122.5996 W	12	coast live oak
9	Muir Woods NM / Mt. Tamalpias SP	Marin	37.9024 N 122.5839 W	10	tanoak
10	Wall Road (Private land)	Napa	38.4092 N 122.4751 W	13	coast live oak
11	Novato (Private land)[1]	Marin	38.0988 N 122.6273 W	13	coast live oak
12	Jack London SP	Sonoma	38.3450 N 122.5616 W	12	tanoak

[1] This site was previously listed as being on Marin County Open Space District land.

Plot selection

At each study location, we established 10 to 16 circular 8 m radius (0.02 ha) fixed-area plots, each of which was centered at a subject tree. The number of plots per location was limited by the time constraints associated with making stem water potential measurements. After determining that symptomatic trees (cases) were present in adequate numbers in the stand, we established a random starting point and searched for the nearest case or control tree starting from that point. This tree became the first subject tree and the center of the first plot. Subsequent tree-centered plots were spaced approximately 20-30 m apart. Actual interplot spacing varied with vegetation and terrain, but to avoid overlap between plots, no two adjacent plots were located closer than 16 m apart. We attempted to alternate case and control plots, but if the designated subject tree type (e.g., control) did not exist within a 4-8 m search radius of the target point, the other subject tree type was selected. Potential cases and controls were rejected if they did not have foliage low enough to be accessed for water potential measurements. In general, we attempted to distribute the plots across a range of topographic positions, slopes, and aspects.

We marked the center subject tree in each plot with a numbered aluminum tree tag. Tags were placed at varying heights, but generally point toward the next successive plot. To help relocate plot center trees within each study site, we recorded the distance and azimuth readings between plots. We subsequently determined the coordinates of the plots at each location using a GPS receiver with an external, mast-mounted antenna, although the position of some plots at location 9 could not be determined with GPS due to poor satellite reception.

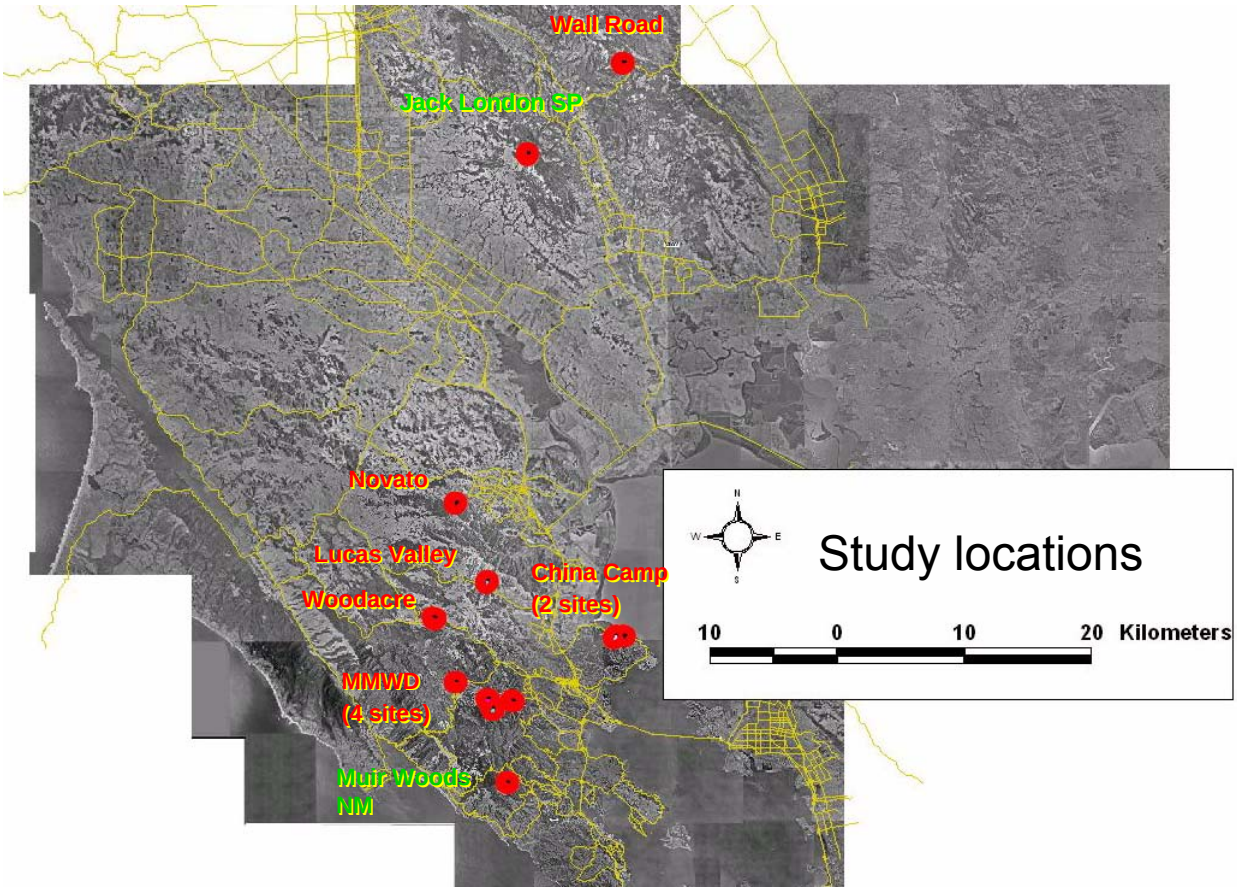

Figure 1. Map showing locations of study areas in Marin, Napa, and Sonoma counties. Background image is a mosaic of USGS digital aerial orthophotos.

Stem water potential measurements

In September of each year from 2000 through 2004, we collected midday stem water potential (SWP) readings on the center subject tree in each plot during the peak midday period (about 1300-1530 PDT). In addition, from 2001 though 2004, we also took SWP readings on additional trees in 45 of the plots (one additional tree per plot except one plot with two additional trees) for comparative purposes. In September 2005, SWP monitoring was reduced to include only three plot center trees at each of the ten coast live oak locations. The selected trees were spread out within each location area. To allow for continued annual measurements on the same trees, we selected trees that had an adequate supply of leaves within reach which could be used for future SWP measurements and avoided trees that appeared likely to die within the next few years.

SWP measurements were made following methods outlined by Shackel (2000). On each tree, we selected a minimum of two shoot tips with several leaves for measurement. We selected shoots and leaves that branched directly off the trunk or from main branches near the trunk, or from basal sprouts (primarily for tanoak). Each shoot tip was sealed in a clear plastic bag and overbagged with a larger opaque reflective plastic bag. These bags prevent the leaves from transpiring and overheating. Bags were left in place for 2 or more hours to allow leaf water potential to equilibrate to that of the subtending stem. At the time of the reading, the outer

opaque bag was removed and the shoot tip was excised and placed into the pressure chamber while still sealed in the inner plastic bag. Two SWP readings were made on most trees. In general, two valid SWP measurements from a single tree were within 0.05 to 0.1 MPa of each other. SWP measurements were made with a pump-up pressure chamber (PMS Instrument Co., Corvallis OR) fitted with a 10.2 cm diameter 40 bar (0.4 MPa) gauge with 1% accuracy full scale. Other methods associated with SWP readings have been described previously (Swiecki and Bernhardt 2005b).

Additional tree and plot variables

Plot center trees and the 47 extra SWP trees were rated for origin class (seed or coppice); stem count; DBH; and amount of crown exposure to overhead sunlight, and other factors (Table 2). Plot variables recorded (Table 3) included plot slope and aspect; total basal area; tree counts by species; plot canopy cover; woody understory cover; cover of selected tree species and poison oak (*Toxicodendron diversilobum*). We also recorded the disease status of all other coast live oak, California black oak, and tanoak trees in the plot with respect to *P. ramorum* and other pathogens, and counts of regeneration of these three species. Coast live oak, black oak, and tanoak trees other than the plot center tree are collectively referred to as plot trees in this paper.

In 2003, we collected data on physical characteristics of the bark of coast live oak plot center and extra SWP trees at all locations except 9 and 12, which did not include coast live oaks (Swiecki and Bernhardt 2004). We used a variety of descriptors to rate bark morphology of the lower bole. Bark characteristics rated included the abundance (none, trace, low-moderate, moderate-dense) and location of epiphytic lichens and mosses; various surface bark morphologies (striate, checkered, smooth, furrowed, irregular); the presence of shallow, medium, or deep bark fissures; the presence of deep bark cracks; and the presence of unweathered, brown bark in the center of furrows or fissures. In the 2004 evaluations, we rated the relative abundance of this last characteristic using the 0-6 scale described below. This variable was reassessed in 2005.

We used the following arcsine-transformed percentage scale for most ocular estimates of percentages: 0 = not seen, 1= less than 2.5%, 2 = 2.5% to 19%, 3 = 20% to 49%, 4 = 50% to 79%, 5 = 80% to 97.4%, 6 = more than 97.5%.

Statistical analyses

We used JMP® statistical software (SAS Inc., Cary NC) for data analysis. Unless otherwise indicated, effects or differences are referred to as significant if $p \leq 0.05$. Repeated measures multivariate analysis of variance (MANOVA) was used to examine the effects of factors on outcomes that have been repeatedly assessed in plots. The likelihood ratio chi square statistic was used to test the significance of difference of proportions in 2×2 contingency tables. We also used the Tukey-Kramer HSD for mean separation following a significant F level in a one-way analysis of variance.

Table 2. Tree variables measured for plot center trees, other plot trees, and selected out of plot trees.

Variable	Trees rated[1]	Year(s) evaluated[2]	Method	Scale/units and notes
General tree descriptors				
Tree species	C,A,P: O:	2000 2002		*Q. agrifolia, L. densiflorus or Q. kelloggii* (plot trees only)
Origin class	C,A: O:	2000 2002	visual assessment	seed (0) or sprout (1)
Distance to plot center	A: P,O:	2001 2002	laser rangefinder	m; recorded for plot trees in 2002
Azimuth to plot center	A: P,O:	2001 2002	compass	degrees; recorded for plot trees in 2002
DBH	C: A: P,O:	2000 2001 2002	flat tape measure	cm
Sky-exposed canopy	C: A: P,O:	2000 2001 2002	visual estimate	pretransformed 0-6 scale[3]; percent of canopy projection area with unobstructed access to direct overhead sunlight
Number of stems from ground	C: A: P,O:	2000 2001 2002	count	stems/tree
P. ramorum canker-related symptoms				
Phytophthora-related symptoms	C,A,P: O:	2000-on 2002-on	visually assess symptoms present	(0) No symptoms (1) Early - bleeding cankers only (2) Late - cankers plus beetle boring and/or *H. thouarsianum* (3) Dead as result of *Phytophthora* infection; evidence of bark cankers present
Recent bleeding from cankers	C: A: P:	2000-on 2001-on 2002-on	visual assessment of exudate	Present (1) scored if bleeding appeared to have occurred within the previous 4-6 months / otherwise absent (0)
Phytophthora canker count	C: A:	2000-on 2001-on	count	Estimated on basis of external bleeding spots and limited inspection of canker margins. In 2000, only an overall count for all stems was made. In 2001, counts per stem for multistemmed trees were also made.
Percent girdling due to *Phytophthora* cankers	C,A:	2000-on	visual estimate	pretransformed 0-6 scale[2] Percent of circumference affected estimated based on projection of cankered areas as if all were viewed on same cross section; some limited chipping of bark done to confirm horizontal extent of canker margins in some trees. In general, girdling ratings are difficult and less reliable on completely dead trees. In 2000, a single overall rating was made for all stems. Starting in 2001, individual ratings were also made for each stem of multistemmed trees.
Height of upper and lower *P. ramorum* canker margins above grade	C,A:	2003	tape measure	Height (cm) above soil level was noted for the upper edge of the highest canker and lower edge of the lowest canker on symptomatic trees.
Stems with Phytophthora symptoms	C,A,P,O:	2000-on	count	infected stems/tree
Dead stems	C,A,O:	2000-on	count	dead main stems/tree and likely cause of stem death (Phytophthora canker or other)

Table 2. Tree variables measured for subject trees, other plot trees, and selected out of plot trees. (continued)

Variable	Trees rated[1]	Year(s) evaluated[2]	Method	Scale/units and notes
P. ramorum canker-related symptoms (continued)				
Tree dead / cause	C,A,P,O:	2000-on	visual assessment	Causes: (0) not dead (1) Phytophthora canker; (2) other agent(s); (3) unable to determine (4) Phytophthora canker plus other agent(s) Tree scored as dead if all main stems are dead, even if small live basal sprouts are present.
Hypoxylon thouarsianum Percent girdling	C: A: P,O:	2000-on 2001-on 2002-on	Visual estimate based on presence of fruiting bodies	pretransformed 0-6 scale[2] Percent of circumference affected estimated based on projection of cankered areas as if all were viewed on same cross section;
Hypoxylon thouarsianum Highest density in 0.1 x 1 m vertical strip	C,A,O:	2002	count	Count of fruiting bodies. Individual lobes counted separately.
Wood boring beetles in main stem	C,A,P,O:	2000-on	Shape and size of exit holes	Type of beetle based on shape of exit holes
Abundance of bark and/or ambrosia beetles in main stem	C: A: P,O:	2000-on 2001-on 2002-on	presence of boring dust and/or holes	(0) none seen (1) low (2) moderate (3) high
Other tree condition variables				
Canopy thinning	C: A: O:	2000-on 2001-on 2002	visual estimate	0-2 Scale: (0) none; (1) slight; (2) pronounced
Canopy dieback	C: A: P: O:	2000-on 2001-on 2002-on 2002	visual estimate	pretransformed 0-6 scale[3] Based on percent dead crown volume
Severe tree decline due to other agents	C,A,P: O:	2000-on 2002	visual assessment	yes (1)/ no (0) Trees scored in decline if overall condition was poor enough that death within 10 years was judged to be likely.
Decay impact	C: A: O:	2000-on 2001-on 2002	visual assessment	0-3 Scale: (0) none; (1) low; (2) moderate: (3) high Decay impact rating (Swiecki and Bernhardt 2001a) assesses the probability that existing decay will have a significant negative impact on tree health or survival. Assessment of decay impact was based on the type(s) of decay present, location of decay within the tree, and the estimated extent of decay as rated by a trained observer. Levels were recoded to three classes as follows for some analyses: (1) none; (2) low or moderate; (3) high
Status change	C,A,P:	2000-on	comparison of 2000 and 2001 data	Evaluation based primarily on canker extent, colonization by secondary organisms, and dieback. (0) no change; (1) improved condition; (-1) degraded condition
Epicormics	C: A: O:	2000-on 2001-on 2002	visual assessment	0-2 Scale: (0) none; (1) few; (2) numerous

Table 2. Tree variables measured for subject trees, other plot trees, and selected out of plot trees. (continued)

Variable	Trees rated[1]	Year(s) evaluated[2]	Method	Scale/units and notes
Other tree condition variables (continued)				
Live basal sprouts	C,A,P:	2000-on	visual observation	presence (1) / absence (0) scored for dead trees only Trees are scored as dead if all main stems are dead even if some live basal sprouts are present.
Other agents and symptoms	C,A,P,O:	2000-on	visual observation	Presence of wood decay fungi fruiting bodies and canker rot or root rot symptoms were noted.
Defect codes	C,A: P: (if failed)	2002-on 2002-on	visual observation	The presence of various structural defects that may contribute to the risk of tree failure were coded. (1) Dead branch or branch stubs (2) Multiple trunks/ codominant stems (3) Hollow branch stubs (4) Dense crown (5) Heavy lateral limbs/ excessive branch end weight (6) Uneven branch distribution: one sided (7) Uneven branch distribution: top heavy (8) Multiple branches from same point (9) Embedded bark in crotch (10) Crook or sweep (11) Leaning trunk (12) Cracks or splits (13) Kinked or girdling roots (14) Cavity (15) Decay column
Tree failure	C,A,P:	2000-on		Failures of bole or branches >20 cm diam noted if present
Failure type	C,A,P:	2001-on		(1) Root (2) Root crown (lower edge of fracture was near soil surface) (3) Bole (main stem) (4) Scaffold (lowest first order branches arising from bole) (5) Branch (all other branches)
Tree condition at time of failure	C,A,P:	2001-on	based on condition of twigs and foliage	(1) Live (2) Dead (3) Uncertain
Estimated failure date	C,A,P:	2001-on	based on weathering of failed surface, degradation of failed part, previous observations, etc.	(1) within previous 6 months (2) 6-12 months prior to rating More precise dates were estimated if supportable by observations (e.g., green foliage on failed part)

Table 2. Tree variables measured for subject trees, other plot trees, and selected out of plot trees. (continued)

Variable	Trees rated[1]	Year(s) evaluated[2]	Method	Scale/units and notes
Bark thickness	C (dead), A,O: C (live):	2003 2004	bark probe	mm
Brown bark from recent bark expansion in fissures	C,A,P:	2003 2004 2005	visual assessment	2003: present/absent 2004: pretransformed 0-6 scale[3] – Percent of cumulative fissure length in lower 2 m of bole showing brown color
Lichen abundance (lower 2m of bole)	C,A:	2003	visual ranking of lichen cover	(0) none; (0.5) trace; (1) low; (2) moderate to high
Moss abundance (lower 2m of bole)	C,A:	2003	visual estimate of moss cover	(0) none; (0.5) trace; (1) low; (2) moderate to high
Moss location	C,A:	2003	visual assessment	(1) basal only (lower 1-2 m of bole) (2) extending up bole into upper bole and/or canopy
Type of bark fissures present	C,A:	2003	visual assessment	(1)shallow; (2) medium; (3) deep
Deep bark cracks	C,A:	2003	visual assessment	present/absent (Unlike fissures, cracks are abrupt discontinuities that extend deep into the bark or to the cambium that are not associated with normal growth patterns.)
Bark texture	C,A:	2003	visual description	bark texture was described using one or more of the following characteristics: smooth, irregular, striate, checkered, corky, furrowed

[1]Tree types: C=plot center tree; A=additional trees used for stem water potential readings starting in 2001; P=other plot trees; O= trees located beyond plot edges used for coring in 2002 (Swiecki and Bernhardt 2003a) and bark probe measurements in 2003. Only asymptomatic trees beyond plots were chosen for coring in 2002.

[2]Variables scored in a single year were reevaluated only for trees which showed a change from the original values.

[3]The 0-6 scale is based on the following arcsine-transformed percentage scale:

(0) Symptom not seen
(1) < 2.5%
(2) 2.5% to <20%
(3) 20% to < 50%
(4) 50% to < 80%
(5) 80% to < 97.5%
(6) 97.5% to 100%

Table 3. Plot and stand variables measured in study plots. Except as noted, all variables were measured in the 8 m radius fixed-area plots.

Variable	Year(s) evaluated[1]	Method	Scale/units and notes
Tree density / species composition	2000	count by species	Trees have at least one stem at least 3 cm DBH located within 8 m of plot center; multi-stemmed trees count as single trees; coppiced redwoods separated by at least 1 m count as separate trees
Plot slope	2000	clinometer	percent
Plot aspect	2000	compass	degrees
Plot drainage	2000	visual observation	none; creek/drainage with surface water; dry creek or drainage
Plot drainage proximity	2000	visual observation	0 if in plot; otherwise estimate meters from plot edge
Plot tree canopy cover	2000	visual estimate	pretransformed 0-6 scale[2]; overall tree cover in plot
California bay cover	2002	visual estimate	pretransformed 0-6 scale[2]; bay cover in plot, including regeneration
Madrone cover	2002	visual estimate	pretransformed 0-6 scale[2]; madrone cover in plot, including regeneration
Woody understory cover	2000	visual estimate	pretransformed 0-6 scale[2]; includes both shrubs and small (<3 cm DBH) tree regeneration
Plot shrub cover	2001	visual estimate	pretransformed 0-6 scale[2]
Poison oak cover	2002	visual estimate	pretransformed 0-6 scale[2]
Overstory canopy trees species in plot	2001	visual assessment	list of species Overstory canopy trees do not have to be rooted within the plot.
Count by general tree health class (trees other than SOD hosts[3])	2000, 2001	tree count by species, subcategorized by symptom class and canopy position (overstory/understory)	Symptom classes: (1) live (2) decline (3) dead
SOD host[3] regeneration	2000-on	count or estimate if >10	regeneration = seedlings and saplings <3 cm dbh
Disease incidence in SOD host[3] regeneration	2000-on	count or estimate percent if count > 10	Disease may be due to *P. ramorum* and/or other agents or factors
Dead SOD host[3] regeneration	2000-on	count	Cause of mortality in regeneration was not determined
Regeneration of trees other than SOD hosts[3]	2000	presence noted by species	regeneration: seedlings and saplings <3 cm dbh
Other pathogens/agents	2000-on	note presence	listing of agents and symptoms observed, including various decay fungi, canker rot, root disease, *H. thouarsianum*, and beetles
Woody understory species	2001	note presence	list shrubs and woody vines present within plot; herbaceous species and grasses were not scored
Disturbance	2000	Note type of disturbance	roads, trails, logging, etc. within plot or near edge of plot were noted
Oak/tanoak failure in plot	2001	count	Bole and large limb failures (>20 cm diam) observed in the plot were noted.
Basal area[4]	2000	survey laser reticle	reticle BAF = 5 m^2/ha

[1]Variables scored in a single year were reevaluated only for trees which showed a change from the original values.

[2]The 0-6 scale is based on the following arcsine-transformed percentage scale:

0: Symptom not seen
1:< 2.5%
2: 2.5% to <20%
3: 20% to < 50%
4: 50% to < 80%
5: 80% to < 97.5%
6: 97.5% to 100%

[3]SOD hosts = hosts of *P. ramorum* stem canker, i.e., coast live oak, black oak, and tanoak

[4]Basal area measurements were made on a variable-radius plot centered at the plot center tree.

RESULTS

Symptom development and disease progress 2000-2005

Overall disease incidence and mortality

Baseline levels of disease in plots were established when the plots were originally evaluated in September 2000. Additional newly symptomatic trees have been noted each subsequent annual evaluation. For tanoak, the greatest numbers of trees with apparently new infections were seen in 2001 and 2004 (Figure 2). In contrast, coast live oak showed the greatest increases in newly symptomatic trees in 2003 and 2005 (Figure 2). These differences may reflect different levels of inoculum produced in these forest types in different years and/or differences in the latent period for the two species.

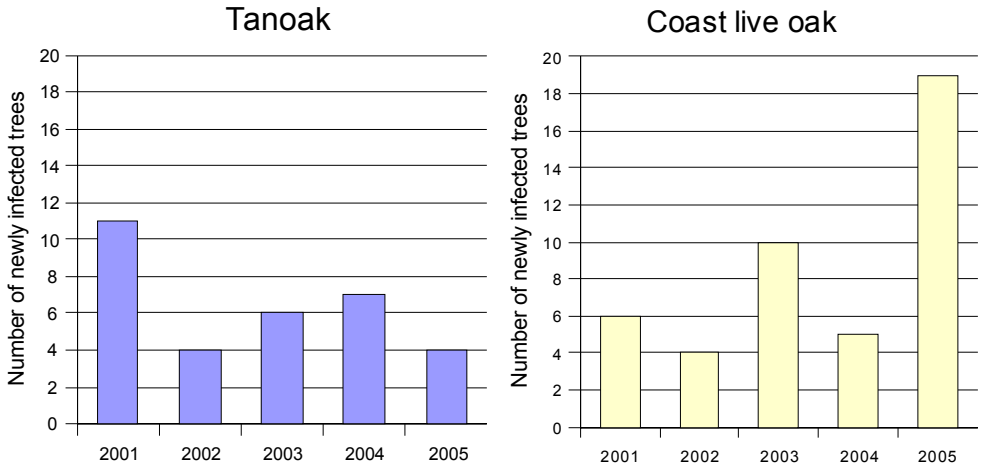

Figure 2. Number of tanoak and coast live oak trees that first displayed *P. ramorum* canker symptoms in September of the years shown in the graph. Total number of live asymptomatic trees present in 2000 was 130 for tanoak, 470 for coast live oak.

Although newly symptomatic trees have been observed among both tanoaks and coast live oaks between 2000 and 2005, only tanoaks show a relatively steady net increase in disease incidence over this period (Figure 3). The overall *P. ramorum* canker incidence among tanoaks has increased from 31% in 2000 to 45% in 2005 (Figure 3). Overall *P. ramorum* canker incidence among coast live oaks has increased only from 23% in 2000 to 26% in 2005 (Figure 3). The increase in disease incidence is significantly higher among tanoaks than coast live oaks (likelihood ratio $p<0.0001$).

In some coast live oaks, symptoms became less obvious over time and appeared to go into remission. The original canker can no longer be detected in some of these trees. Therefore, some coast live oaks previously classified as showing early symptoms of infection (bleeding cankers only) have been reclassified as asymptomatic because the cankers have become inactive and indistinct in succeeding years. Although a total of 44 coast live trees have become newly symptomatic over the course of the study, reclassification of 26 previously symptomatic trees to asymptomatic has resulted in a net increase of only 18 symptomatic trees between 2000 and 2005.

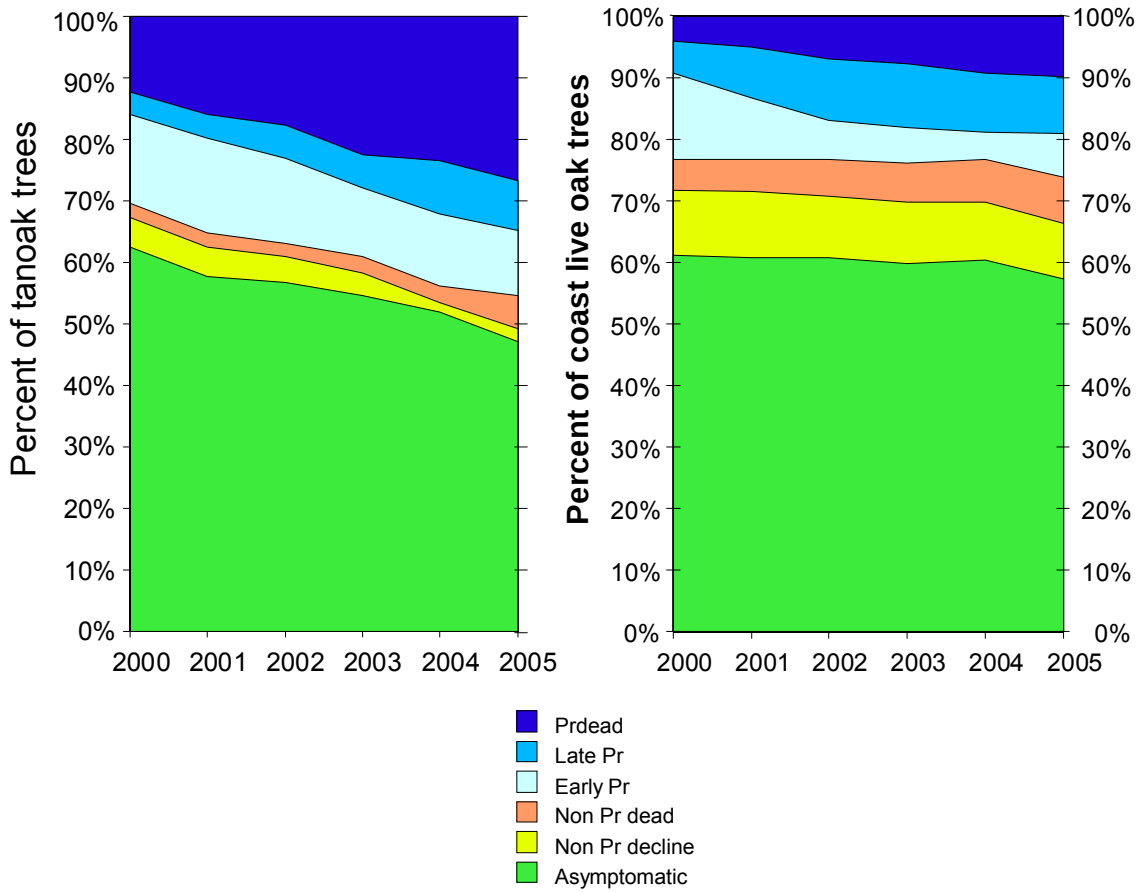

Figure 3. Changes in health of all tanoak (n=187) and coast live oak (n=655) study trees from September 2000 to September 2005. **Dead Pr** = tree dead as a result of *P. ramorum;* **Late Pr** = live trees with *P. ramorum* cankers plus beetle boring and /or *H. thouarsianum* fruiting bodies; **Early Pr** = live trees with *P. ramorum* cankers only; **Other dead** = tree dead due to agents other than *P. ramorum;* **Other decline**=tree in severe decline due to agents other than *P. ramorum;* **Asymptomatic**= no evident symptoms of *P. ramorum* infection or decline due to other agents.

The overall percentage of the study tree population killed by *P. ramorum* is also significantly higher for tanoak than coast live oak (likelihood ratio $p<0.0001$). This trend has been evident from the beginning of the study (Figure 3).

Percent mortality due to *P. ramorum* has increased in a relatively linear fashion over the study period for both coast live oak and tanoak (Figures 3, 4). Linear regression models for mortality assessed in September 2000 through 2005 are shown in Figure 4. Under the assumption that the linear trend in mortality applied prior to 2000, we estimate that mortality of tanoak due to *P. ramorum* began around 1995 at the two tanoak study locations. A similar extrapolation of the regression line for the ten coast live oak study locations suggests that mortality of coast live oak due to *P. ramorum* possibly began around 1996 in the study locations.

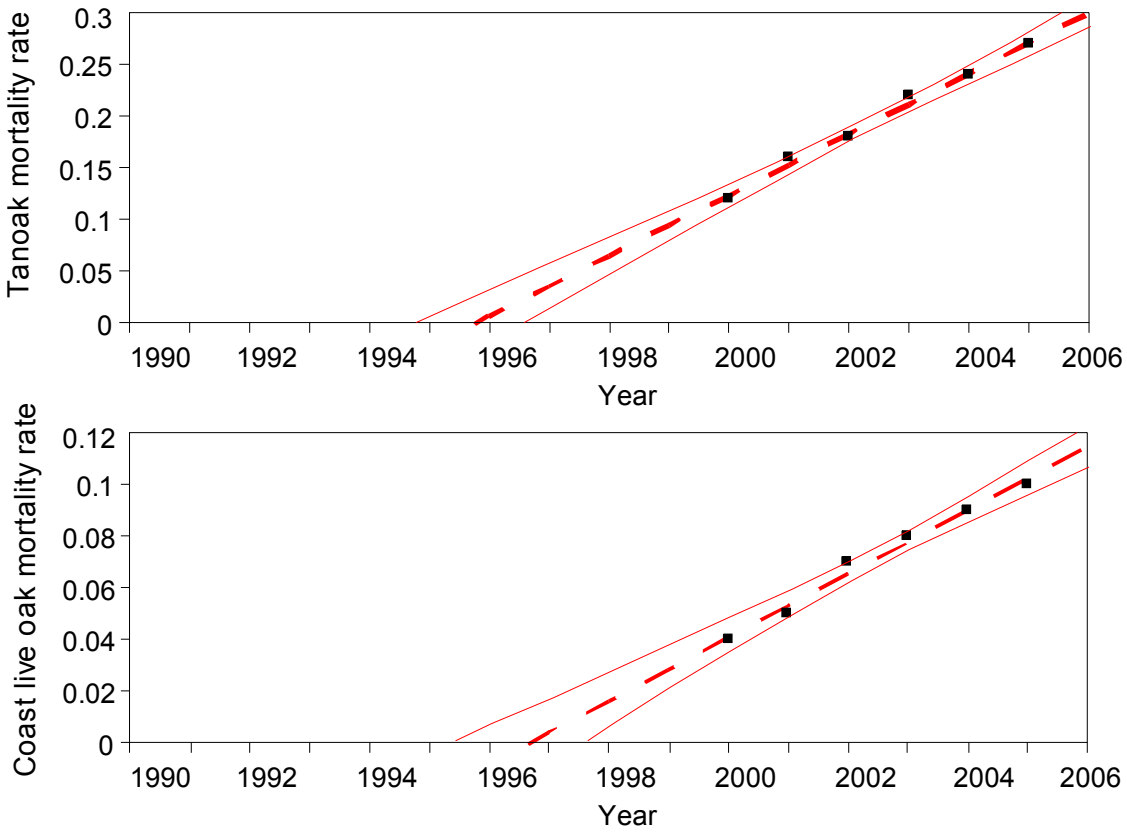

Figure 4. Extrapolated beginning date for mortality due to *P. ramorum* in tanoak (top) and coast live oak (bottom). Fitted regression lines (dashed lines) are based on data for *P. ramorum*-related mortality shown in Figure 3. Tanoak: *Prdead* = -58.73238 + 0.0294286 *Year* (R^2=0.992); Coast live oak: *Prdead* = -24.53048 + 0.0122857 *Year* (R^2=0.984). Curves (solid lines) around the regression lines indicate the 95% confidence interval.

The overall *P. ramorum* infection rate for tanoak has also been linear over the study period (Figures 3, 5). Extrapolation of the linear regression line to the x intercept suggests that *P. ramorum* canker symptoms in the study tanoak population may have been visible as early as 1989 (Figure 4). Because the net *P. ramorum* infection rate for coast live oak had not changed appreciably between 2000 and 2005 (Figure 2), we did not attempt to use a linear extrapolation of coast live oak infection levels to estimate a date for the onset of symptoms in this species.

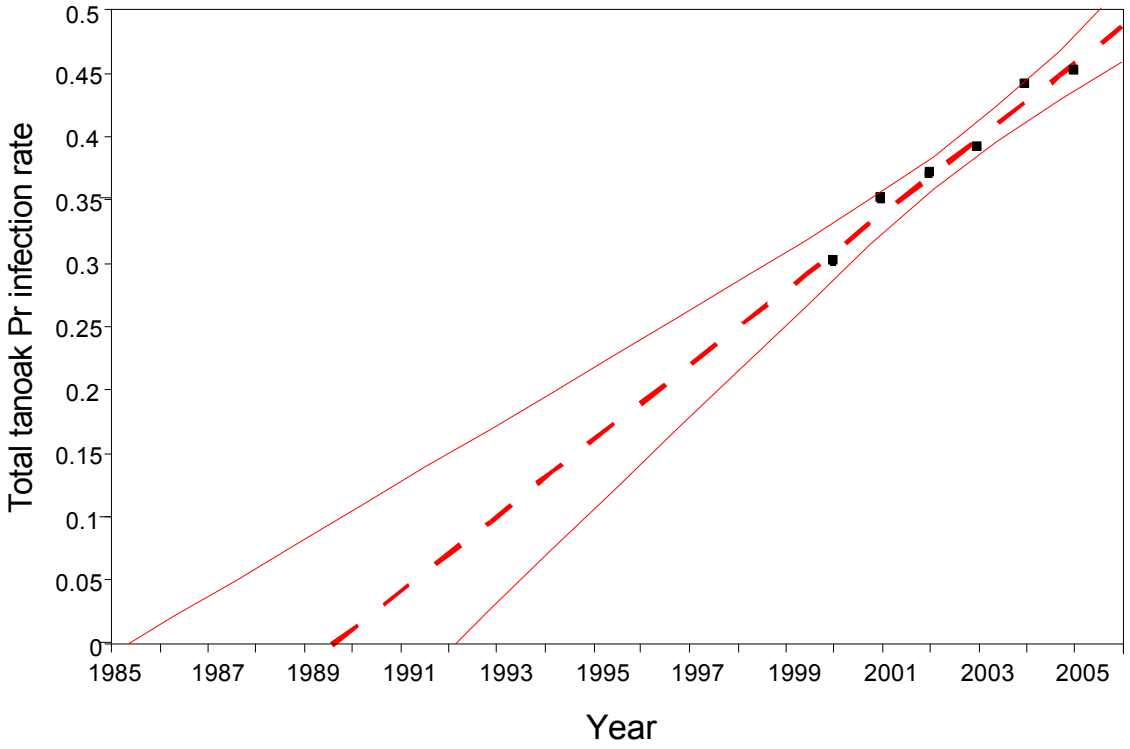

Figure 5. Extrapolated date for the first appearance of *P. ramorum* canker symptoms in tanoak based on overall tanoak *P. ramorum* infection rates from 2000 to 2005. *Overall Pr rate* = -59.11952 + 0.0297143 *Year* (R^2=0.969). Curves (solid lines) around the regression line (dashed) indicate the 95% confidence interval.

Time from symptom onset to mortality

Most of the *P. ramorum*-related mortality that has occurred since 2000 was seen in trees that had *P. ramorum* canker symptoms in 2000; 58% of the tanoaks and 90% of the coast live oaks killed by *P. ramorum* between 2001 and 2005 were symptomatic in 2000. Overall, symptomatic tanoaks tend to survive for a shorter time after the onset of symptoms than do coast live oaks. Among live trees that had *P. ramorum* canker symptoms in 2000, 73% of coast live oaks but only 50% of tanoaks were still alive in 2005 (Figure 6).

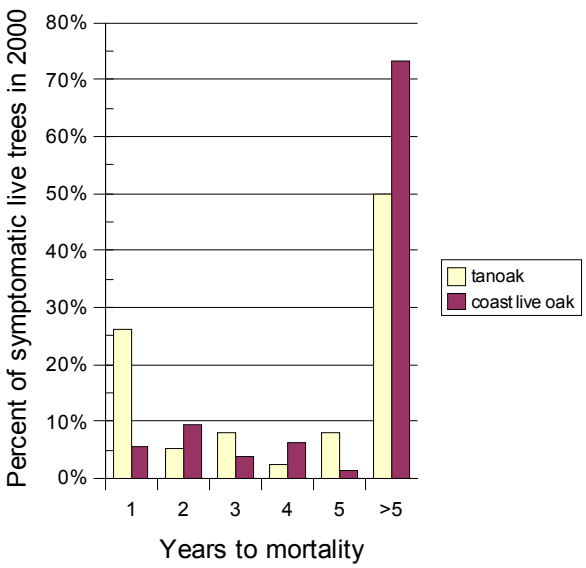

Figure 6. Years to mortality for living trees that had symptoms of *P. ramorum* infection in September 2000. The date of initial symptom development for this group of trees is unknown. N=38 for tanoak and 128 for coast live oak

The same overall trend in survival time is apparent among trees that first developed visible symptoms after 2000 (Figure 7). For tanoaks which first became symptomatic during the period from 2001-2005 (n=32), 34% died within two years after symptoms were first observed; the remaining trees have survived at least one to four years (Figure 7). Among 44 coast live oaks which first became symptomatic in 2001-2005, only 5% died within the first two years after symptoms were first observed; the remainder had not died by 2005 (Figure 7).

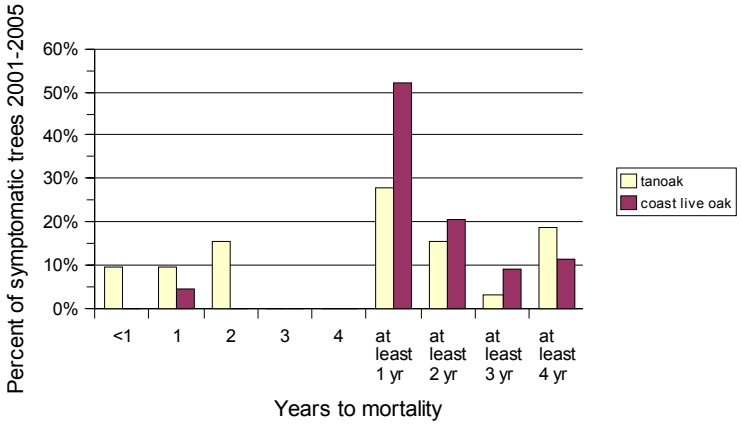

Figure 7. Years from symptom onset to mortality of trees which became symptomatic in the period 2001-2005. For surviving time-censored trees, the minimum possible survival period is noted. N = 42 for tanoak and 44 for coast live oak

Disease incidence and mortality by study location

Figure 8 shows changes in *P. ramorum* symptom status between 2000 and 2005 for trees at each of the 12 study locations. At locations 2 and 6, no asymptomatic trees developed *P.*

ramorum canker symptoms between 2000 and 2005. At all other locations, at least some trees developed *P. ramorum* canker symptoms over this period. Between 2000 and 2004, the two tanoak locations (9 and 12) had the highest percentage of trees that developed new *P. ramorum* canker symptoms. However, due to new symptoms that were observed in 2005, coast live oak location 7 now has a greater percentage of trees which have become infected with *P. ramorum* since 2000 than does tanoak location 9 (Muir Woods). Between 2000 and 2004, disease incidence at location 7 had changed little, increasing from 21% in 2000 to 22% in 2004. However, in 2005, *P. ramorum* canker incidence nearly doubled, increasing to over 40% of the canker host tree population (Figure 8).

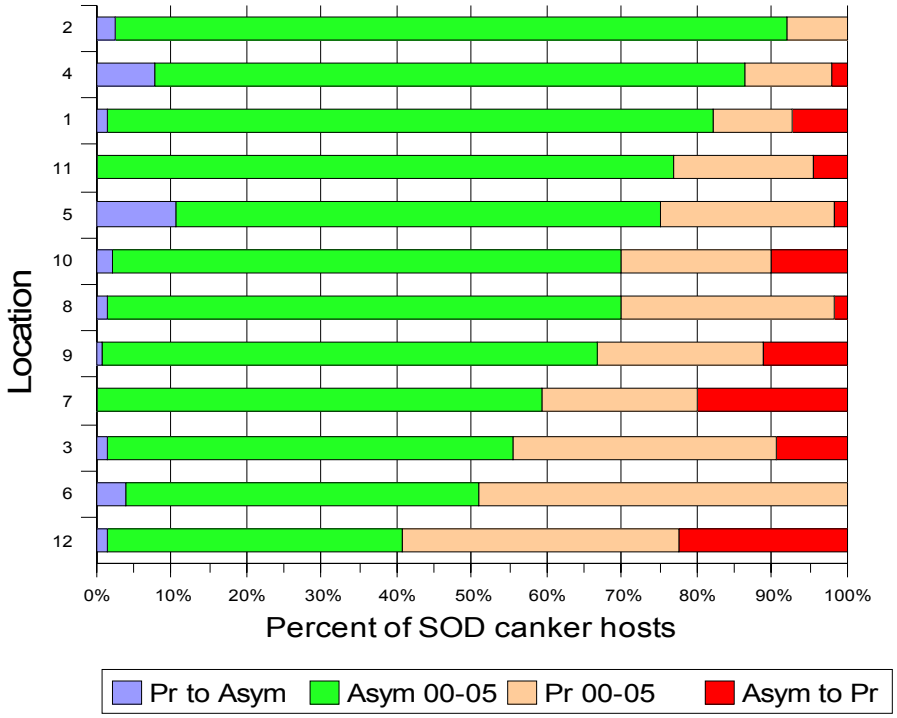

Figure 8. Percent of SOD canker hosts (coast live oak, California black oak, and tanoak) showing changes in overall *P. ramorum* canker symptom status between 2000 and 2005 ratings. Percentages include trees that were dead in 2000 or died between 2000 and 2005. **Asym to Pr**= trees which have developed *P. ramorum* canker symptoms since 2000; **Pr 00-05** = tree with *P. ramorum* canker symptoms in 2000 through 2005; **Asym 00-05**= trees without *P. ramorum* canker symptoms in 2000 through 2005 (includes trees scored as having *P. ramorum* canker symptoms only in 2000); **Pr to asym**= trees scored with *P. ramorum* canker symptoms in 2000 and at least one additional year but asymptomatic in 2005. Location numbers are shown in Table 1 and are sorted in order of increasing *P. ramorum* canker incidence in 2005.

Figure 9 shows the overall contribution of *P. ramorum* canker to mortality rates at each of the 12 study locations. Mortality rates from all causes vary greatly across the study locations. Mortality rates at the coast live oak study locations vary from a low of about 8% of the study trees at locations 2 and 4, to a high of about 30% of the study trees at locations 3, 6, and 10. The overall mortality rates at the two tanoak study locations are 21% and 45%. *P. ramorum* has been the leading cause of study tree mortality at both tanoak locations (locations 9 and 12, Figures 2, 4) and has caused the majority of tree mortality at 6 of the 10 coast live oak locations (locations

3, 4, 5, 6, 8, and 11). Wood decay fungi, particularly species of *Inonotus* and *Ganoderma*, have been important causes of tree mortality at several of the coast live oak locations.

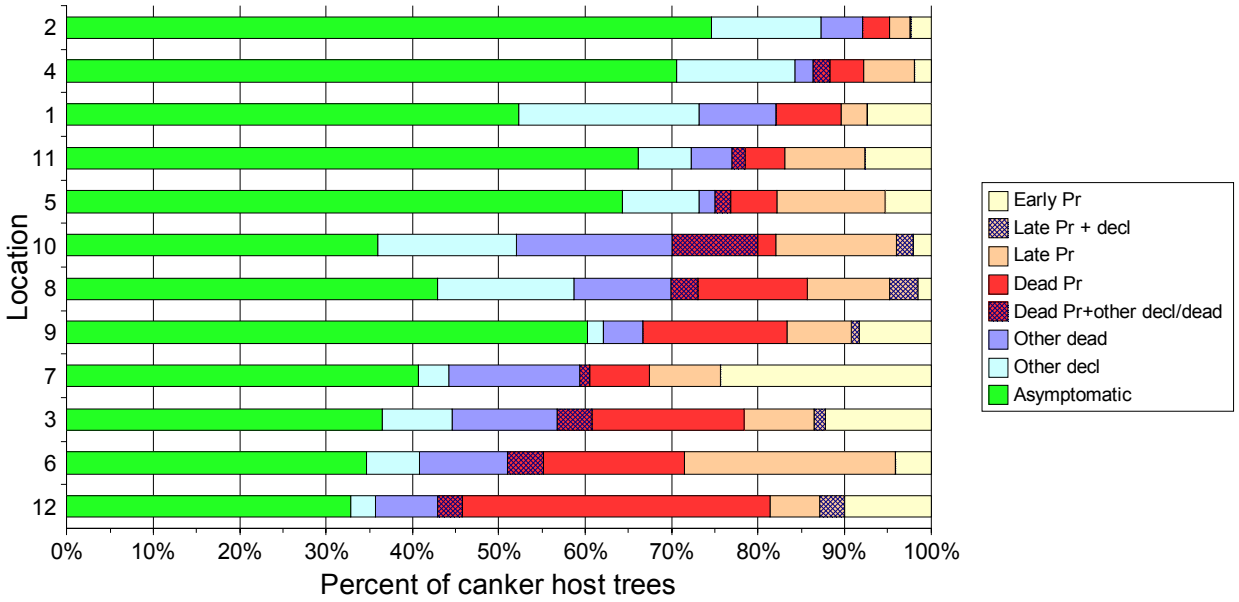

Figure 9. Overall disease status of SOD canker hosts (coast live oak, California black oak, and tanoak) in 2005. Pr = tree with *P. ramorum* canker symptoms; early = bleeding cankers only, late = bleeding cankers with beetles and/or *H. thouarsianum;* other decl/dead = tree declining or dead due to agents other than *P. ramorum*. Hatched bars indicate trees that have *P. ramorum* canker symptoms and are also declining due to other agents other than secondary agents typically associated with *P. ramorum* canker. Location numbers are shown in Table 1 and are sorted in order of increasing *P. ramorum* canker incidence. Trees with *P. ramorum* symptoms that were killed when nearby trees fell on them have been grouped into the 'other dead' category for the purposes of this graph.

Canopy dieback in coast live oak

Starting in 2000, we scored canopy dieback in the plot center trees in all plots. In 2002, we began evaluating dieback on all other coast live oak and tanoak trees in the plots as well. Although the observation interval is shorter for the full data set (plot trees plus center trees), the sample size is much greater than the center tree data set, which was used in our previous analyses of canopy dieback (Swiecki and Bernhardt 2005b).

In the full data set, trees that were either healthy or had only early symptoms of *P. ramorum* canker (bleeding cankers only) in September 2005 showed no overall change in their average levels of canopy dieback since September 2002 (Figure 10). Trees that had late symptoms of *P. ramorum* infection (cankers plus beetle boring and or wood decay fungi) or were in decline due to other factors in 2005 showed increasing levels of canopy dieback between 2002 and 2005 (Figure 10). Year, disease status, and the interaction between year and disease status were all significant at $P<0.0001$ in a repeated measures analysis of variance for the canopy dieback ratings.

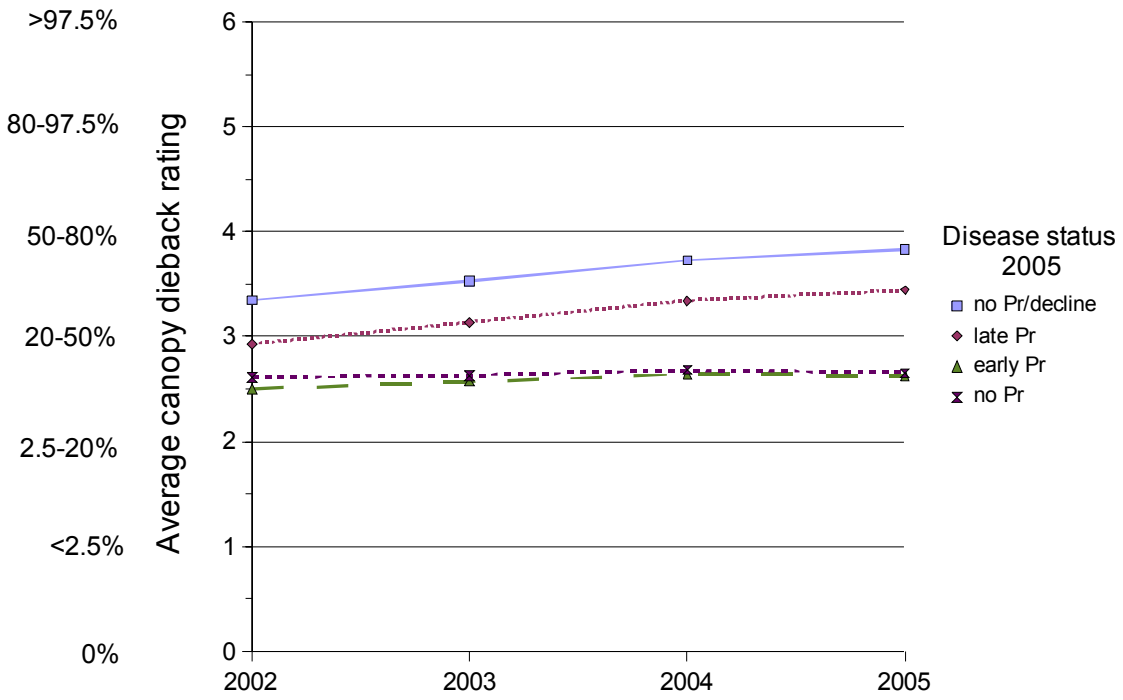

Figure 10. Canopy dieback ratings in September for coast live oak trees in each disease class as of Sept 2005; **decline/no Pr** =trees in severe decline due to agents other than *P. ramorum*, **late Pr**=live trees with *P. ramorum* cankers plus beetle boring and /or *H. thouarsianum* fruiting bodies; **early Pr**=trees with *P. ramorum* cankers only, **no Pr**=non-declining trees with no evident symptoms of *P. ramorum* infection.

Tree failure

Between 2000 and 2005, 66 of 153 coast live oaks with *P. ramorum* symptoms had initial failures above the minimum size threshold scored in this study (branch failures ≥20 cm diameter; and bole, root or root crown failures of trees greater than ≥ 3 cm DBH). This 36% failure rate in *P. ramorum*-infected trees was significantly greater (likelihood ratio $p<0.0001$) than the 4% failure rate seen over this period among 484 coast live oaks without *P. ramorum* symptoms. Bole and branch failures were the most common type of failures in trees with *P. ramorum* canker symptoms (Figure 11). No root failures occurred among trees with *P. ramorum* symptoms over this period.

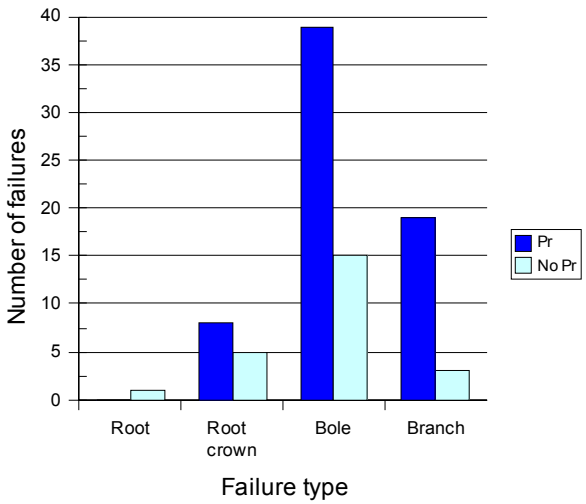

Figure 11. Number and location of failure for initial failures above threshold size (20 cm diameter at break for branch failures; any main stem ≥ 3 cm DBH for bole, root, and root crown failures) occurring between September 2000 and September 2005 among coast live oaks with (**Pr**) and without (**No Pr**) symptoms of *P. ramorum* canker at the time of failure.

Based on characteristics of the failed part (weathering and accumulation of detritus on broken surfaces, etc.), we estimated the date that each failure occurred to the nearest six-month interval. As shown in Figure 12, the number of initial failures (first failure above threshold size for a given tree) among coast live oaks increased dramatically over time in the first two years of the study but has been decreasing since 2002 (Figure 12). For trees with branch or high bole failures or multi-stemmed trees, additional failures over the threshold size are possible after the initial failure. The number of such later failures occurring each year has increased substantially since 2002. These later failures constituted the majority of the observed failures for the first time in the 2004-2005 observation interval (Figure 12). Seventy four percent of the later failures have occurred in dead trees.

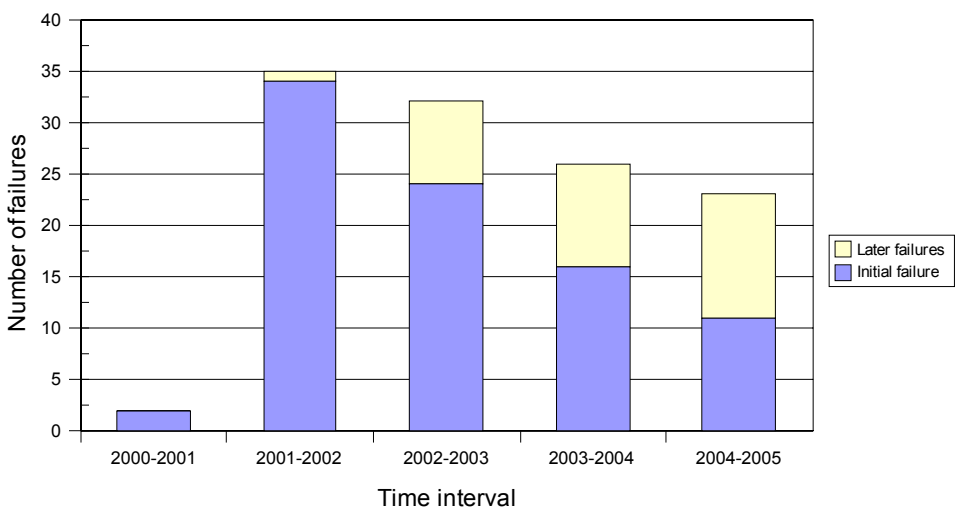

Figure 12. Number of initial (blue) and subsequent (yellow) failures above threshold size occurring in annual observation intervals between September 2000 and September 2005 among 629 coast live oaks that had not failed prior to September 2000. Annual observation intervals are from October of the first year through September of the following year.

Overall, most failures have involved trees or portions of trees that were dead at the time of failure. However, among living trees with *P. ramorum* canker, more than half of the failures occurred in live branches and stems (Figure 13). The majority of the failures in trees declining due to other factors and most of the failures in asymptomatic trees also occurred in live branches and stems.

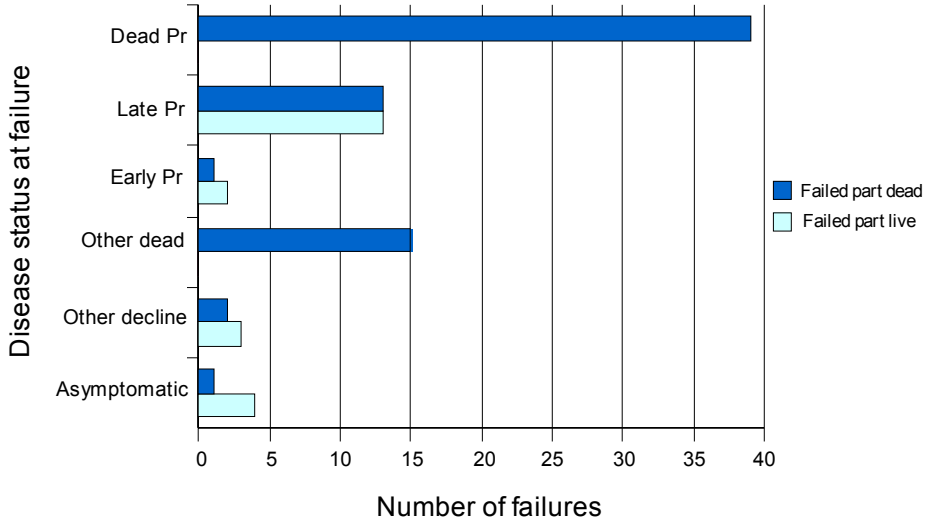

Figure 13. Tree disease status and condition of the failed part (live or dead) at the time of failure for initial failures above threshold size occurring between September 2000 and September 2005 in coast live oak study trees.

Calculation of failure rates for trees in different disease classes is complicated by the fact that the disease status of many trees has changed over the course of the study. In Figure 14,

cumulative failure rates over time for coast live oak study trees are plotted according to the disease symptom class observed in 2000. The disease status of the trees at the time of failure is also shown. These data exclude dead trees that had already failed by 2000.

As has been the case since 2002, trees killed by *P. ramorum* canker exhibited the highest failure rates (Figure 14). With only two exceptions, virtually all trees with *P. ramorum* canker symptoms had either died or progressed to the late disease stage (with *H. thouarsianum* sporulation and/or wood boring beetles) prior to failure (Figure 14). Trees that were asymptomatic in 2000 showed very low rates of failure over the study period (Figure 14).

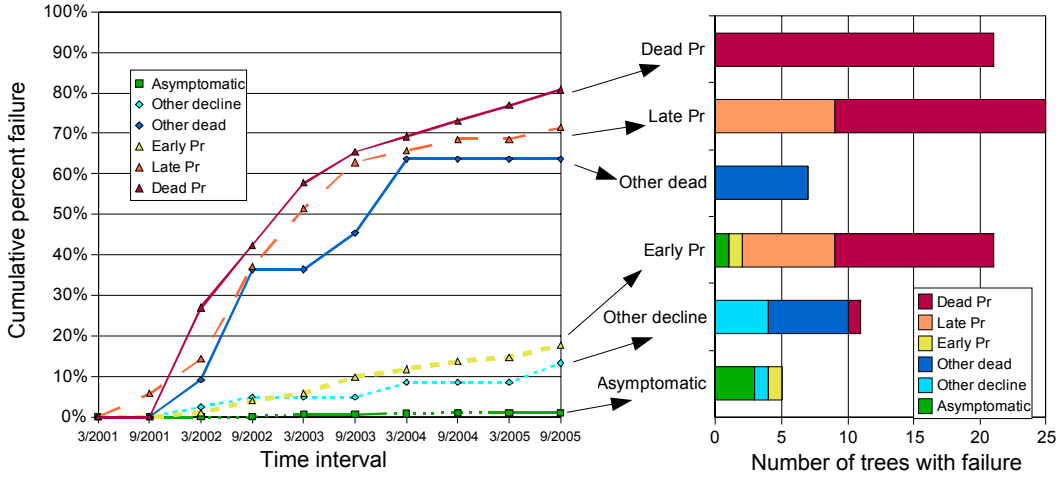

Figure 14. Cumulative failure rates (left) and tree disease status at failure (right) for coast live oaks by initial disease status in 2000. Only initial failures above the size threshold are shown. Failure dates were estimated to the nearest 6 month interval and failure percentages are plotted at the end date of each interval. Trees that had symptoms of both *P. ramorum* (**Pr**) and decline due to other agents (**other decline / dead**) are pooled with the respective *P. ramorum* symptom class. Dead trees that had failed prior to September 2000 are excluded.

Cumulative mortality and failure rates for *P. ramorum*-infected coast live oaks are compared in Figure 15. Since plots were first observed in September 2000, complete tree failure data is not available for the first time interval in the graph. However, for the six locations included in our tree failure study (Swiecki and Bernhardt 2003b, Swiecki and others in press), the failure rate in *P. ramorum*-affected trees prior to 2000 was 0.5%. Figure 15 shows that although mortality had increased in a relatively linear fashion over the interval from September 2000 to September 2005, failure rates showed an initial lag followed by a steep increase in the first half of this period. For the past several years, the percentage of *P. ramorum*-infected coast live oaks with initial failures has approximated the mortality rate due to *P. ramorum* (Figure 15). Because failures are also common in living trees with late *P. ramorum* canker symptoms (Figure 13), the failure rate among *P. ramorum*-infected coast live oaks could eventually exceed the mortality rate.

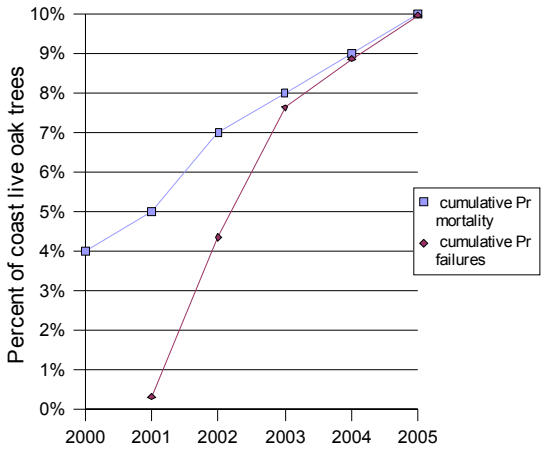

Figure 15. Cumulative initial failures in *P. ramorum*-infected coast live oak trees compared to the cumulative mortality rate due to *P. ramorum*. Mortality and failure percentages shown are cumulative to September of the year shown on the x axis.

For coast live oaks, we also calculated the time interval that elapsed between tree mortality and the initial failure above the study threshold size for cohorts of trees that died n 2001, 2002, 2003, and 2004 (Figure 16). Trees that were dead in 2000 could not be included in the analysis because we could not be certain of the year that these trees died. Among trees dying in 2001 through 2003, at least two-thirds of the trees failed within two years of mortality (Figure 16). However, the interval from tree death to failure appears to be rather variable; two of the nine trees that died in 2001 had still not failed after four years. Among the 16 trees that died in 2004, five had failed prior to tree mortality by up to two years (Figure 16). Furthermore, in all years, many of the trees that failed in the year of mortality (0 years from mortality to failure) were live at the time of failure but died as the result of a low bole or root crown failure.

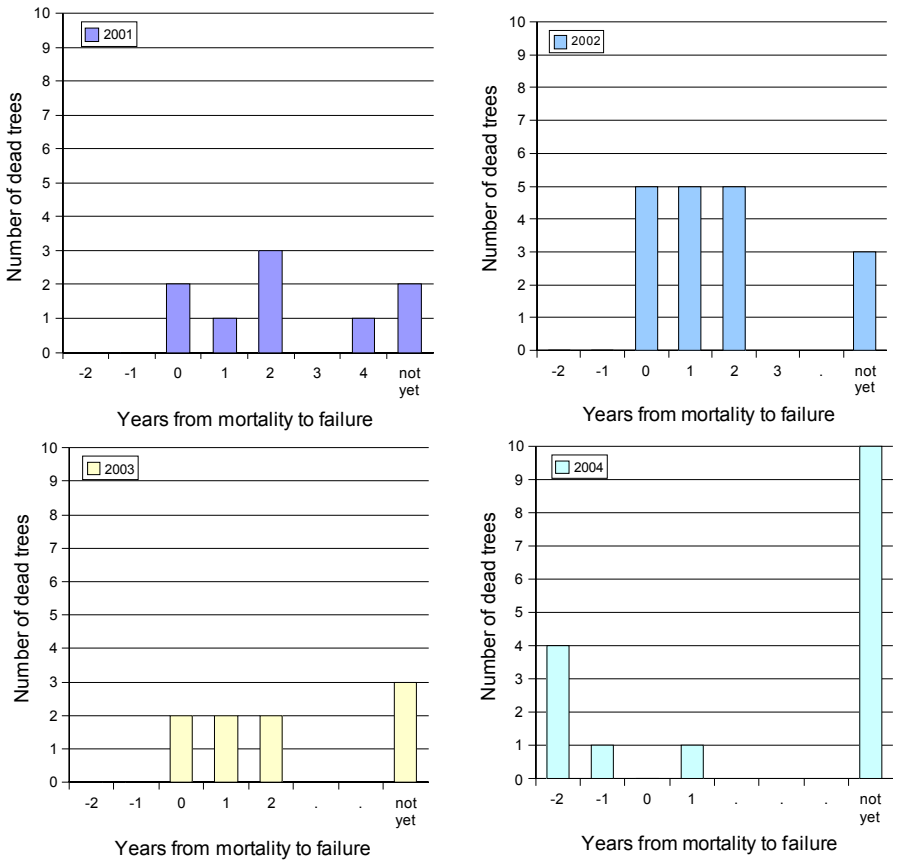

Figure 16. Years from mortality to failure for coast live oak trees that were first recorded as dead in September of 2001, 2002, 2003, and 2004. For all mortality years, some trees had not failed by 2005 ("not yet" column).

Regeneration

To determine the potential for killed SOD canker hosts to be replaced by seedlings of the same species, we counted the number of seedlings (plants with DBH<3 cm) of coast live oak, California black oak, and tanoak in the study plots during each September evaluation.

Coast live oak

Almost all of the 128 plots with coast live oak overstory had coast live oak seedlings at some point between 2000 and 2005. Only five of these plots had no coast live oak seedlings in any of the six years of the study. Seedling numbers fluctuated widely, both within plots at the same location, and also in the same plots from year to year (Figure 17). Forty six percent of the plots had no seedlings in at least one year. Average seedling counts per plot reached a maximum of 80 at location 8 in 2001 (Figure 17). In half of the plots, no more than ten coast live oak seedlings were observed in any year.

The number of dead seedlings tallied in plots each year was almost always much smaller than the drop observed in live seedling numbers from one year to the next, presumably because small dead seedlings do not persist in the plots. This would certainly be the case for seedlings destroyed by herbivores. Hence, counts of dead coast live oak seedlings within plots did not provide an accurate picture of seedling mortality from year to year.

Seven locations showed peak seedling populations in 2001, the only year in which average seedling counts per plot differed from other years (Table 4). Location 8 was unique in having exceptionally high seedling counts (up to about 200) in some plots in 2001 (Figure 17). In locations that showed strong increases in seedling counts in 2001, the mean number of seedlings per plot has decreased sharply in subsequent years (Figure 17). It appears that this decrease is due to mortality of many of the seedlings established in 2001.

We used a repeated measures analysis of variance of plot seedling counts to determine whether various factors were associated with changes in seedling populations over time. These analyses indicate that seedling counts varied significantly over time and between locations (F test $p<0.0001$ for both factors) and are somewhat negatively correlated with the number of live coast live oak trees in the plot (F test $p<0.0258$). The interaction between location and time was also highly significant (Pillai's Trace, Wilk's Lambda $p<0.0001$). Factors that were not significantly related to seedling population levels included whether coast live oaks with *P. ramorum* canker symptoms were present in the plot, overall plot canopy cover, shrub cover in the plot, and overall vegetation type (using grouped CALVEG alliances).

The overwhelming majority of coast live oak seedlings in the plots were very small, typically less than 10 cm tall (Figure 18). A few larger seedlings, generally up to about 30 cm tall were found in some plots. These larger seedlings were generally multistemmed and shrubby in appearance. In plots that have experienced significant canopy loss due to tree mortality and/or failure over the course of the study, larger shrubby seedlings have become more common, and some of these were up to about 50 cm tall in 2005. Such seedlings are often but not exclusively found near or among failed branches and other woody debris that provide some protection from deer browsing (Figure 18, cover photo). Over the five years of this study, none of the coast live oak seedlings in the plots have been recruited to the tree stage (DBH at least 3 cm).

Table 4. Mean number of coast live oak seedlings or tanoak seedlings per plot (standard deviation) by year. Means are calculated from plots with overstory of coast live oak (128 plots) or tanoak (39 plots).

Year	2000	2001	2002	2003	2004	2005
Coast live oak	3.9 (5.8)	17 (32)*	9.8 (15.8)	10 (20.5)	8.7 (11.3)	7 (9.1)
Tanoak	11.8 (13.4)	11.5 (9.5)	13.8 (12.9)	13.1 (13.3)	12.4 (10.3)	13.8 (12.3)

* significantly different from all other means within species according to Tukey-Kramer HSD.

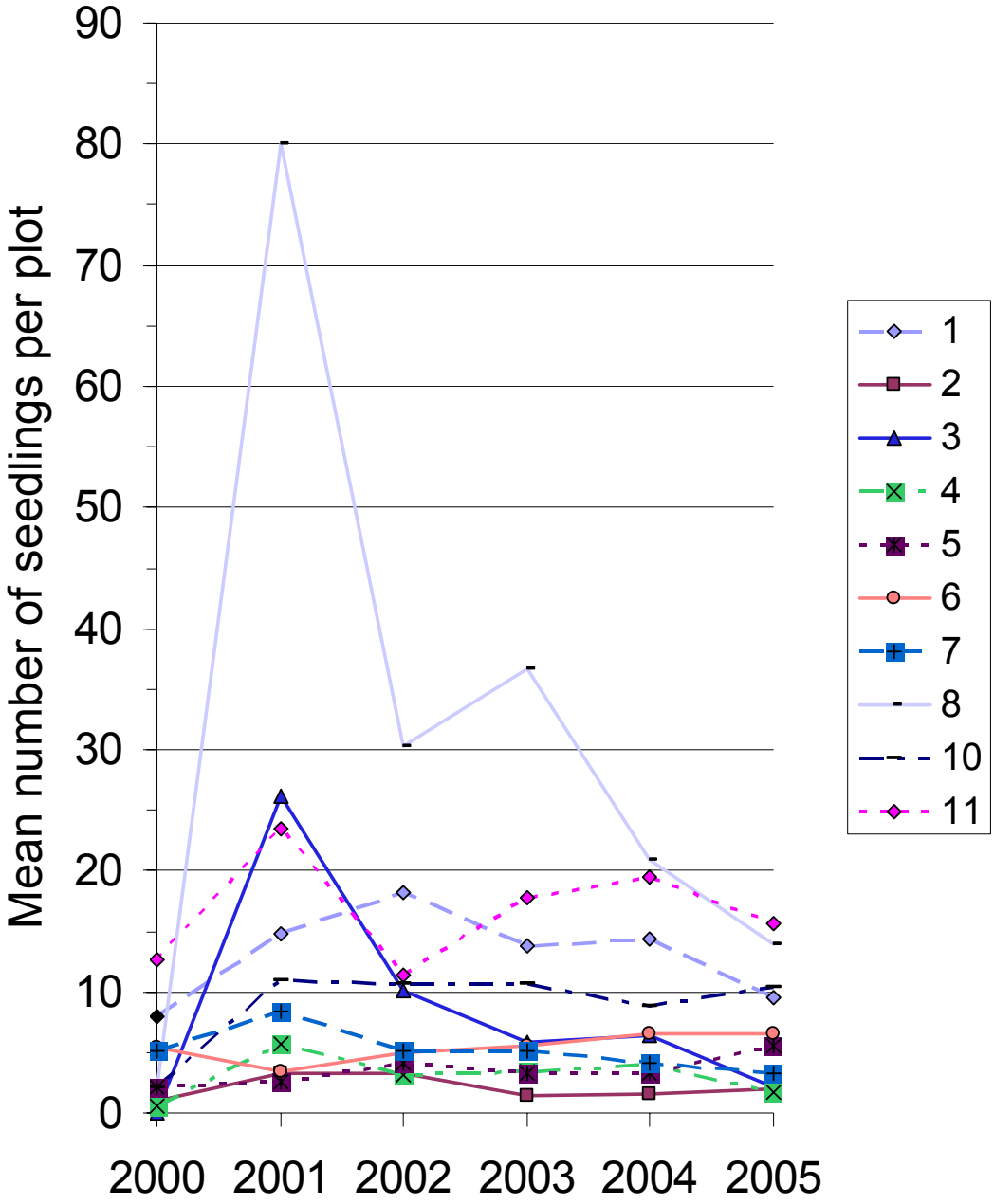

Figure 17. Mean numbers of coast live oak seedlings per plot at each of 10 locations with coast live oak overstory from 2000 through 2005. Location numbers shown in the legend correspond to locations in Table 1.

Figure 18. Typical small (left) and larger (right) coast live oak seedlings observed in plots.

Tanoak

Tanoak seedlings were present in 23 or more of the 25 plots with tanoak overstory between 2000 and 2005. At some coast live oak locations, tanoak seedlings were also found in plots lacking overstory tanoak. Tanoak seedlings most commonly occurred as shrubby seedling-sprouts with multiple stems, and were commonly at least 50 cm tall, although smaller seedlings were also present in some areas. Consequently, tanoak seedling counts in plots were more stable from year to year than coast live oak seedlings counts (Figure 19, Table 4). For coast live oak, the annual coefficients of variation of mean seedling counts per plot ranged from 42% to 224% between 2000 and 2005, compared with coefficients of variation ranging from 83% to 113% in tanoak over this period. At least three tanoak seedlings in the study plots have attained a DBH of 3 cm or more over the study period and are now classified as trees.

Unlike coast live oak twigs, tanoak twigs are susceptible to *P. ramorum*; understory tanoak seedlings commonly showed tip dieback and/or mortality of individual stems. However, over the period of the study, this damage has not resulted in a reduction in tanoak seedling populations (Figure 19). According to a repeated measures analysis of variance, seedling counts per plot have not changed significantly between 2000 and 2005.

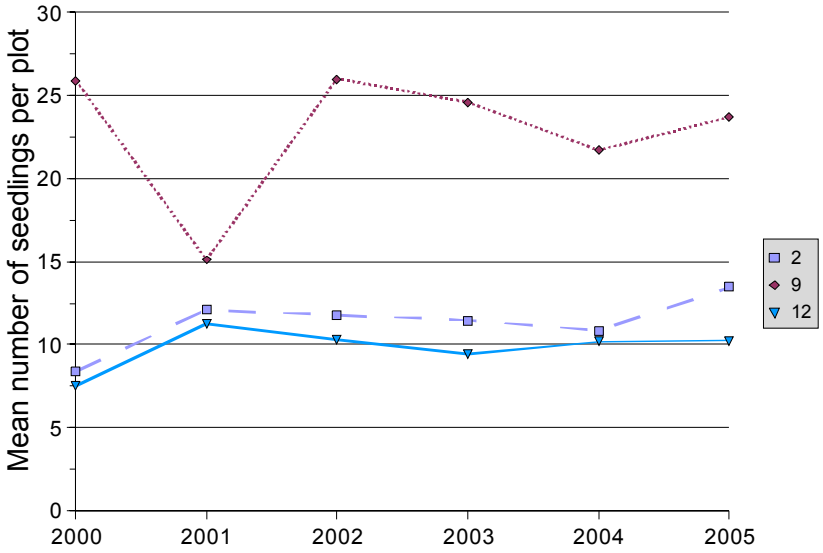

Figure 19. Mean number of tanoak seedlings and seedling sprouts per plot at each of three locations with tanoak overstory. Location numbers shown in the legend correspond to locations in Table 1.

Balance between tree mortality and seedlings in coast live oak and tanoak

By killing overstory coast live oak and tanoak trees, *P. ramorum* has the potential to change the composition of infested forests both over the short term and the long term. Loss of coast live oak or tanoak canopy due to *P. ramorum* canker is more likely to lead to a long-term shift in forest composition if regeneration of these species is lacking in areas where overstory canopy is lost. Hence, the presence of seedling advance regeneration is most critical in plots that have lost overstory trees due to *P. ramorum* canker.

We compared plot-level data on tree mortality with seedling counts to provide a measure of the adequacy of advance regeneration. If overstory mortality was present within a plot but understory seedlings of the same species were lacking, we considered the level of advance regeneration to be inadequate. However, depending on the rate at which seedlings are successfully recruited to the tree stage, advance regeneration could also be inadequate in plots with overstory mortality that had very low numbers of seedlings. Figure 20 shows the relative balance between tree mortality and seedling presence for coast live oak and tanoak in 2000 and 2005.

The percentage of plots with at least one dead coast live oak tree approximately doubled between 2000 and 2005, from 27% to 49% (Figure 20). Over the same interval, the percentage of plots without coast live oak seedlings decreased by half, from 40% in 2000 to 20% in 2005. Due to the increase in the number of plots with seedlings, the percentage of plots that had coast live oak overstory mortality but no coast live oak seedlings decreased slightly over this time interval, from 12% to 8%. Hence, according to this particular measure, the potential for coast live oak mortality to be replaced though regeneration has not changed substantially between 2000 and 2005, despite the great increase in tree mortality due to *P. ramorum* that has occurred during this period. Since there is no guarantee that seedlings present in plots with coast live oak

mortality will be recruited, it is possible that long-term reductions in coast live oak cover may occur in more than the 8% of plots that lack regeneration. Although coast live oak seedlings in some plots with recent mortality-related canopy gaps have grown substantially, none of these seedlings have grown large enough to escape browsing of the leader by deer.

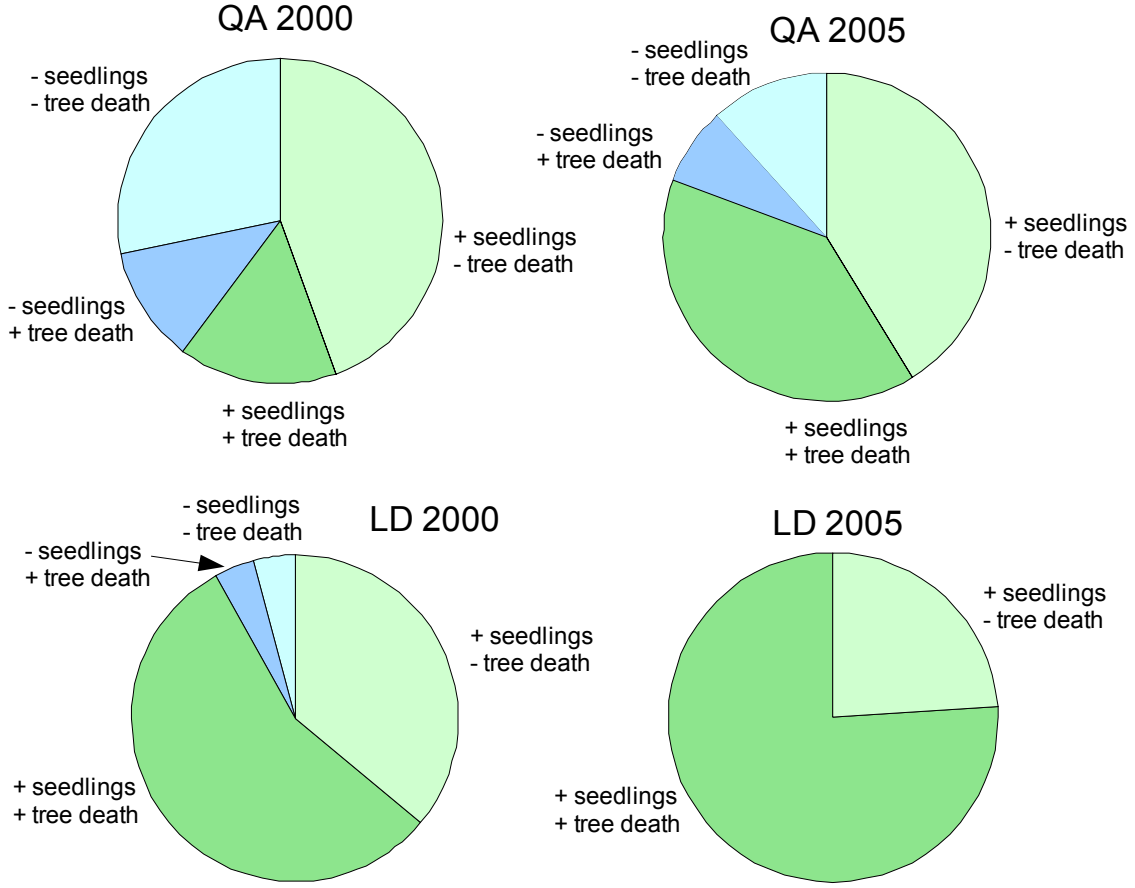

Figure 20. Percent of plots with and without tree mortality and regeneration of the same species in 2000 and 2005. QA=coast live oak (128 plots). LD= tanoak (25 plots). Plots with regeneration are shown in green. Plots without regeneration are shown in blue. Plots with dead trees are shown in dark shades and those without dead trees are shown in lighter shades.

Among plots with tanoak trees, the percentage of plots with tanoak mortality increased from 60% in 2000 to 76% in 2005. Over the same interval, the percentage of plots without tanoak seedlings decreased from 8% in 2000 to 0% in 2005. Due to the decrease in the number of plots without seedlings, the number of plots with both tanoak mortality and no seedlings decreased from 4% in 2000 to 0% in 2005 (Figure 20). Hence, it does not appear that *P. ramorum* has substantially impacted the tanoak seedling bank or overall regeneration potential at this point. However, because *P. ramorum* infects and kills tanoak saplings and small diameter tanoak trees, sapling regeneration may not be successfully recruited to mature trees in many of these plots.

California black oak

The study locations were chosen for the presence of coast live oak and tanoak. California black oak rarely occurs in the study plots, but co-occurs with coast live oak at six locations. Only 16 of the trees in the study are California black oaks and they occur in 11 study plots.

Three California black oaks were dead at the start of the study and three have died since the study began; two of these deaths were due to *P. ramorum*. Of the remaining 10 California black oaks, four are healthy, one is in decline due to wood decay fungi, and five have *P. ramorum* symptoms.

California black oak seedlings were extremely scarce in the plots. Only five plots had California black oak seedlings present in low numbers (1 to 3 seedlings) in 2001. By 2005, only two plots had California black oak seedlings (1 seedling in each plot); neither of these was found in a plot with overstory California black oak trees.

Stem water potentials of coast live oaks

We collected data on changes in stem water potentials (SWP) during the first five years of the study and have previously reported on this data (Swiecki and Bernhardt 2001ab, 2002ab, 2003a, 2004, 2005b). Because SWP readings provide information on the degree to which winter rainfall affects late season water stress, we continued collecting SWP data on a subset of the previously monitored trees from each coast live oak location in September 2005.

The average SWP for the three monitored trees at each location are shown in Figure 21. The pattern of annual changes in SWP is very similar among the locations, and correlates well with the annual changes in rainfall. The 2004-2005 rainy season was the wettest within the study area since we began collecting data in 2000 (Figure 21, top). For all of the coast live oak locations, 2005 SWP readings were at or near the highest levels (indicating low water stress) observed.

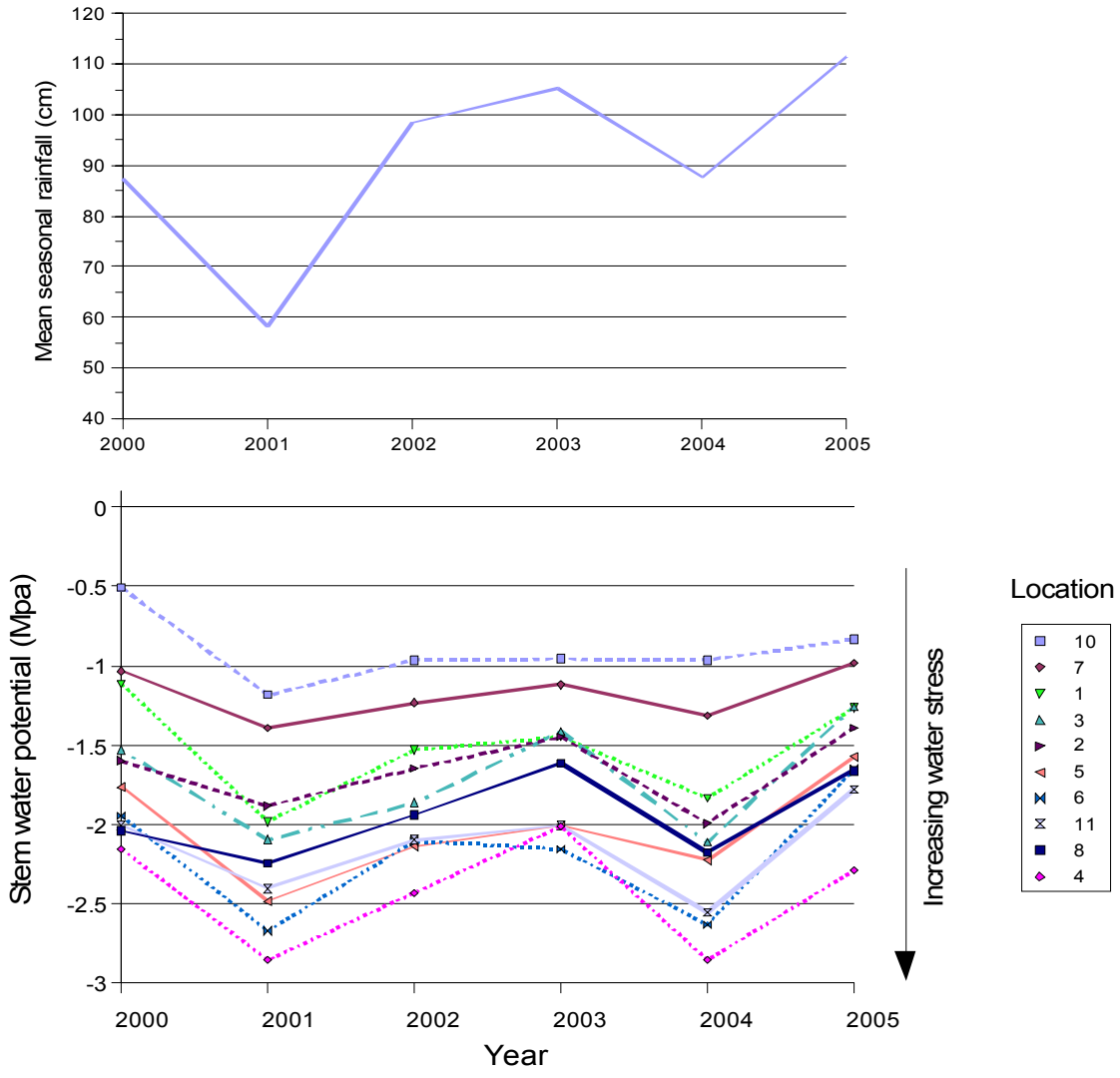

Figure 21. Average stem water potential for coast live oaks at each location across all years compared with average seasonal rainfall (top graph). Rainfall is for September of the previous year through August of the listed year; and is the mean for all study locations calculated from the nearest weather station data for each location. Location numbers correspond to those given in Table 1.

DISCUSSION

Since the start of this study in September 2000, annual mortality related to *P. ramorum* has increased in a linear fashion in both coast live oak and tanoak (Figure 4). Under the simplifying assumption that this trend has been constant from the onset of the epidemic, we estimated that the first tanoak mortality could have occurred in the study plots in 1995 and the first coast live oak mortality in 1996. These dates agree well with the first reports of *P. ramorum*–related mortality in Marin County, CA: 1995 for tanoak and 1997 for coast live oak (Svihra 2001). Because the Marin County reports were among earliest known occurrences of *P. ramorum* in

California, this suggests that the study plots are located in areas that were among the first to be affected by *P. ramorum.*

P. ramorum was first recognized as causing a disease of rhododendrons in Europe in 1993 (Werres and others 2001). Recent genetic analyses of *P. ramorum* isolates (Ivors and others 2006) indicate that *P. ramorum* was introduced into California forests recently, most likely by way of contaminated nursery stock located in or near forested areas. *P. ramorum* canker was first recognized by the appearance of unusually high numbers of dead trees, and therefore it is not known when the earliest canker symptoms appeared in Marin County.

Because disease incidence in tanoak increased linearly over time between 2000 and 2005, extrapolation of this line back to the x axis (0% disease incidence) may provide a reasonable estimate of the date that symptoms originally appeared in the tanoak plots. Using this method, we estimate that the first *P. ramorum* symptoms in tanoak may have appeared as early as 1989 (Figure 5). If the assumption of linearity for overall infection in tanoak prior to 2000 is not valid, this estimate could be off by more than is implied by the 95% confidence interval bounds.

An alternative method for estimating the initial date of symptom appearance uses the estimated date of initial mortality (1995) from Figure 4. Given that approximately 30% of tanoaks in the study that developed visible symptoms between 2000 and 2005 died within two years (Figure 7), visible canker symptoms may have preceded initial tanoak mortality by as little as two years. This suggests that visible canker symptoms should have been evident by about 1993, although it does not preclude an earlier date of initial symptom appearance.

Bole cankers are unlikely to have developed until sufficient levels of inoculum had been produced on infected tanoak twigs or other foliar hosts. Hence, the original introduction of *P. ramorum* into the plot areas probably preceded the appearance of bole canker symptoms by at least several years.

Disease epidemiology in tanoak and oak stands differs in several ways that are reflected in the plot data. Artificial inoculations (Rizzo and others 2002) have shown that tanoak is more susceptible than coast live oak to *P. ramorum* canker development. This attribute is reflected in our field data in both the higher overall disease incidence (Figure 3) on tanoak and the shorter interval between symptom onset and mortality seen in tanoak relative to coast live oak (Figures 6, 7).

Furthermore, *P. ramorum* infects and sporulates on tanoak twigs, providing a source of inoculum within a given tree (Rizzo and others 2005). Coast live oak has not been shown to develop foliar or twig infections that produce *P. ramorum* inoculum. This observation and patterns of disease in the field (Swiecki and Bernhardt 2001ab, 2002ab) indicate that *P. ramorum* cankers on coast live oak are initiated by spores produced on other hosts, primarily California bay. Conditions that favor heavy spore production on bay and the transport and deposition of spores under conditions that favor disease initiation appear to occur somewhat episodically in the coast live oak study locations. This is reflected in the high level of variation in the appearance of newly symptomatic trees over time (Figure 2) and the variation in the numbers of newly symptomatic hosts between locations (Figure 8).

High levels of inoculum production on California bay are correlated with wet conditions and mild temperatures during the spring months (Davidson and others 2005). However, infected bay leaves are shed more quickly than healthy leaves (Rizzo and others 2005), especially in water-stressed bay trees. Hence, in some locations, high levels of *P. ramorum* inoculum may be produced on bay foliage only when wet years occur in succession. Coast live oak study locations that have shown little or no increase in disease incidence since 2000 may represent sites where

most of the susceptible oaks close to California bay were infected by that time. Locations showing greater increases since then may represent sites where inoculum levels had not built up to high levels on California bay trees during the previous favorable rainfall periods that occurred between 1995 and 1998. In years that favor high spore production, some of the at-risk coast live oaks at these locations have subsequently been infected and developed symptoms.

Temporal variation in the appearance of visible canker symptoms may also be related to differences in the amount of time that elapses between infection and external symptom development. Such variation has been observed even in artificially inoculated trees (Rizzo and others 2002). Compared with artificially-inoculated trees, a population of naturally-infected trees will have much greater variation in factors that may influence disease development, including initial inoculum density, the number and locations of infection sites, seasonal timing of infection(s), and host characteristics (e.g., bark thickness, water stress level, carbohydrate reserves). Hence, we expect that the apparent latent period associated with natural infections may vary more widely than that seen in artificially inoculated trees, and may range from less than one year to two or more years. Therefore, newly symptomatic trees observed in a given year may or may not have been infected during the preceding wet season.

Conditions for inoculum production in spring 2006 were highly favorable. Furthermore, the 2005-2006 rainy season was the second successive year of relatively high rainfall. We therefore anticipate new canker symptoms associated with this infection period will become evident in 2006 and possibly succeeding years.

Failures

Most recent coast live oak failures in oak woodlands affected by *P. ramorum* have occurred in trees infected by *P. ramorum* (Figures 13, 14; Swiecki and Bernhardt 2003b, 2004, 2005b, Swiecki and others in press). We previously reported that the highest number of live stem failures occurred among trees with late *P. ramorum* canker symptoms, i.e., symptomatic trees colonized by secondary organisms including *H. thouarsianum* and/or bark or ambrosia beetles (Swiecki and Bernhardt 2003b, 2004, 2005b, Swiecki and others in press). Data from the most recent failures have followed these same trends (Figures 13, 14).

After an initial lag, cumulative failure rates and mortality rates of *P. ramorum* infected trees have been converging over time (Figure 15). Due to failures in live infected trees, it may be that eventually failure rates will slightly exceed mortality rates.

The time interval between tree death and the first substantial failure (i.e., above threshold size) has been relatively variable in coast live oaks (Figure 16). This is probably associated with the fact that disease progress can also be quite variable (Figures 6, 7). In the tree population included in this study, multiple patterns of disease progress have been seen (Swiecki and Bernhardt 2005b), ranging from a relatively quick decline to apparent symptom remission. Furthermore, failure potential is strongly correlated with the presence of secondary wood-decay fungi and wood-boring beetles (Swiecki and Bernhardt 2003b, Swiecki and others in press). The Colonization of *P. ramorum* canker–infected trees by these secondary organisms is variable with respect to both timing and extent. This variability explains why tree failure can occur over a wide span of time relative to tree mortality.

Regeneration

Data for the period from September 2000 to September 2005 indicate that *P. ramorum* has not negatively impacted seedling populations of either coast live oak or tanoak. This suggests

the possibility that mortality of these species due to *P. ramorum* could be replaced in kind through regeneration, but this outcome is far from certain. As acorn-producing trees are eliminated from the overstory, new seedlings are less likely to become established. Therefore, successful regeneration may depend largely on the successful recruitment of existing seedlings.

This process appears to be occurring at some coast live oak locations in canopy gaps that have formed due to *P. ramorum*-related mortality. However, various factors could inhibit successful recruitment: seedling growth may be stalled by excessive browsing by deer or other herbivores; seedlings may be killed by fire or other damaging agents; regeneration of other species, including Douglas-fir and California bay may overtop and suppress seedlings. In addition, due to the high stocking levels found in many of the plots, growth of remaining overstory or sub-overstory trees is likely to refill relatively small canopy gaps even without regeneration. The chance that coast live oak will refill *P. ramorum*-induced gaps could be reduced unless these stands are monitored and managed to promote regeneration.

Furthermore, it is unlikely that coast live oak seedlings or saplings are exposed to significant levels of selection pressure for resistance to *P. ramorum* because these stages do not appear to be infected by *P. ramorum.* Therefore, coast live oak trees arising through natural regeneration may be as susceptible to *P. ramorum* as were their predecessors.

For tanoak, the potential for natural regeneration is further complicated by the fact that sapling stems can be killed by *P. ramorum* cankers. As long as the root crown remains viable and the root system is healthy, many of these saplings will persist by continuing to produce new sprouts. It remains to be seen whether tanoaks can grow to maturity in areas where the disease has been especially intense. However, because tanoaks are susceptible to *P. ramorum* at all growth stages, high disease pressure should tend to favor selection for more resistant tanoak genotypes, which would have the potential of reducing disease impact in the forest over the long term.

In both this study and a study in Sonoma County which included hardwood forests dominated by California black oak, seedling counts of black oak were low (Swiecki and Bernhardt 2005a). Given that California black oak seedlings and trees are so rare in the study areas, it appears likely that additional mortality of California black oak overstory trees due to *P. ramorum* is unlikely to be replaced by the current population of California black oak seedlings, and that seedling numbers will continue to decline. Concerted management efforts may be necessary to ensure the persistence of California black oak in forests affected by *P. ramorum.*

Stem water potentials

Previous annual reports have presented data on changes in stem water potential (SWP) readings for all subjects trees (the center tree in each plot) for tanoaks and coast live oaks (Swiecki and Bernhardt 2001, 2002b, 2003a, 2004, 2005b). Briefly stated, results have shown that most coast live oaks with *P. ramorum* canker maintained relatively high SWP levels and did not show progressive increases in water stress as disease progressed. For coast live oak, trees with low water stress (high SWP) were more likely to develop *P. ramorum* canker than were more water-stressed trees.

September SWP readings were highly correlated from year to year for individual coast live oak and tanoak trees. Although mean SWP varied from year to year, the SWP of most trees shifted up or down by an amount that approximated the overall mean year to year difference. After removing the overall year to year differences, the SWP of individual trees were quite consistent from year to year. Hence, we believe that the reduced sample of SWP measurements

made in 2005 provides a reasonable estimate of the trend that would have been observed among the entire monitored tree population.

The 2004-05 and 2005-06 rainy seasons provide the first example of two successive high rainfall years over the course of the study. By monitoring the SWP of the reduced tree sample in 2006 we will gain a better idea as to whether there is a significant year to year carry-over effect that could result in especially high September SWP readings.

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