

Sudden Oak Death Update for Foresters and Landowners

Ukiah & Eureka, California

May 12-13, 2010

Biology and epidemiology of *P. ramorum*

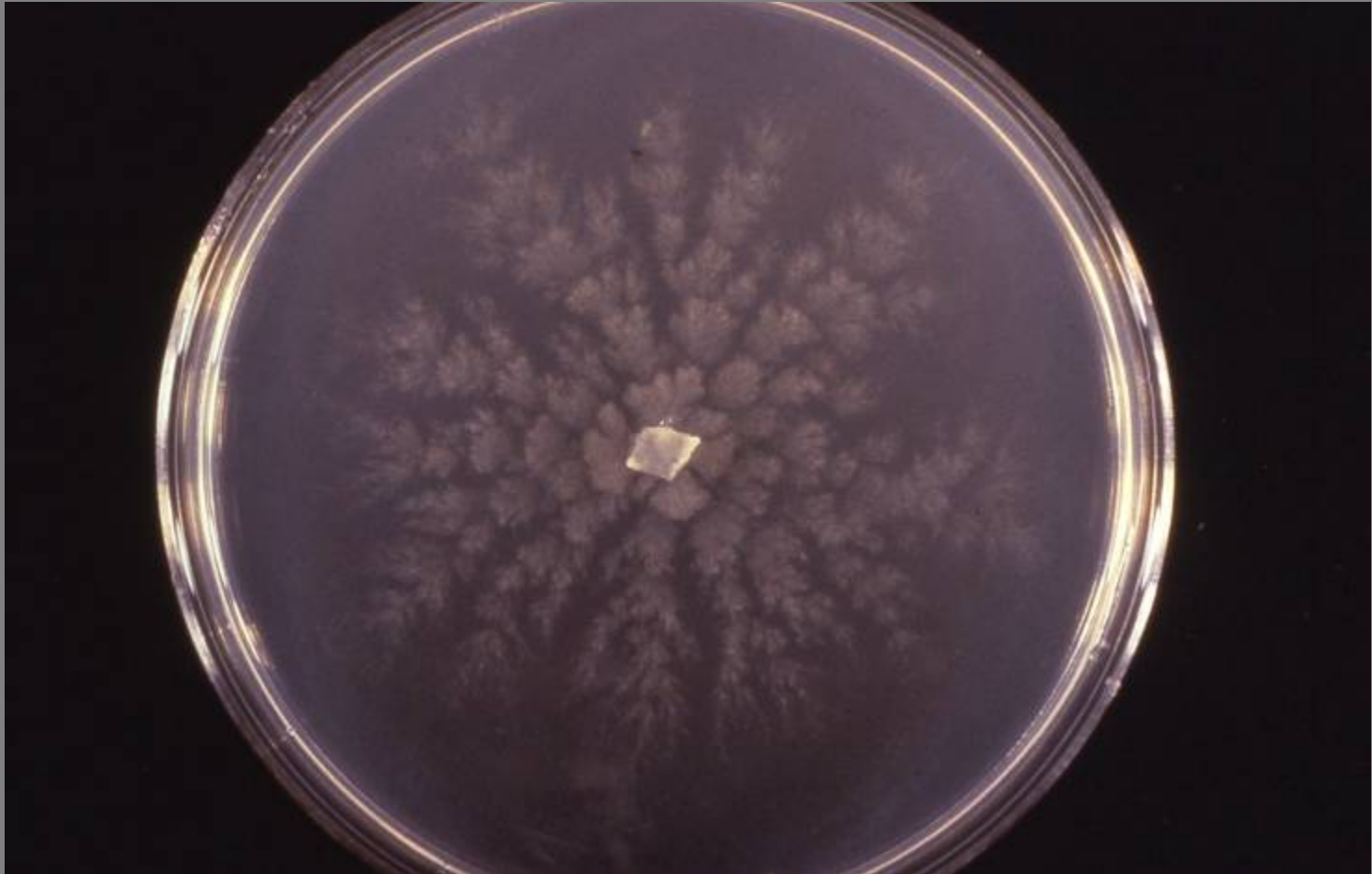
Matteo Garbelotto, U.C. Berkeley



The year was...

70's, 80s?

P. ramorum growing in a Petri dish



Organism new to science

- Origin unknown
- Biology unknown
- Symptoms caused unknown
- Immediately though highly regulated





Stem canker

Rhododendron:
In EU mostly a nursery
issue, but also present in
nurseries in US and Canada



Leaf necrosis



Symptoms not always present



- Early stages of infection
- Cryptic infection in rhododendron roots
- Plants chemically treated
- Leaves plucked



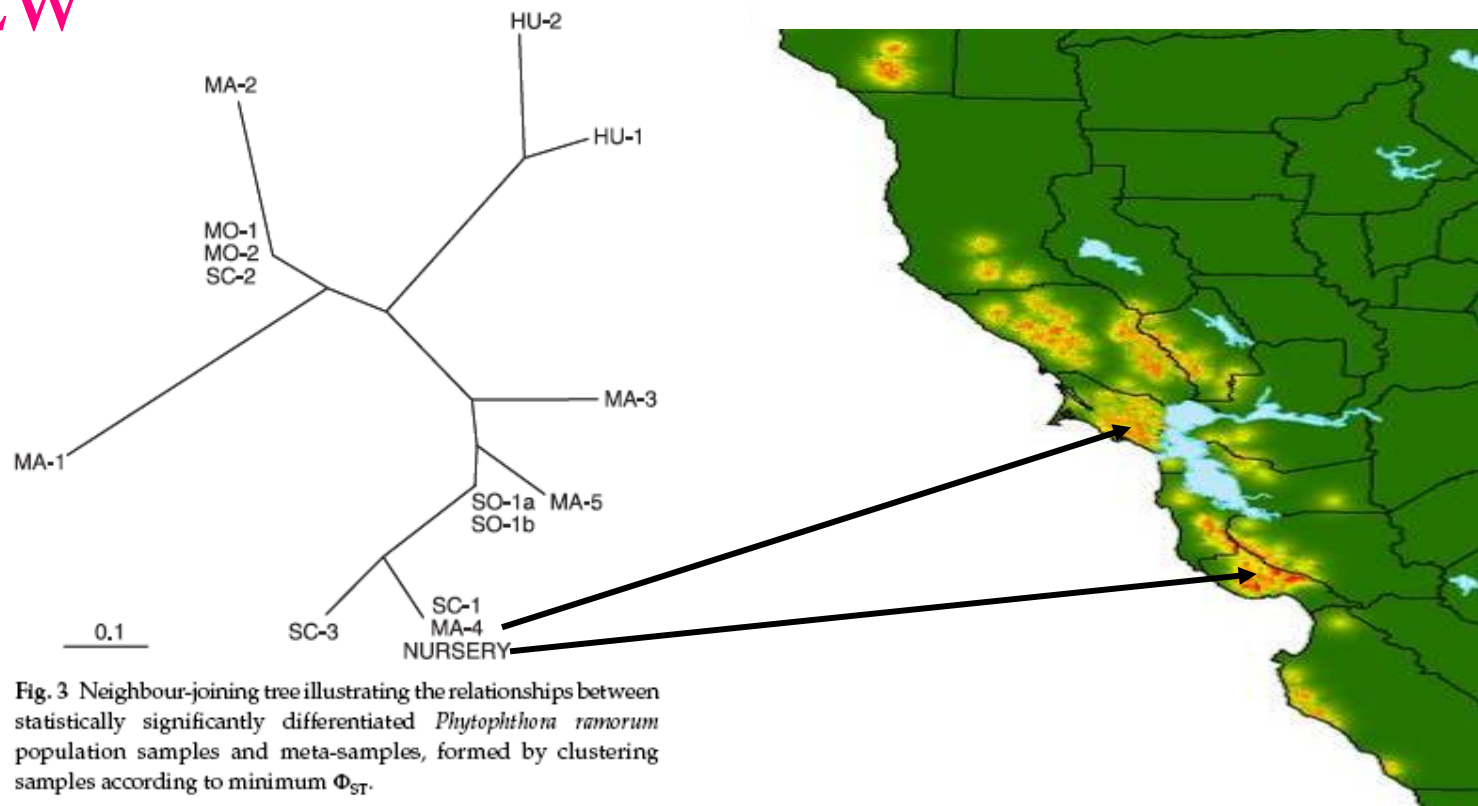


Pathogen

- Q: Where does it come from?

A: While area of origin is still unknown, commercial plant nurseries were unequivocally identified as sources of original introduction

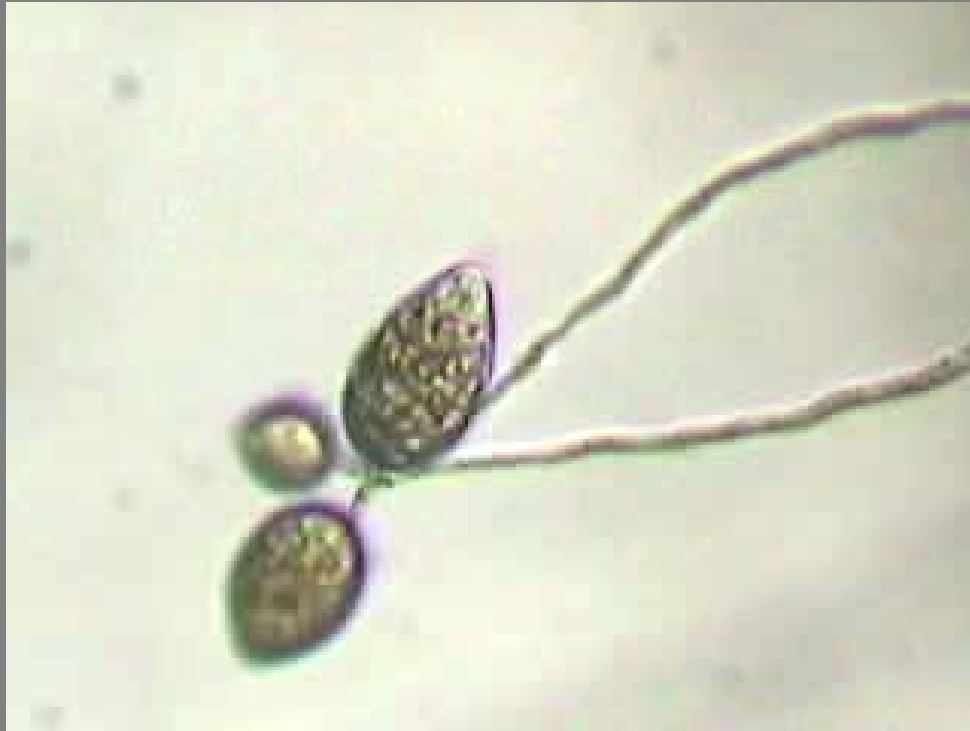
NEW





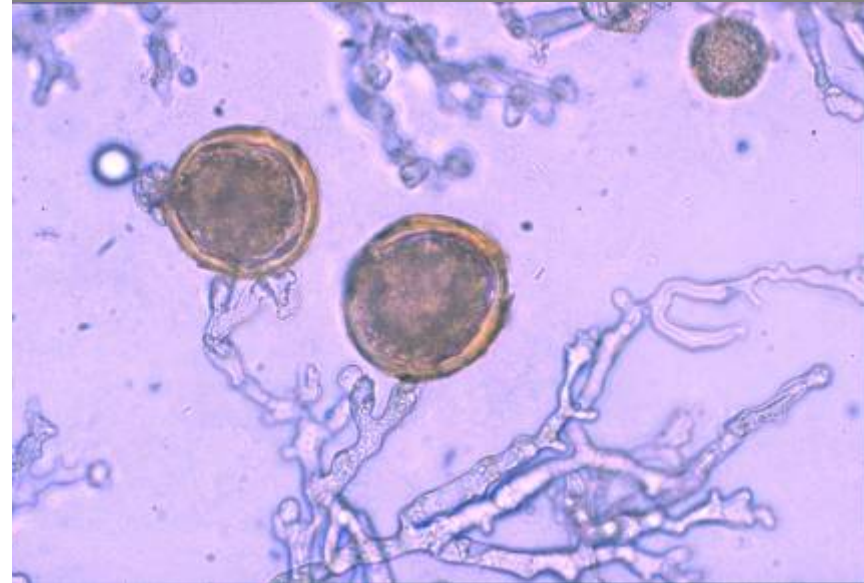
Umbellularia

Phytophthora ramorum



**Sporangia: produced when
It is wet**

Chlamydospores



How do sporangia move?

- Airborne
- Waterborne/soilborne/ movement of infected plants
- Can move 100-200 m, but in strong winds up to 2 km

Reconstruction of the Sudden Oak Death epidemic in California through microsatellite analysis of the pathogen *Phytophthora ramorum*

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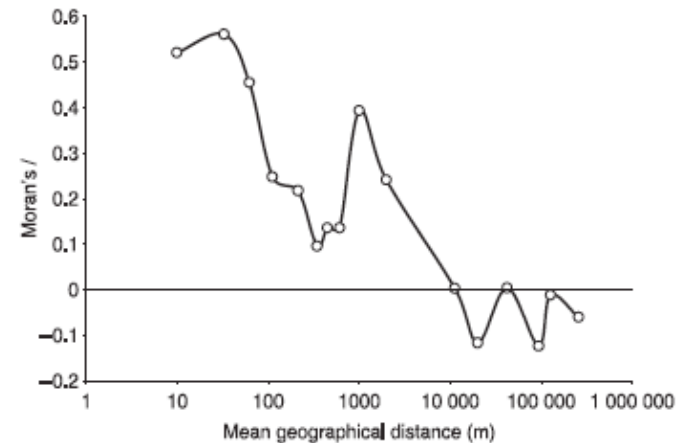
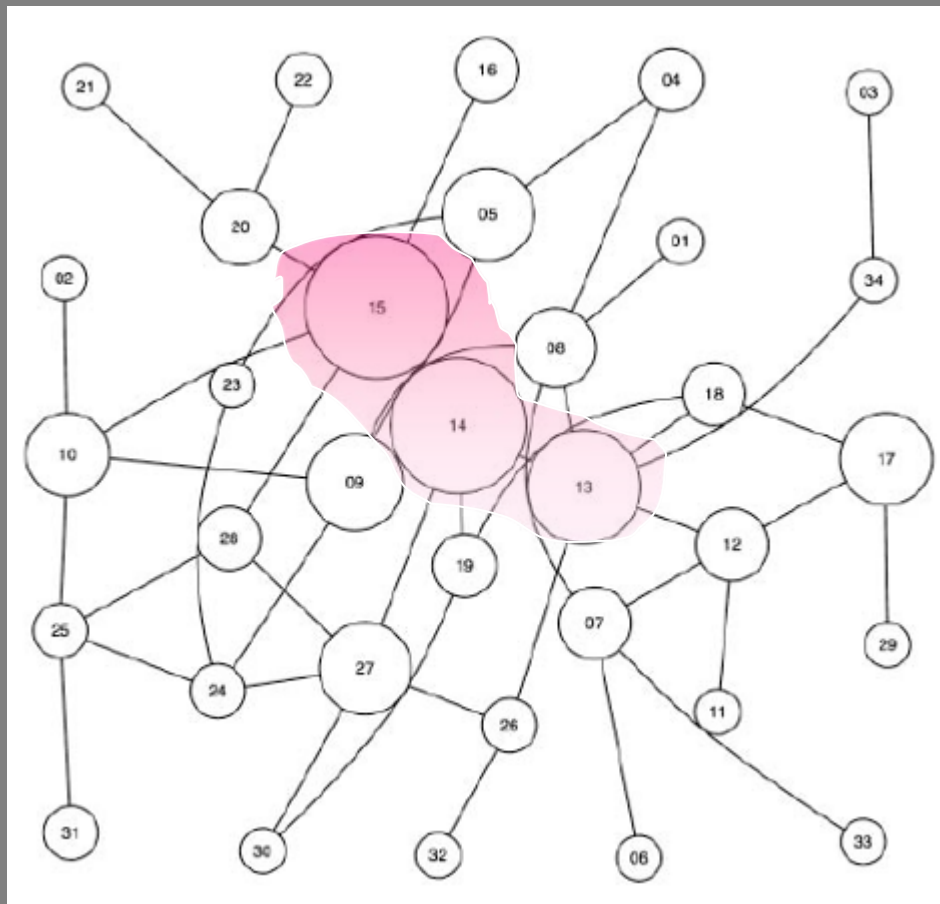


Fig. 4 Spatial autocorrelation analysis of genetic and geographical distance in *Phytophthora ramorum*. Moran's *I*-index, averaged over loci, was calculated from the repeat number at each of four variable microsatellite loci.

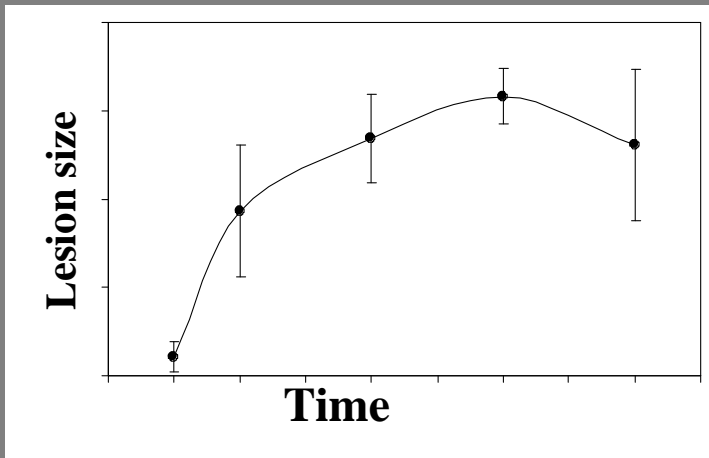


← Circles represent genotypes: red shade indicates those that were introduced, are common in all infested sites, and are the progenitors of all other genotypes. Relatedness is indicated by connecting lines

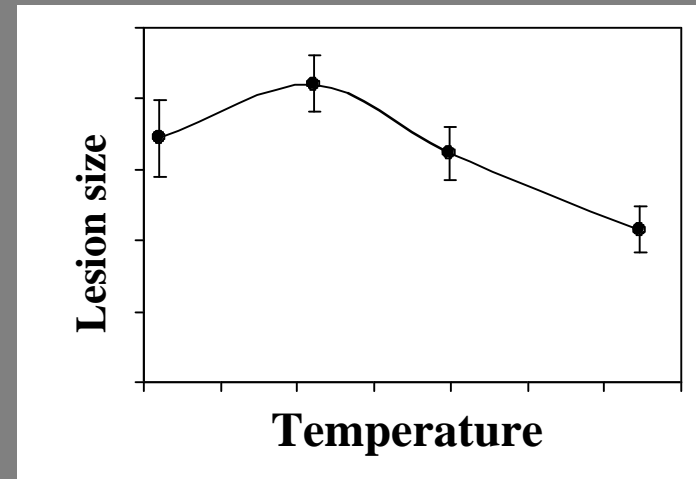
When do sporangia infect new hosts?



- Temperature and leaf wetness



↑
12 hours



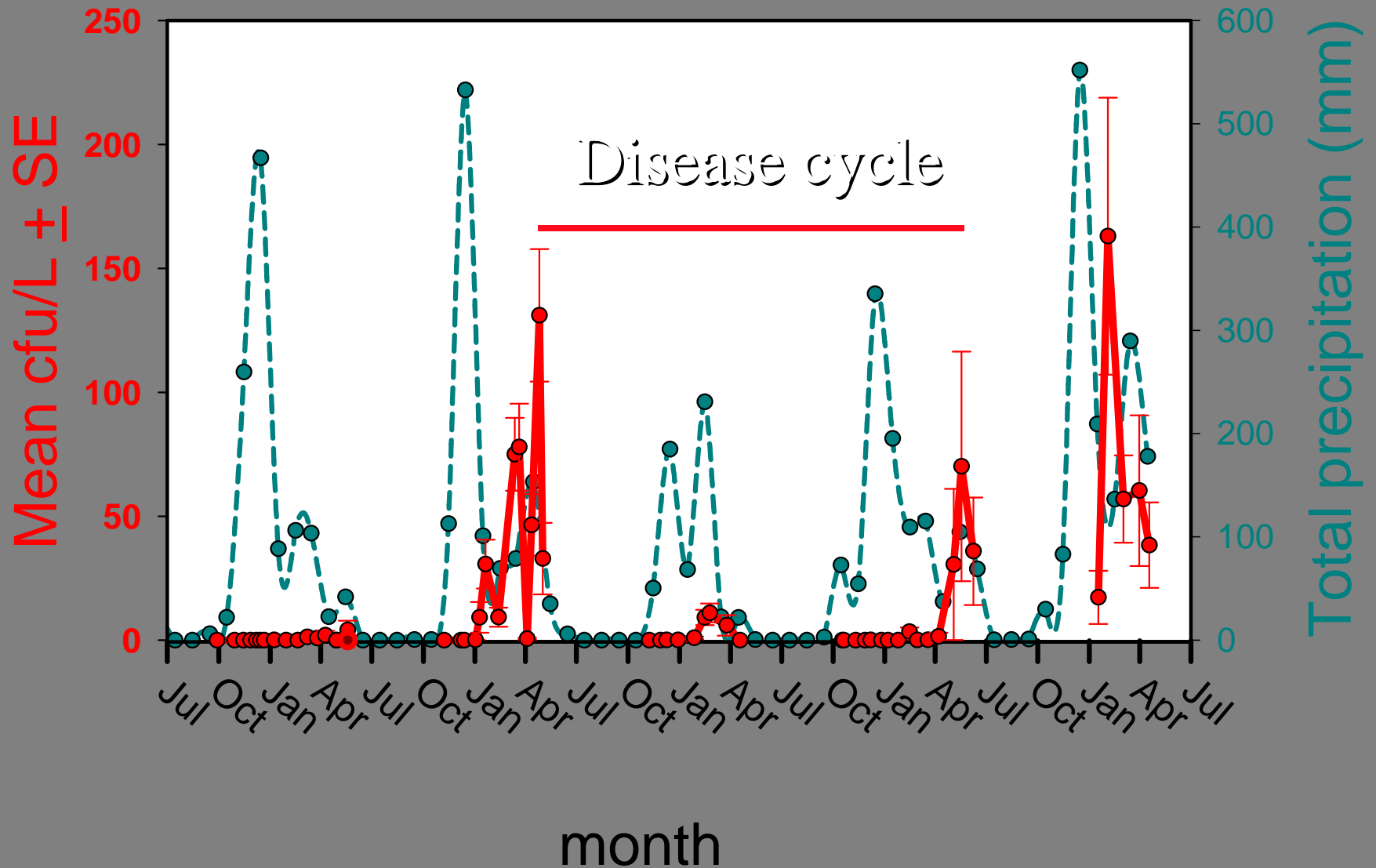
↑
20 C

Infection occurs....

- Via zoospores produced by airborne sporangia.....
- Requires water: limited infection with fog, high with rain
- Needs warm temperatures



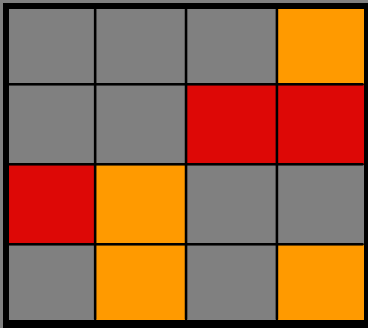
Spores in rainwater 2001-06



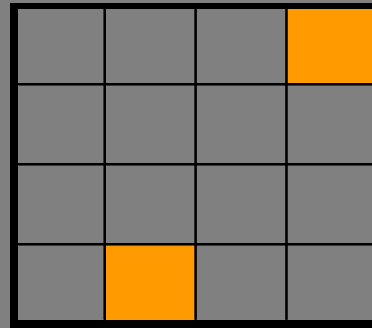


Buckets with water
and healthy bay
leaves as bait are
placed on the vertices
of a grid

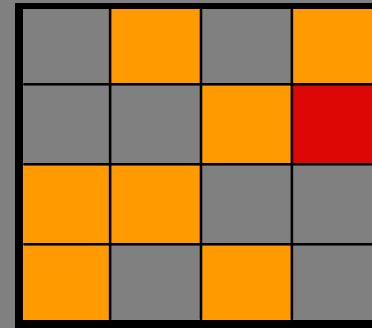
Medium (yellow) and high (red) inoculum levels in bucket baits in 8 plots in June



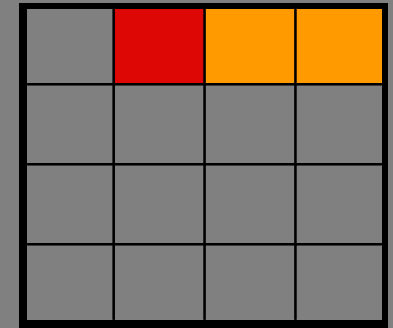
L1C



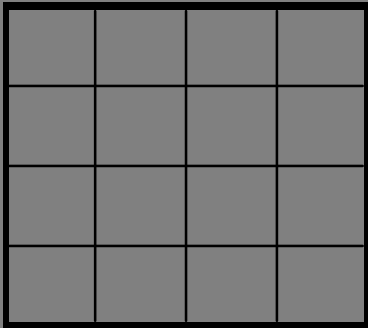
L1T



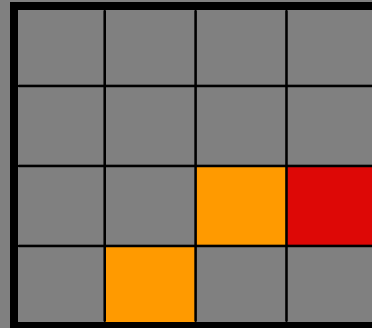
L2C



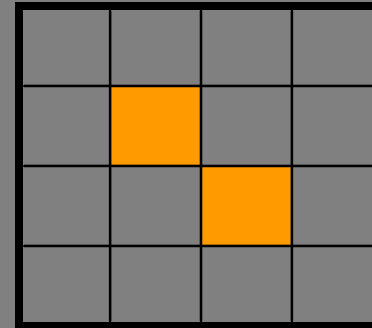
L2T



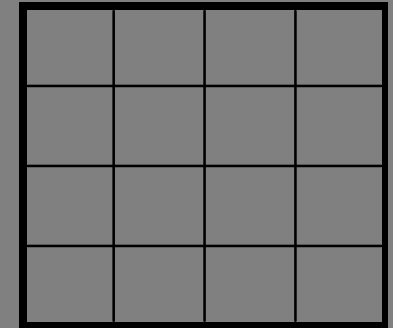
LT1C



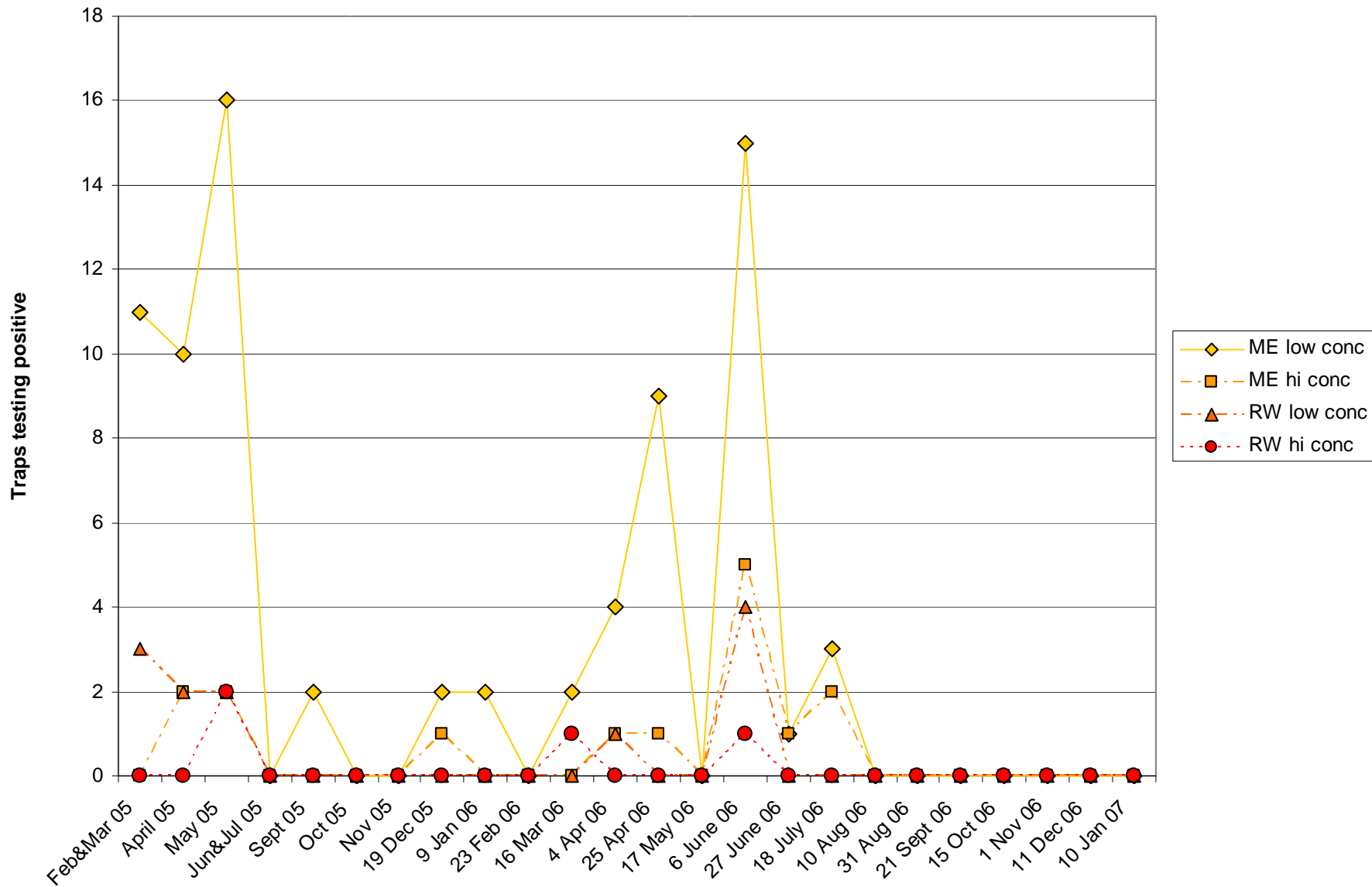
LT1T



LT2C



LT2T





Douglas-fir sapling branch tip wilted by *P.*
ramorum



Symptoms on Buckeye leaves and petioles





Autumn

Scorching of maple
leaves caused by *P.*
ramorum

Spring



Most severe levels of disease and many plants species infected

- In proximity of infected bay laurels
- Tanoaks started were reported dead in 1995
- Coast live oak and black oaks in 2000



Girdling aerial 'cankers' removed from roots



Oaks

Quercus agrifolia

Black oak

Q. kelloggii

Shreve's oak

Q. parvula var. *shrevei*

Canyon Live oak

Q. chrysolepis



Cankers end at soil line



Redwood-Tanoak forest

Tanoaks of all ages
can be infected

Tanoaks mortality
rate of adults can
locally be 100%

Tanoaks can be
infectious

Mixed-evergreen forest

Only mature oaks
are
Infected

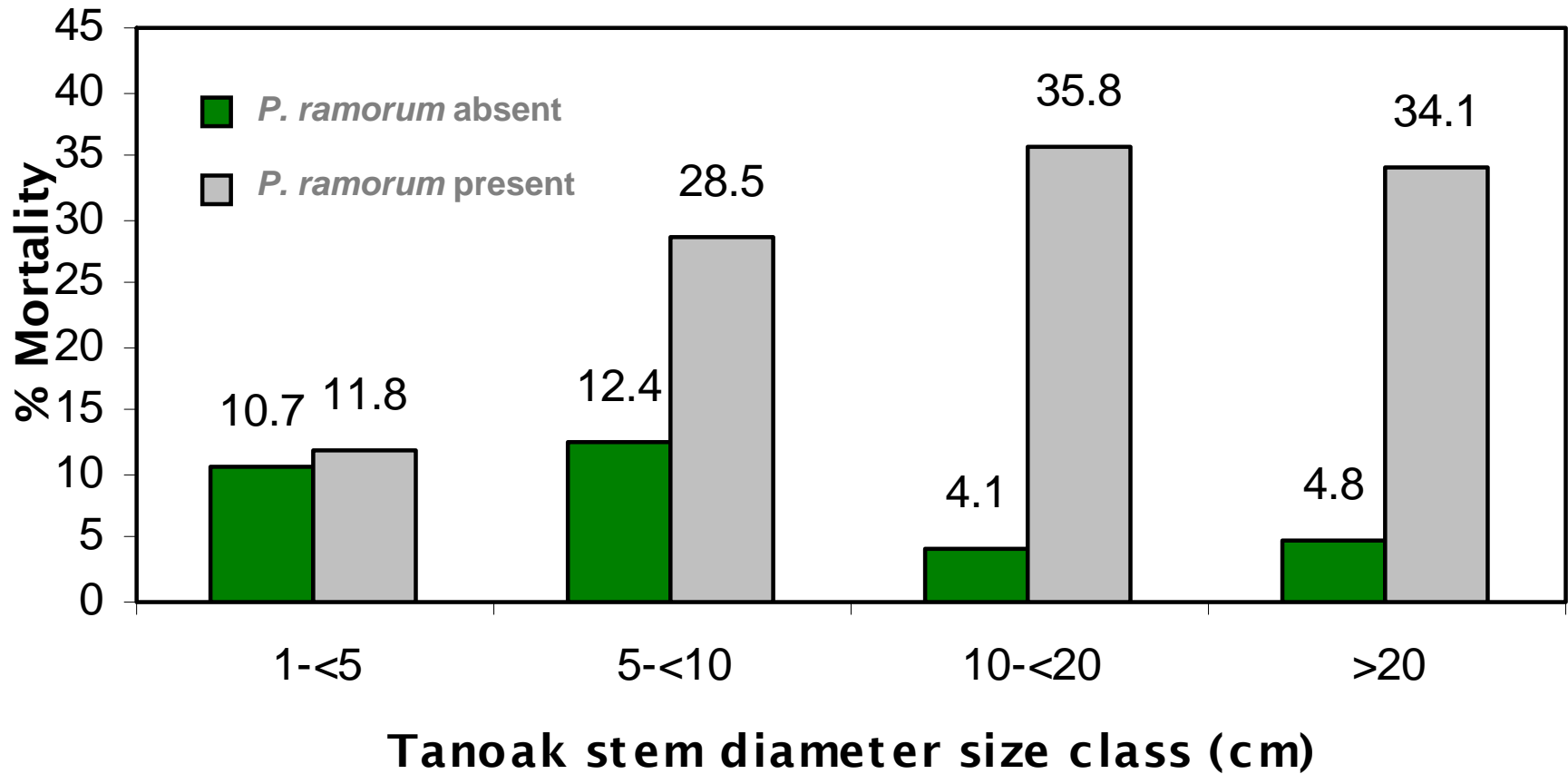
Oak mortality still
under 50%

Oaks are not
infectious and their
infection normally
occurs near other
infectious hosts and



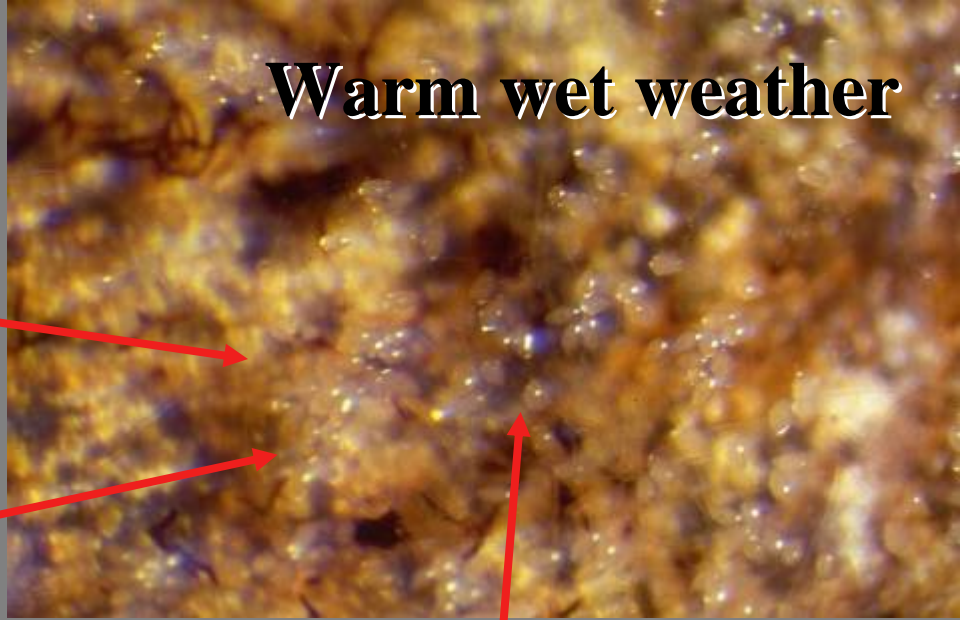
Big Sur
2006
K. Frangioso

% Mortality of Tanoak by Stem Size Class





Warm wet weather



Bay/Oak association

Bay



Coast Live Oak (no sporulation)



Canker margin in phloem



Bleeding canker



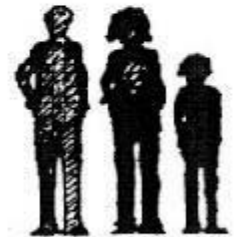
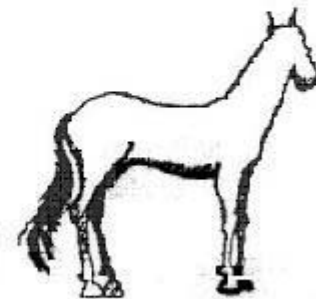
Sporangia

SOIL, WATER

Life Cycle of the West Nile Virus

SUMMER

Warm, wet weather
produces large mosquito
populations



Dead-end hosts



Some birds die

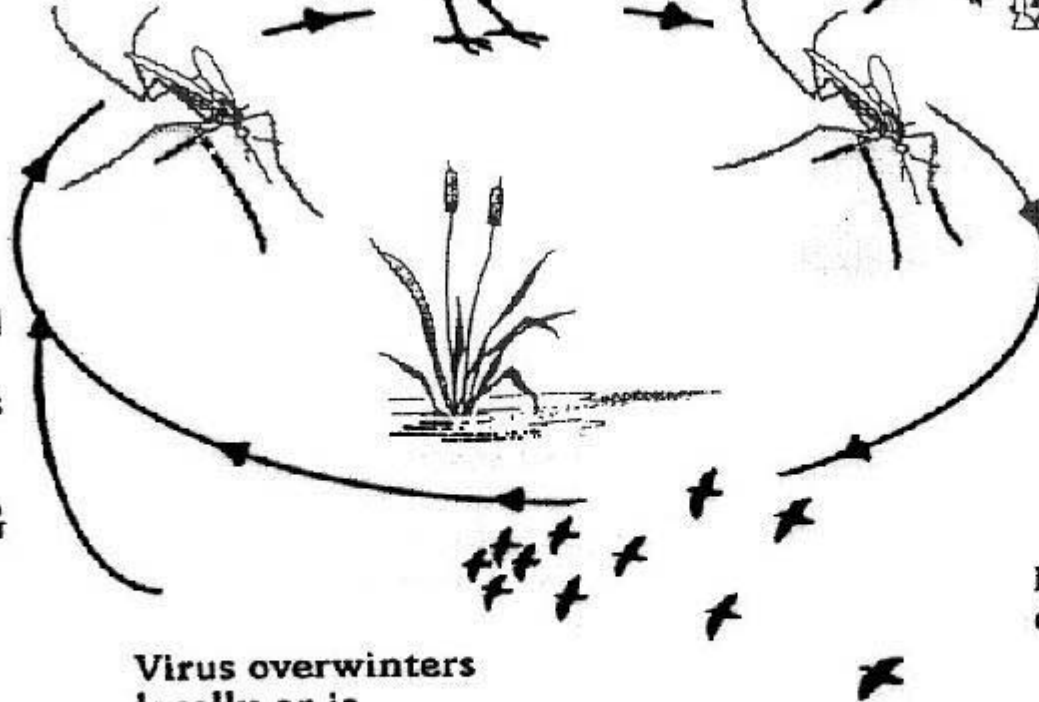
FALL

Mosquito populations
decline, birds migrate

Virus amplified
among birds
and mosquitoes

SPRING

Virus overwinters
locally or is
reintroduced



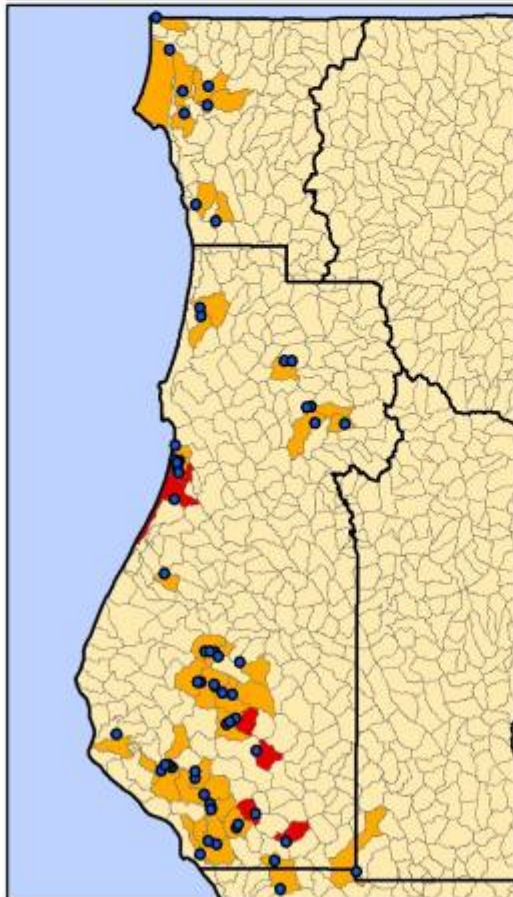
Stream Monitoring Detection of *Phytophthora ramorum* 2004-2009

Legend

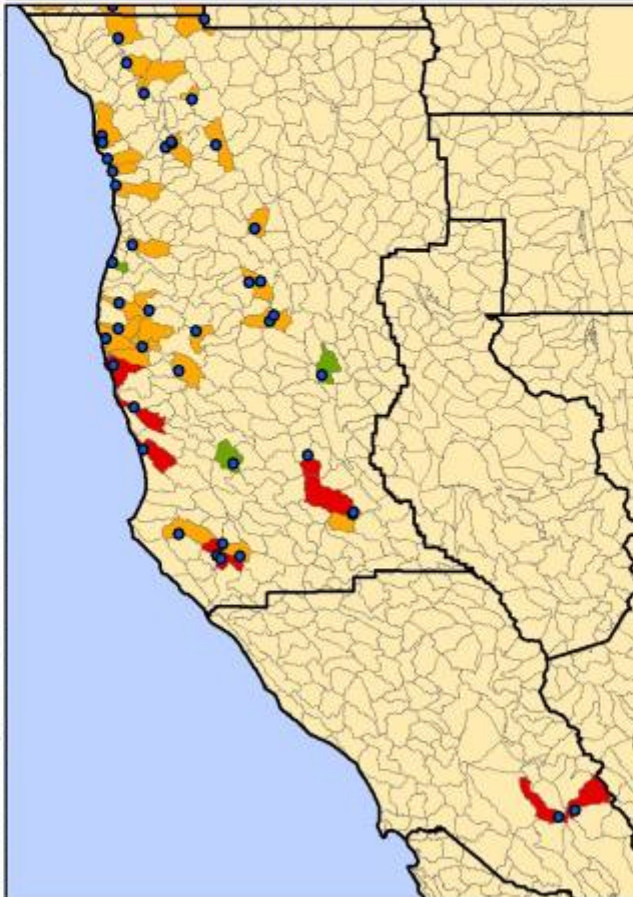
- all stream monitoring sites
- new monitoring watersheds 2010
- P. r. negative watersheds
- P. r. positive watersheds



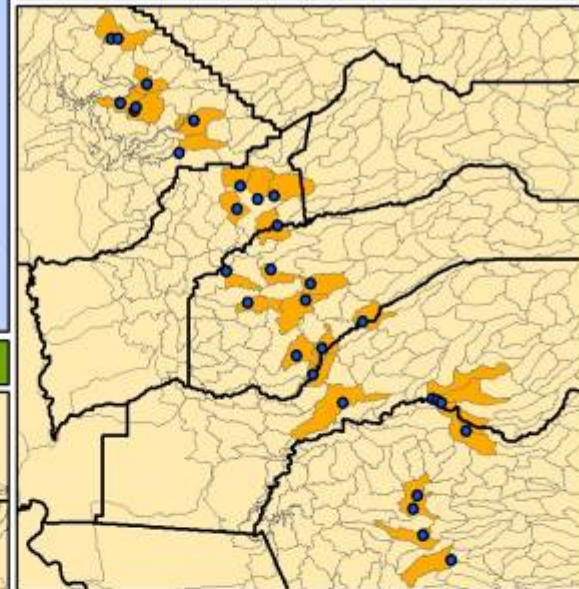
1. Del Norte & Humboldt Co.



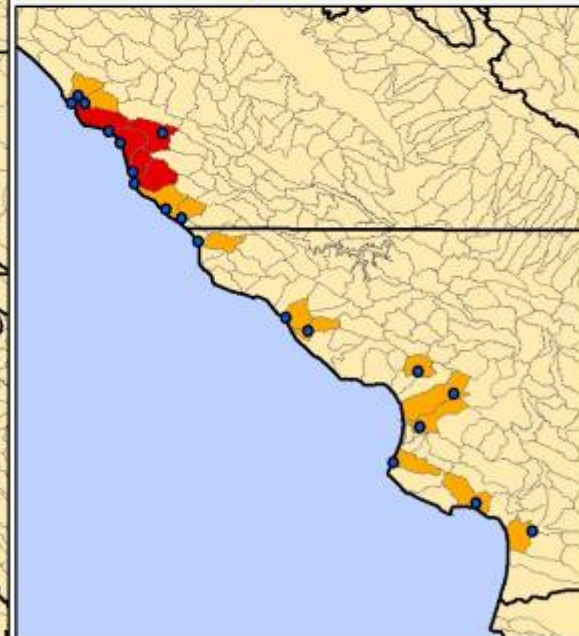
2. Mendocino & Sonoma Co.

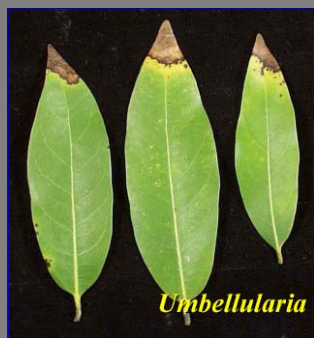


3. Northern Sierra Nevada



4. South Monterey & San Luis Obispo Co.





Host(s)

Evidence for the role of synchronicity between host phenology and pathogen activity in the distribution of sudden oak death canker disease

Richard S. Dodd¹, Daniel Huberli², Waiima Mayer¹, Tamar Y. Harnik¹, Zara Afzal-Rafii¹ and Matteo Garbelotto¹

¹Department of Environmental Science Policy and Management, 137 Mulford Hall, University of California, Berkeley, CA 94720, USA; ²Centre for Phytophthora Science and Management, School of Biological Sciences and Biotechnology, Murdoch University, Murdoch, WA 6150, Australia

Summary

P. ram active

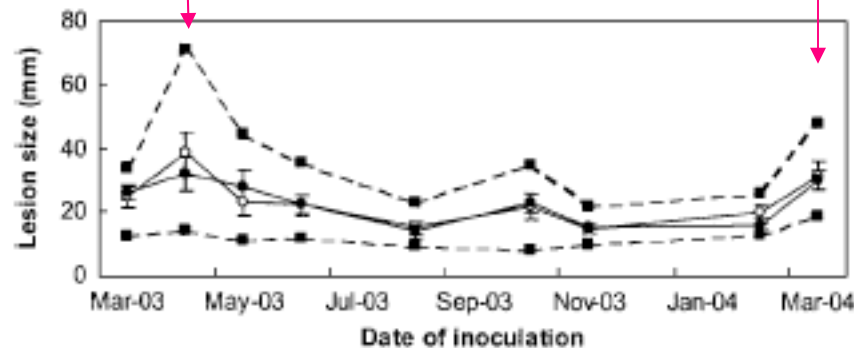


Fig. 1 Size of lesions at different inoculation dates in coast live oak (*Quercus agrifolia*) after inoculation with *Phytophthora ramorum*. Open circles, Chicken Coop site; closed circles, Miwok site. Overall maximum and minimum lesion sizes are shown as dotted lines with closed rectangles. Standard errors are shown as vertical bars.

Peaks of susceptibility correspond to peaks of cambial activity

Is variation in susceptibility to *Phytophthora ramorum* correlated with population genetic structure in coast live oak (*Quercus agrifolia*)?

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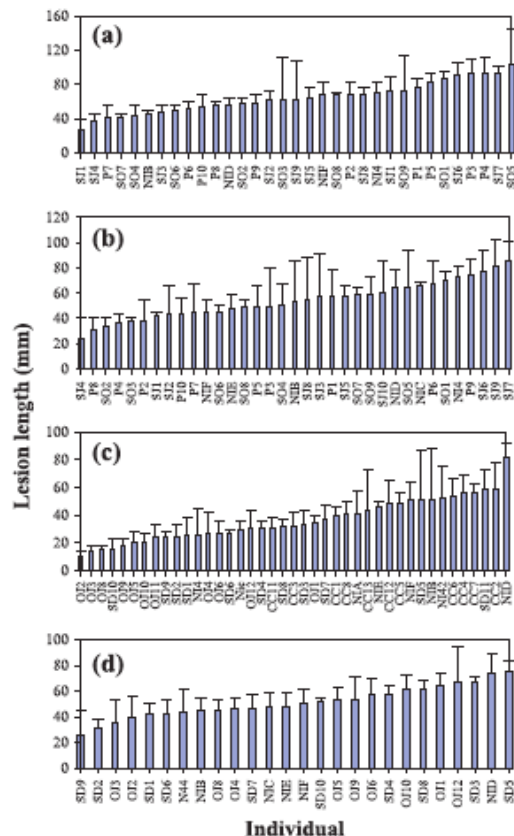


Host(s)

Significant variation in susceptibility (up to 20x) highlighted through inoculation studies

Although variation is mostly amongst individuals within a population, some populations differ as well. Population-level significance may allow us to predict local disease severity.

It is unclear what drives the observed difference (genetics vs. environment). First understanding of genetic structure of coast live oaks



Phytophthora ramorum

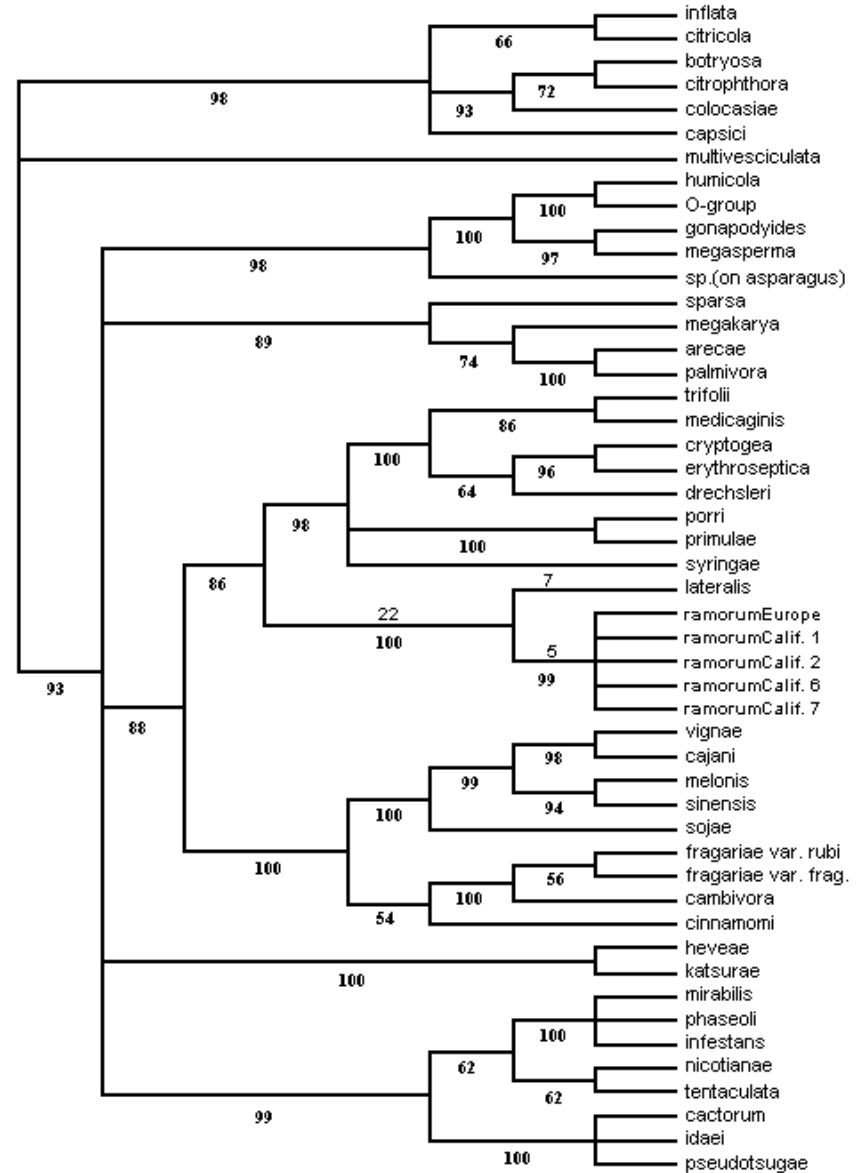
Sporangia



Chlamydospores



Phytophthora - ITS



Known Host Range of *Phytophthora ramorum*

Andrew's clintonia bead lily

Bigleaf maple

Blueblossom

California bay laurel

California black oak

California buckeye

California coffeeberry

California hazelnut

California honeysuckle

California maidenhair fern

California nutmeg

California wood fern

Camellia species

Camphor tree

Canyon live oak

Cascara

Chinese witchhazel

Coast live oak

Coast redwood

Douglas fir

Drooping leucothoe

European ash

European beech

European turkey oak

European yew

Evergreen huckleberry

False Solomon's seal

Formosa firethorn

Fetterbush

Goat willow

Grand fir

Griselinia

Holly olive

Holm oak

Horse chestnut

Hybrid witchhazel

Japanese evergreen oak

Laurustinus

Lilac

Madrone

Magnolia varieties

Manzanita

Michelia

Mountain laurel

Northern red oak

Oleander

Oregon ash

Osmanthus

Pacific yew

Persian ironwood

Pieris varieties

Planetree maple

Poison oak

Portuguese laurel
cherry

Red fir

Red tip photinia

Redwood ivy

**Rhododendron
species**

Roble beech

Rugosa rose

Salal

Salmonberry

Scotch heather

Sessile oak

Sheep laurel

Shreve oak

Southern red oak

Spicebush

Spreading
euonymus

Star magnolia

Strawberry tree

Striped bark maple

Sweet bay laurel

Sweet chestnut

Sweet Cicely

Sweet olive

Tanoak

Toyon

Viburnum varieties

Victorian box

Vine maple

**Western maidenhair
fern**

Western starflower

White fir

Winter's bark

Witch hazel

Wood rose

Yew

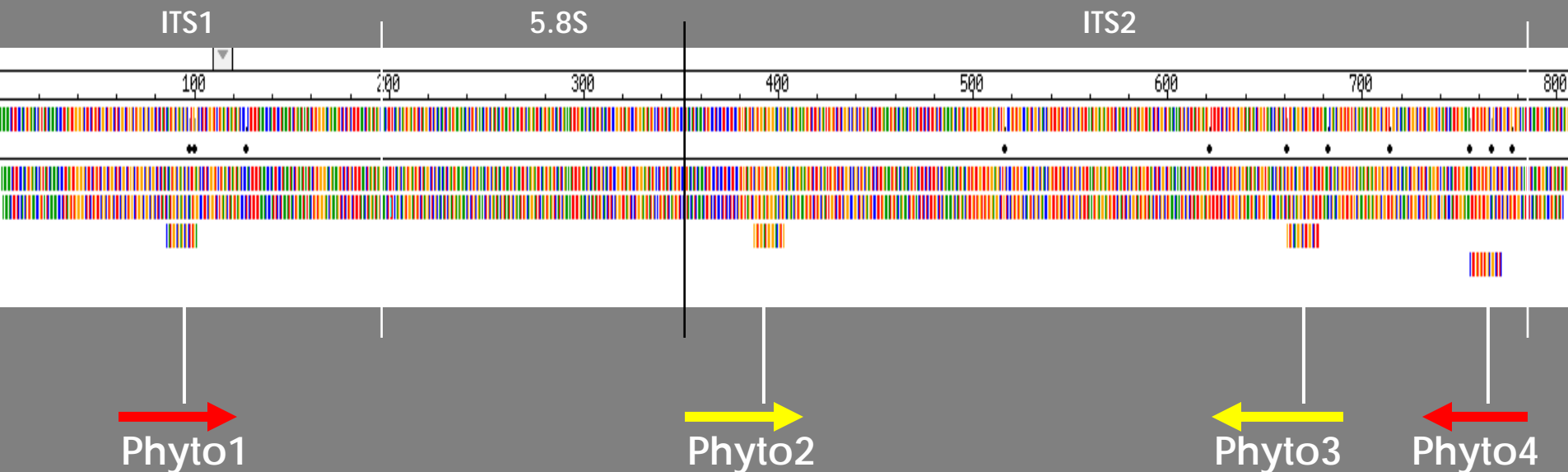
Problems

- Host lists started expanding (now over 100) in all plant families and ferns
- Symptoms looked extremely different on different hosts
- Isolation of organism from symptomatic tissue often not possible
- Isolation success extremely different in different seasons

DNA-based diagnostics

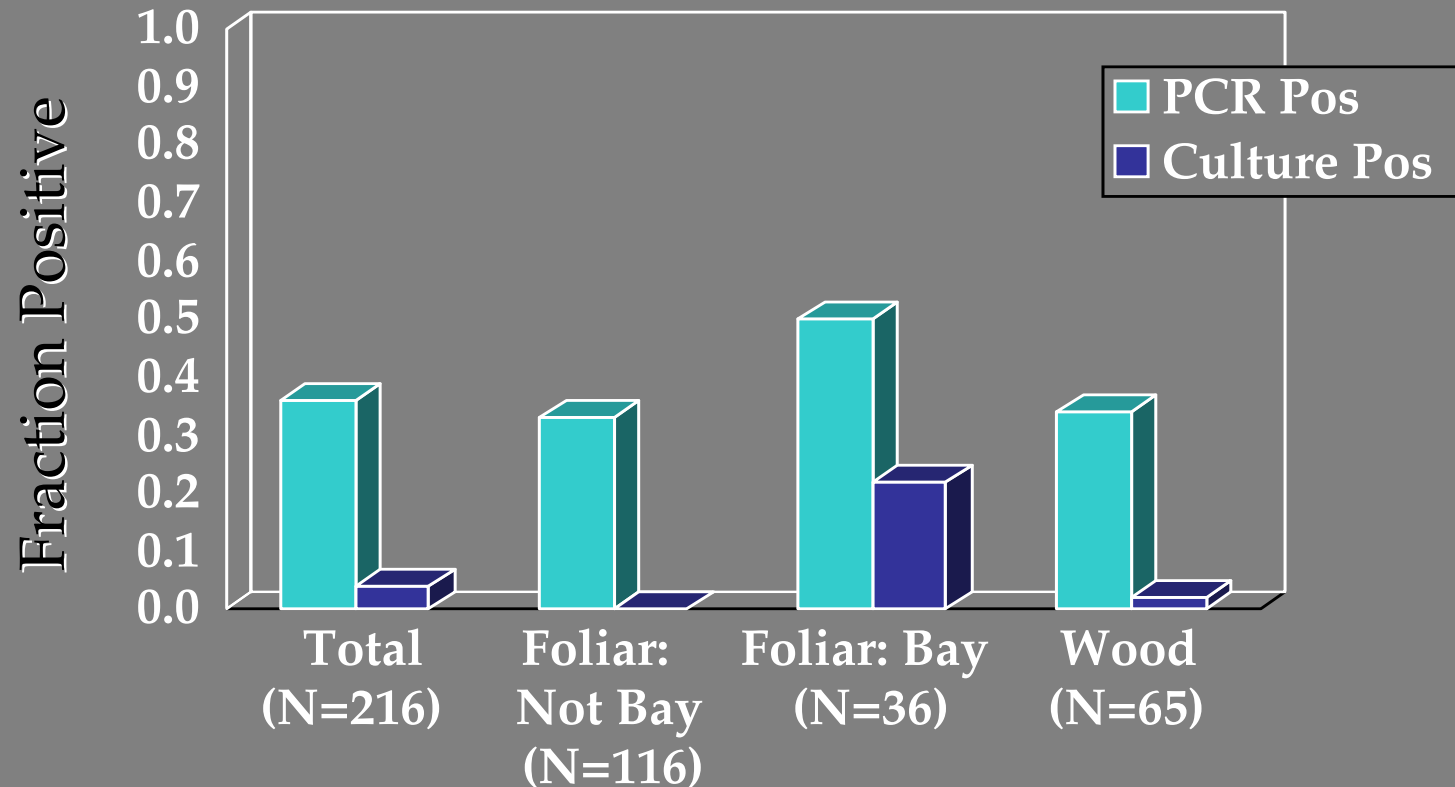
Designed 2 sets of *P. ramorum* specific primers (www primer3 software)

- phyto1-phyto4 (1st round PCR)
 - highly specific for *P. ramorum*
 - 687 bp fragment (in between red arrows)
- phyto2-phyto3 (2nd round PCR)
 - nested in phyto1-4 amplicon; specific for *Phytophthora* spp.
 - 291 bp fragment (in between yellow arrows)





Culture versus nested PCR



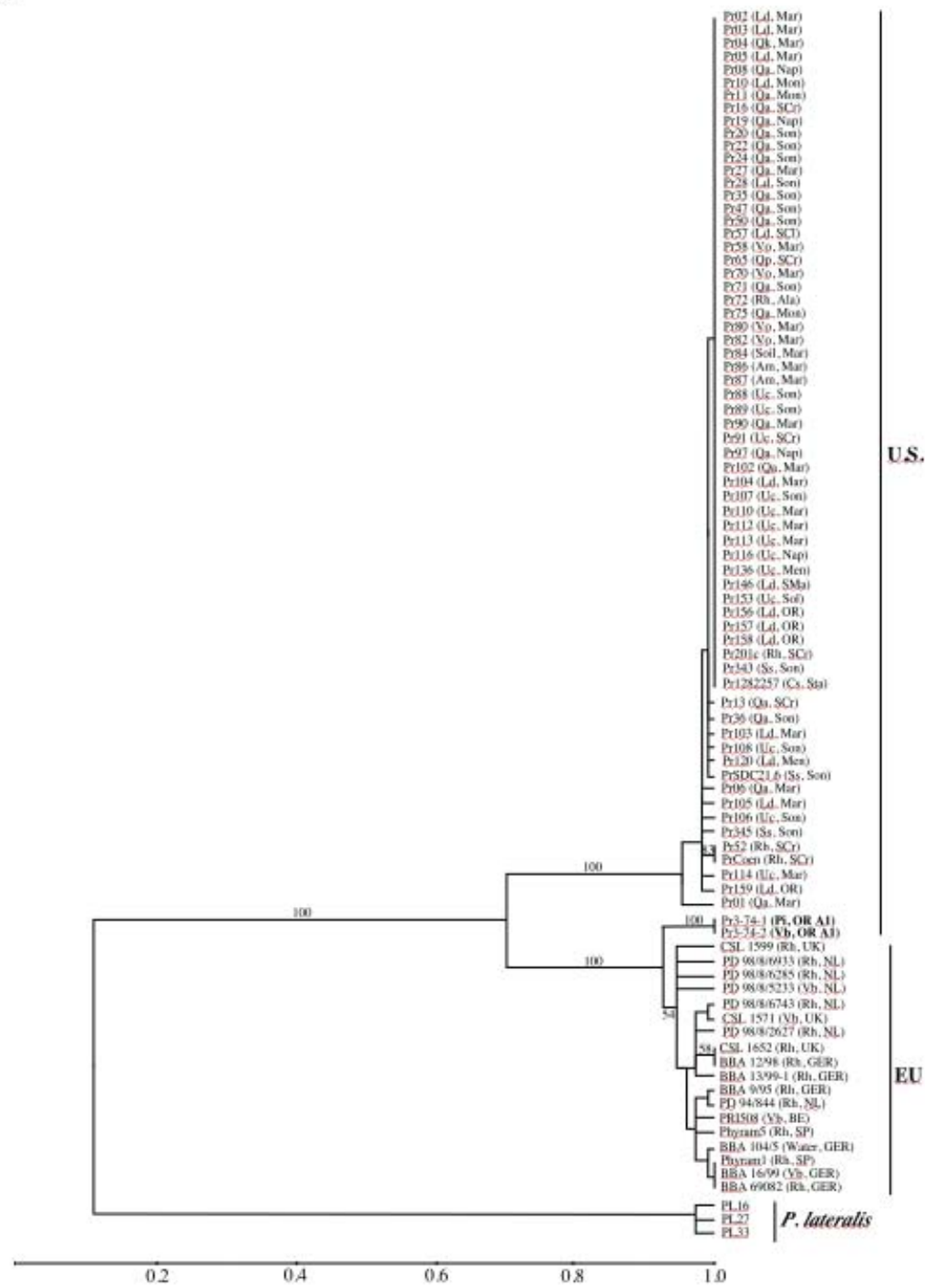
Significant effect of diagnostic type ($P < 0.001$) and sample type ($P = 0.0036$)

The assay we developed became the first DNA assay to diagnose non viral plant pathogens. Now diagnosis of most microbes will be DNA based

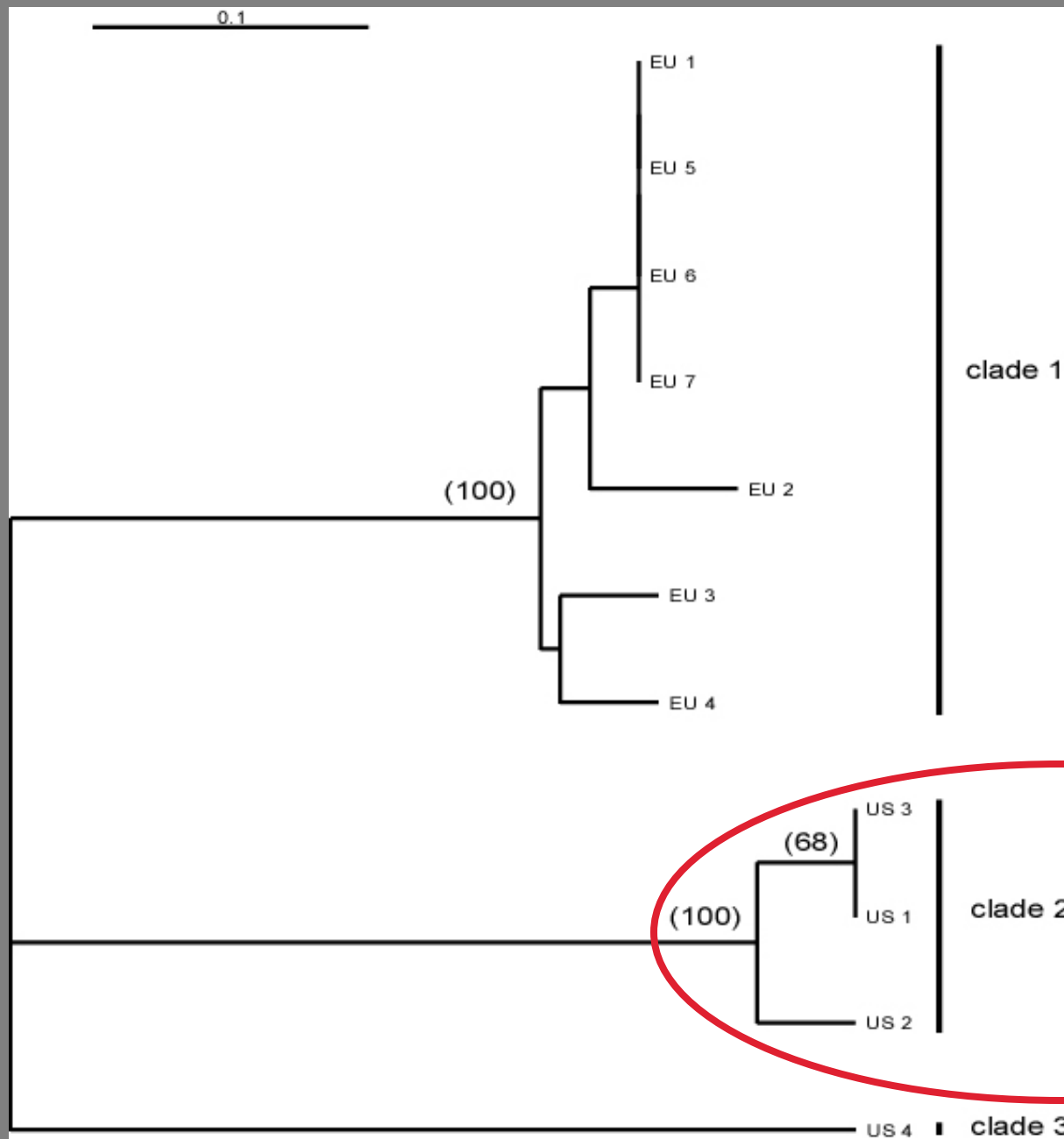
What else have we learned thanks to DNA analysis

- P. ram is introduced
- P. ram reproduces clonally in CA forests
- P. ram comes in three “varieties”
- P. ram in CA forests is not the same “variety” as in European nurseries
- P. ram is evolving through mutations
- P. ram was introduced multiple times in California from the same nursery source
- P. ram is “breathing” even if it does not grow

Fig. 1.



- US forest isolates clearly distinct from EU nursery isolates, also have different mating type
- Isolates from nurseries in WA, OR, & BC both of the US and EU types
- Potential for XXX sex and recombination in US nurseries
- US forest population is genetically very homogeneous, trademark of an introduced species



Mating
Type

Growth
Rate

A1

Fast

A2

Slow

A2

Fast

Forest structure



Wildlife



Fire



Microorganisms



EFFECTS OF SOD

- Ecological and sociological impacts: tanoaks at risk of extinction: change in forest structure and composition
- Biota linked to vanishing trees highly impacted
- Hydrogeological impact
- Climate and nutrient cycle impact
- Hazard to humans and properties
- Added cost because of intense regulation of pathogen: cleaning, prescriptions, testing
- Affecting trade

Outlook

- **Pathogen**
 - No clear trade- off between virulence and transmission. Predict an increase in virulence, already supported by some preliminary data
- **Host**
 - Bays: seem to be favored by SOD, their presence likely to increase
 - Oaks: climate change predicted to lengthen the window of maximum susceptibility
 - If resistance present it will take thousands of years to become significant, hampered by long distance host gene flow
- **Environment**
 - Warmer winters may mean infection may be rampant in winter

Yana Valachovic
UC Cooperative Extension

Sudden Oak Death in Forestlands: Management Options?

Outline

- What you can expect from this disease
- Is there hope
- Results of management trials
- Stand-scale management approaches
- Landscape management approaches

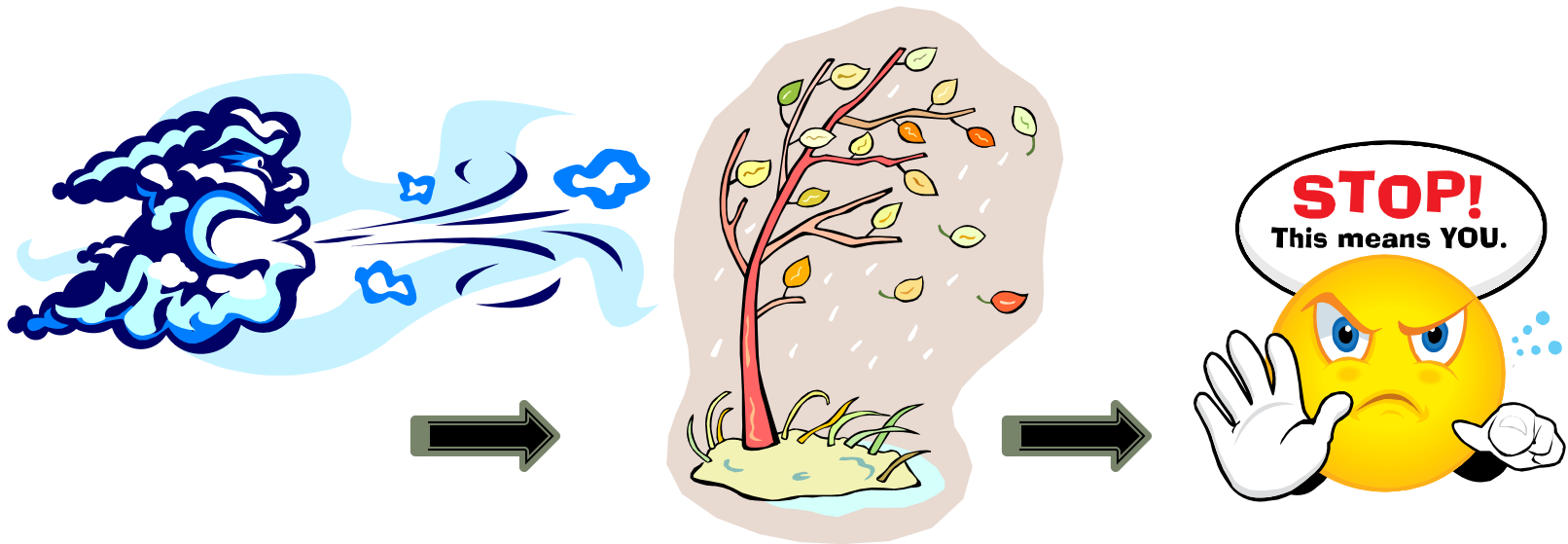
What you can expect

Tanoak mortality in high numbers

- Fuels and fire risk problems
- Regeneration problems?
- Market restrictions?
- Wildlife habitat and forage loss, how will this affect listed species?
- Loss of cultural resources
- Restrictions on movement of plant materials/soils
- Costs to businesses for regulatory compliance and sanitation
- Aesthetic issues
- Continued spread unless management occurs

Is there hope?

(infected tree)

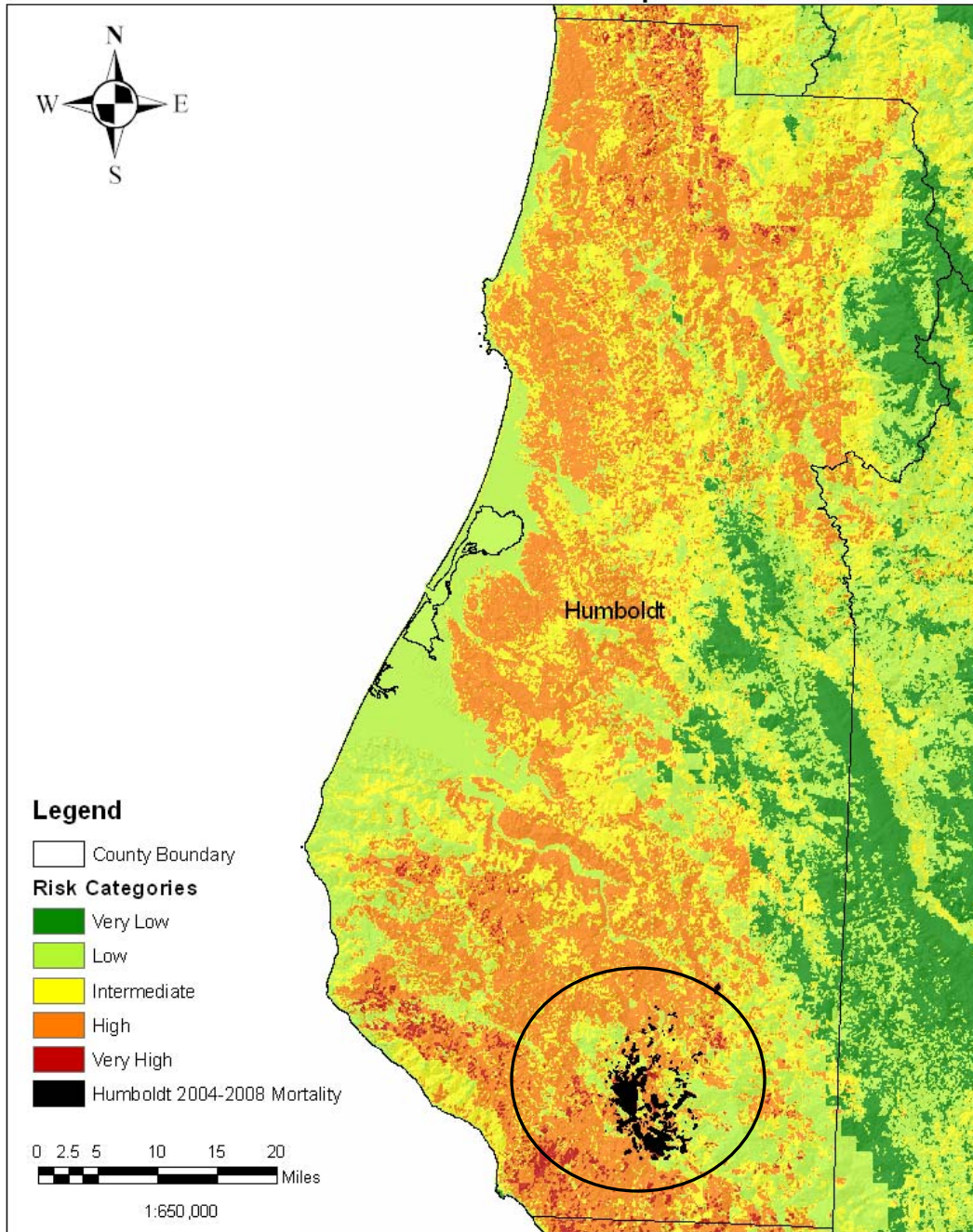


It depends, on what you are trying to achieve . Requires an understanding of the stand and the landscape dynamics .

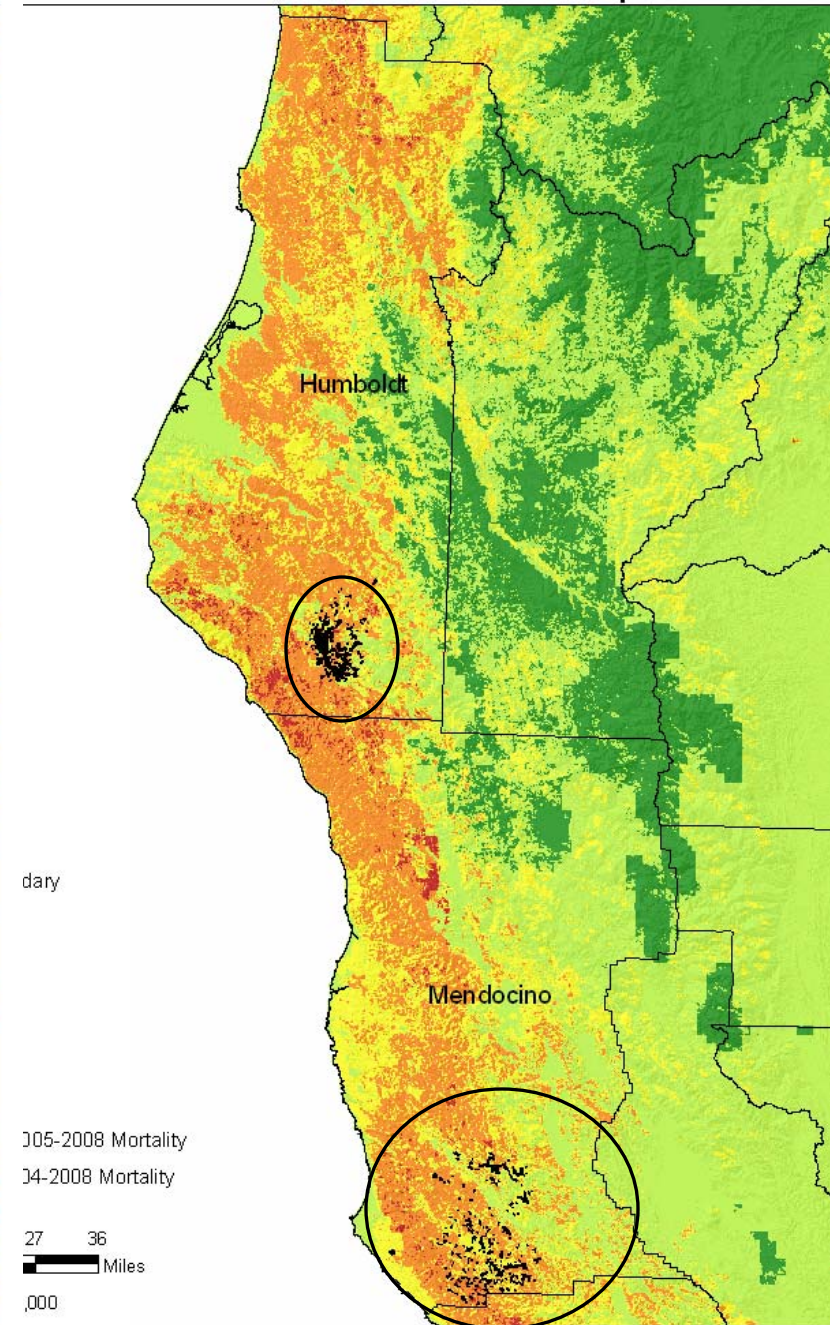
Is there hope?

- More options outside of the infested area
- Some tools are emerging within the infested areas
- Always risk and uncertainty in any treatment effort
- Not much time to take action
- Only 10% of at-risk area in CA has been impacted and much less in the north coast

Humboldt Risk Map



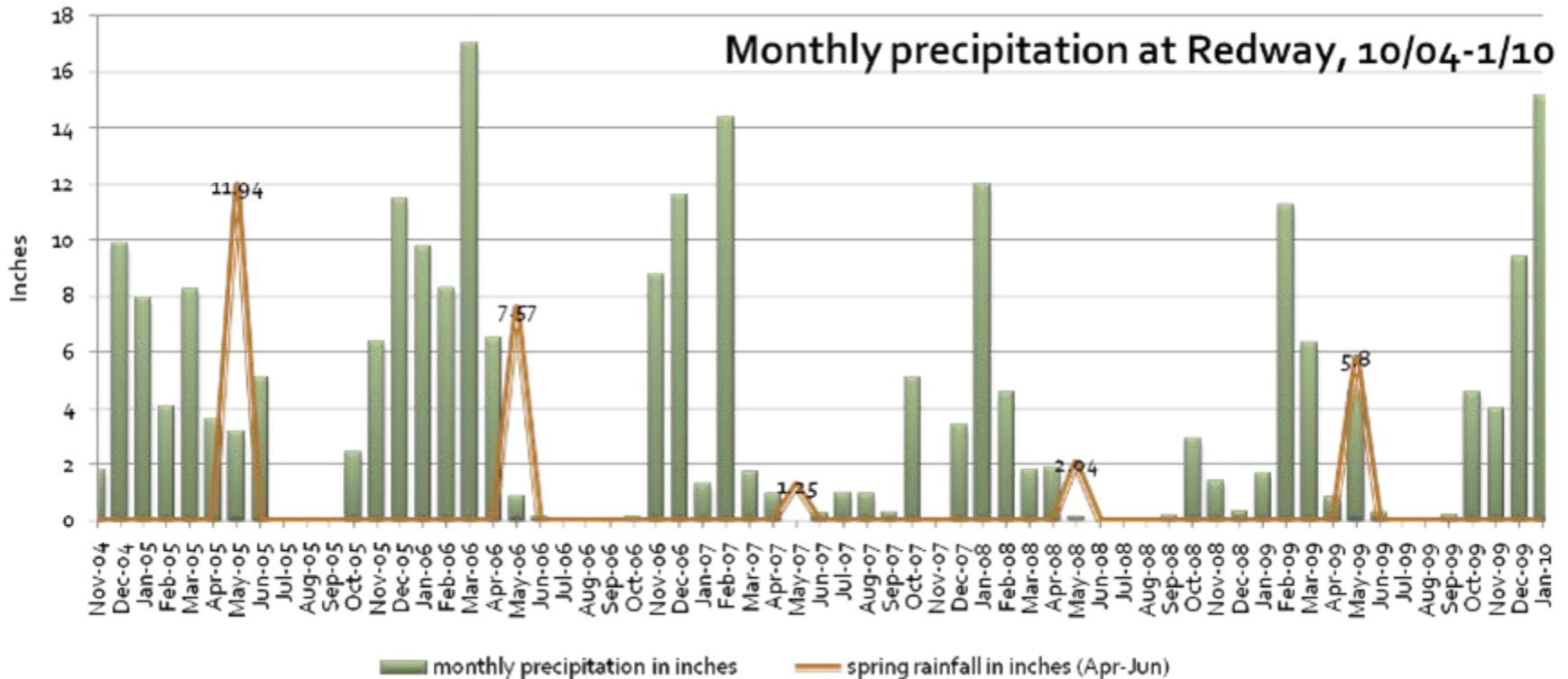
Humboldt - Mendocino Risk Map



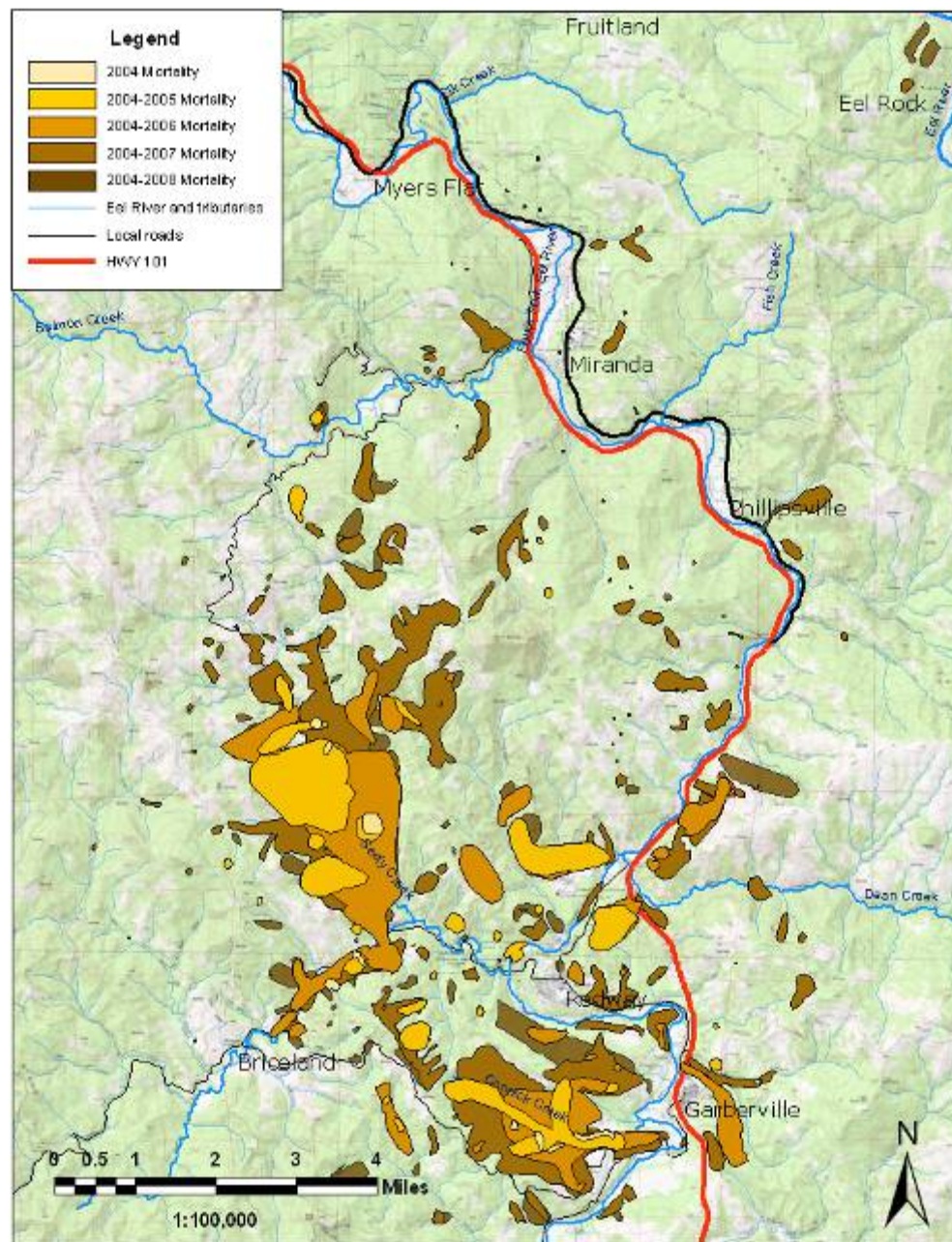
P. ramorum Management Considerations

- Treatments have to be designed to address:
 - Stand structure: pathogen needs moisture to survive
 - Species composition: favors bay and tanoak, which serve as reproductive platform (aka- seed head)
- Treatments must be responsive
 - Disease expands rapidly during wet spring conditions
- Monitoring is critical
 - Small infestations can grow rapidly
- Long-term habitat needs
 - No tanoak resistance observed to date

2005 and 2006 were highly supportive for pathogen spread, what will happen this year?



Southern Humboldt Tanoak Mortality -2004-2008



Outline

- What you can expect from this disease
- Is there hope
- Results of management trials
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- Landscape management approaches

Management Tools

Tree/ plant
removal by:

- chainsaw
- herbicides
- fire
- girdling

■ Reforestation

- planting
- promoting
seed trees

■ Agri-Fos

■ Creativity





Treatment Evaluation Process

- Bucket traps
- Soil samples
- New sprouts- bioassay

Oregon:

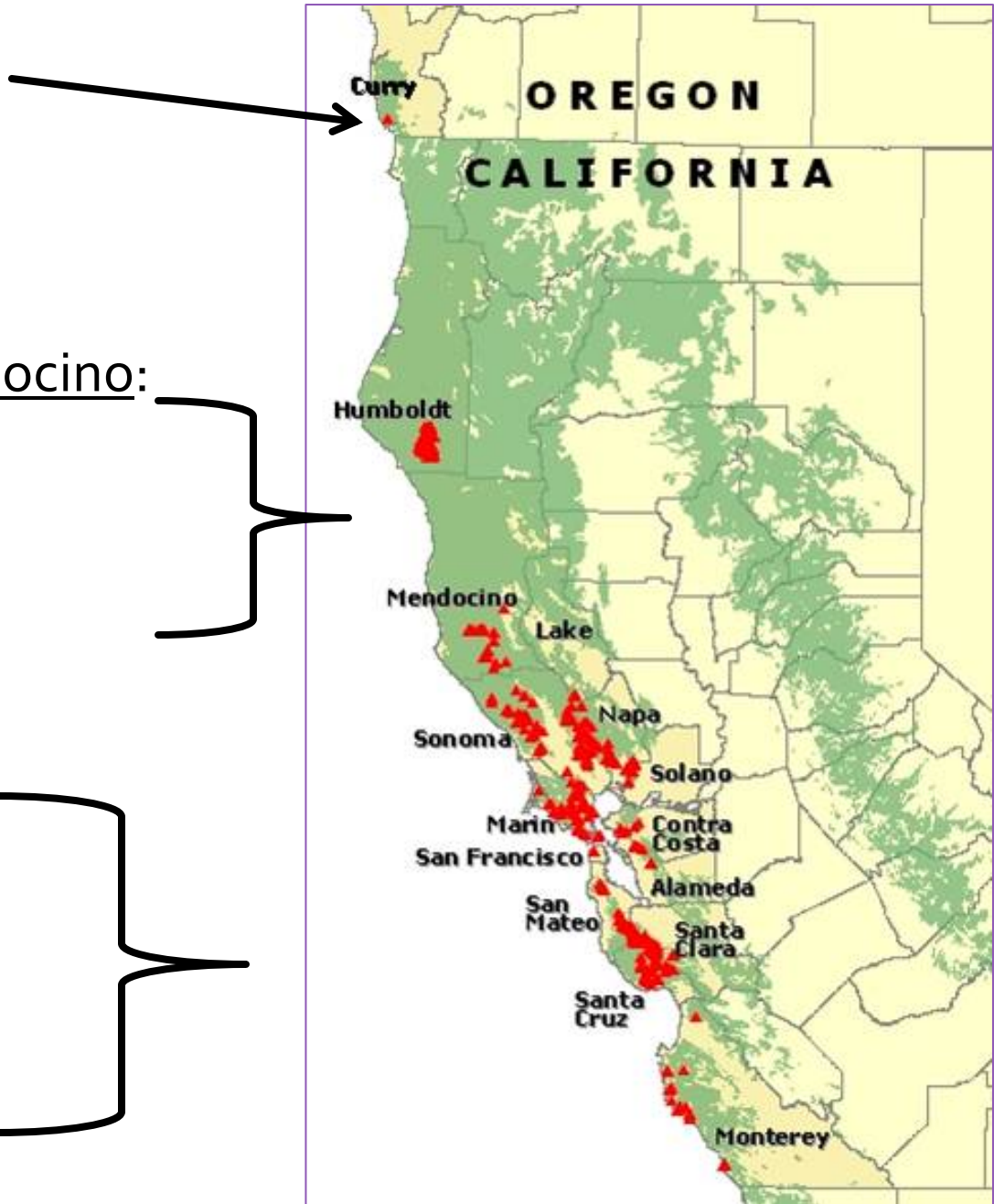
- Eradication

Humboldt and Mendocino:

- Hot spot approach
- Slow the spread
- Tanoak survival

Greater bay area:

- Impact management
- Tanoak/ oak survival



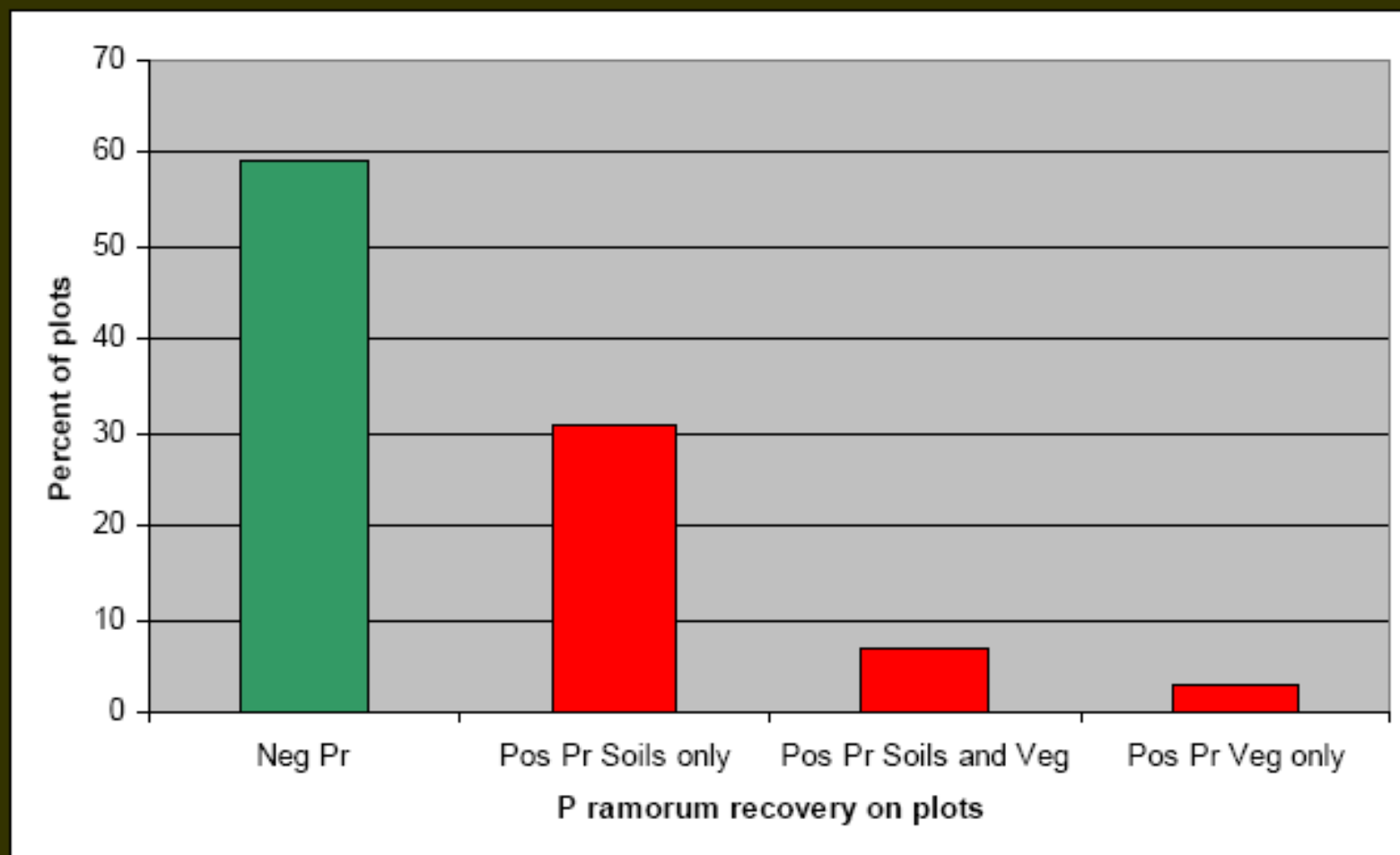
Treatment



1. Hack & squirt tanoak to prevent stump sprouting (except on BLM land)
2. Cut tanoak, rhododendron, huckleberry, sometimes myrtle.
3. Burn (piles or broadcast)
4. Plant, follow-up treatments



All Plots on Sites Treated 2001-2007 (n = 119)



59 percent plots No *Pr* detected

31 percent *Pr*+ soils* only

7 percent *Pr*+ soils* and vegetation

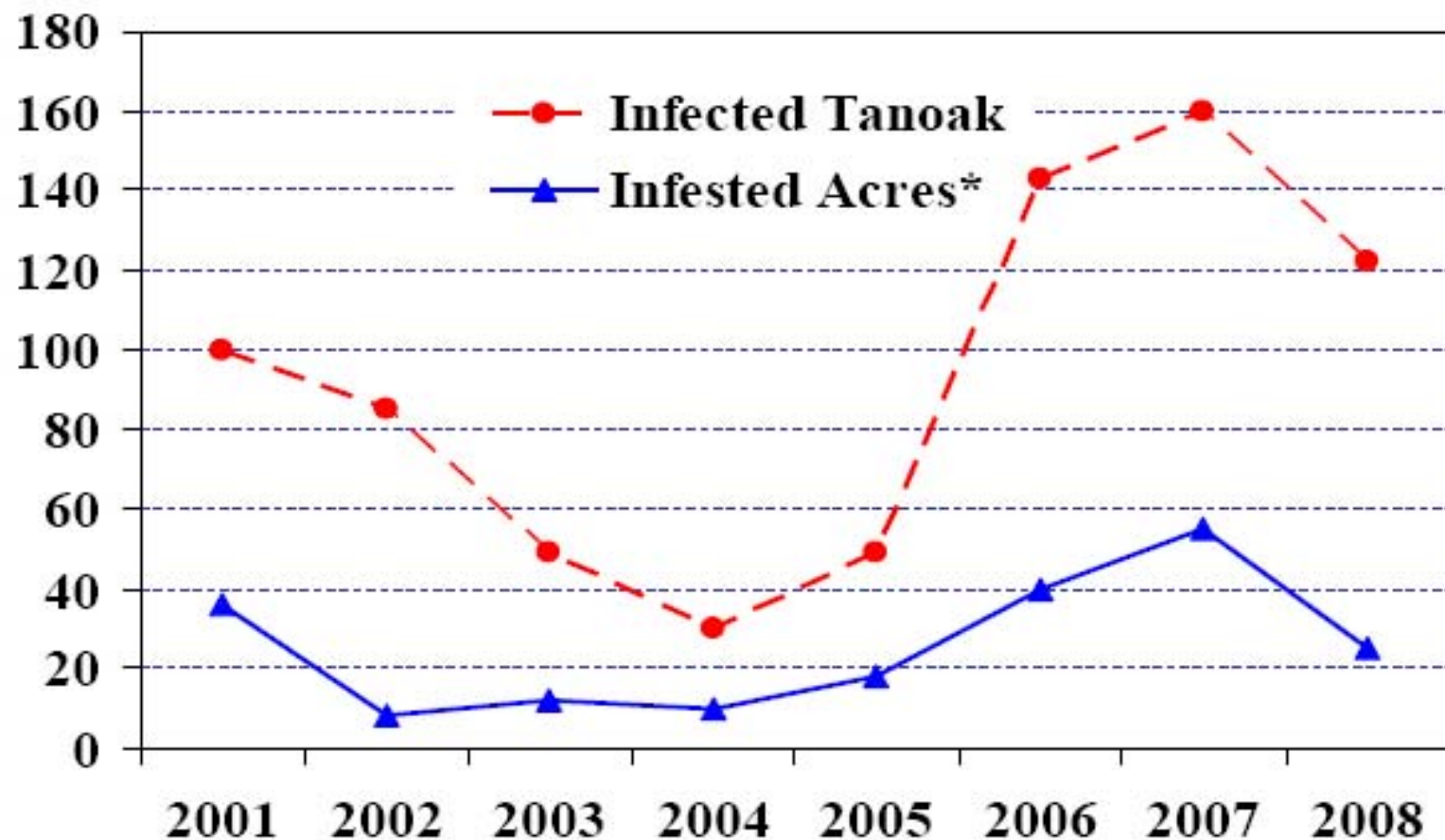
3 percent *Pr*+ vegetation only

*OSU soil baiting only

Sudden Oak Death in Oregon Forests

31 December 2008

Acres or Trees



California Approaches

<i>Goal</i>	Minimizing Property Impacts from Sudden Oak Death	Strategic Protection of Tanoak Islands, Old-Growth Tanoak, or Particular Geographic Areas	Suppression of <i>Phytophthora ramorum</i> and Limitation of Spread
<i>Treatment</i>	Dead tree removal	Manual removal of bay laurel only	Manual removal of bay laurel and tanoak (+/- prescribed underburning)
	Reforestation	Agri-fos® application	Modified fuel hazard reduction removal (+/- bay girdling)
	Maintain some tanoak with thinning (manual or by Agri-fos®)	Combination of manual removal of bay laurel and Agri-fos® application	Herbicide host removal (bay laurel and tanoak)
	Combination treatments to address site specific goals		

Known hosts in north coast

- Bigleaf maple
- Blue blossom ceanothus
- **California bay**
- California black oak
- California buckeye
- California hazel
- Canyon live oak
- Coast redwood
- Coffeeberry
- Evergreen huckleberry
- Hairy and common manzanita
- Inside-out flower
- Maidenhair fern
- Pacific yew
- Salmonberry
- Sweet cicely
- **Tanoak**
- Toyon
- Vine maple
- Western star flower
- Wood rose



Mendocino Only

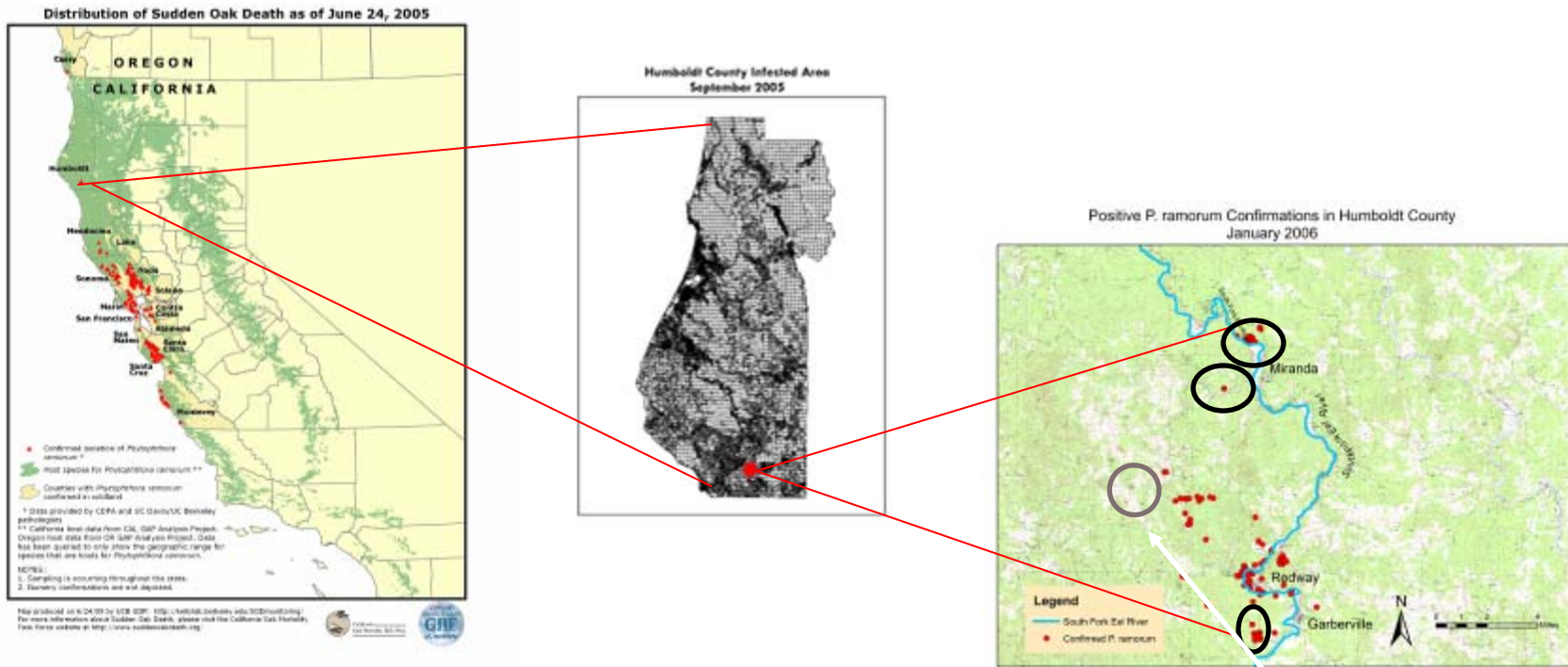
- Pacific Yew,
- Coast Live Oak
- Shreve Oak



Example of mortality observed in Southern Humboldt

Hot Spot Treatment Strategy

"Fire Control Model"



Forth project tied to a community fire protection project

Before



Before



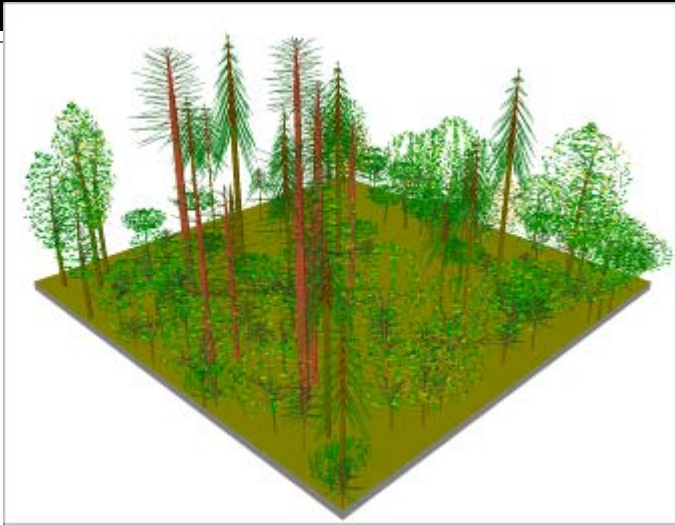
After



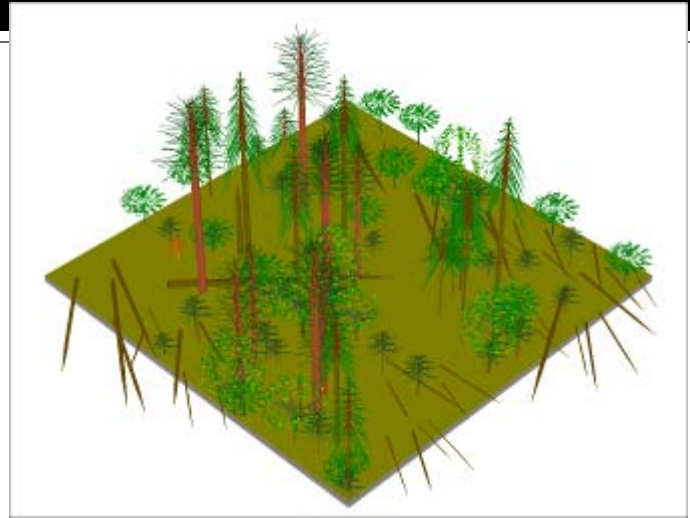
After



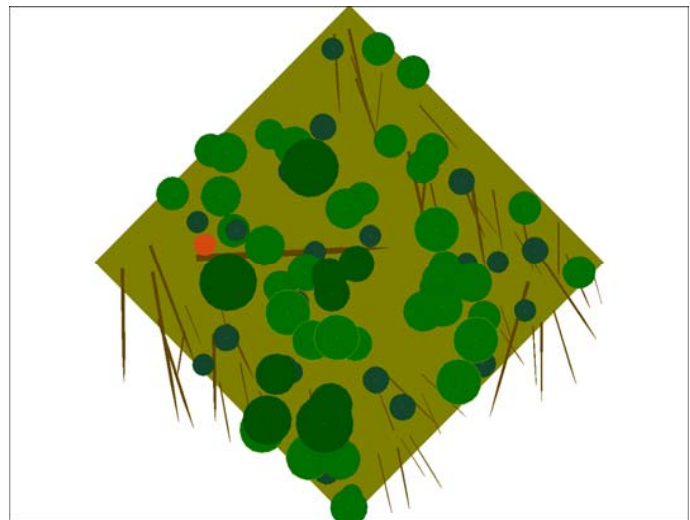
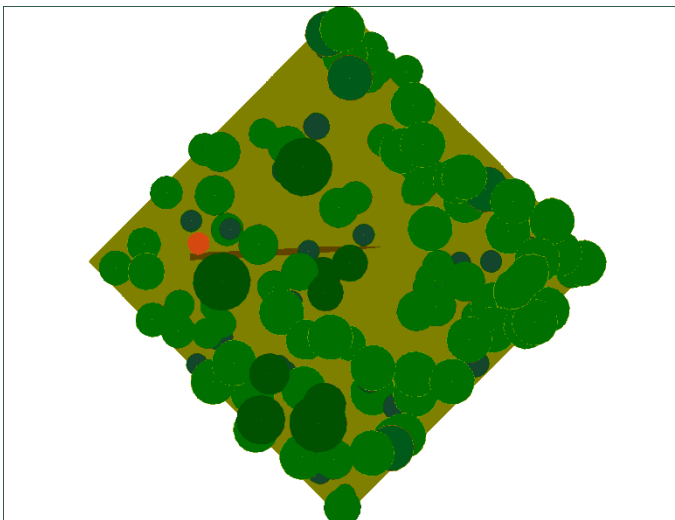
P. ramorum management



156 TPA, 187 Ft² BA, 61% Canopy Cover



108 TPA, 131 Ft² BA, 51% Canopy Cover



Bay's miracle



What does it take?



Humboldt Summary

- Data after 4 seasons post-treatment
 - Significant re-sprouting, but few symptoms observed
 - Prescribed burn sites had the most material consumed, likely more effective
 - No spread observed north of bay removal zone on Ave of the Giants
 - Evidence mounting to be able to control the pathogen at the backyard to smaller site scale
 - Bay removal appears most effective (though not appropriate in all situations). Herbicide bay removal is questionable. Imazapyr + glophosate.
 - Results are very weather dependent
 - Recommendations still very site specific and reflect landowners long-term goals.

IMG_0040





Agri-Fos

Wildland trials

Six counties, treatment by
Agri-Fos versus control

Island treatment strategy,
assessing treatment in two
old-growth stands of
tanoak



Outline

- What you can expect from this disease
- Is there hope
- Results of management trials
- Stand-scale management approaches
 - What is practical?
- Landscape management approaches

Stand-scale approaches

- Prioritize your property
- What is most at risk, least at risk?
- Are you inside or outside of the infested area?
- Work strategically
- Don't bring it home

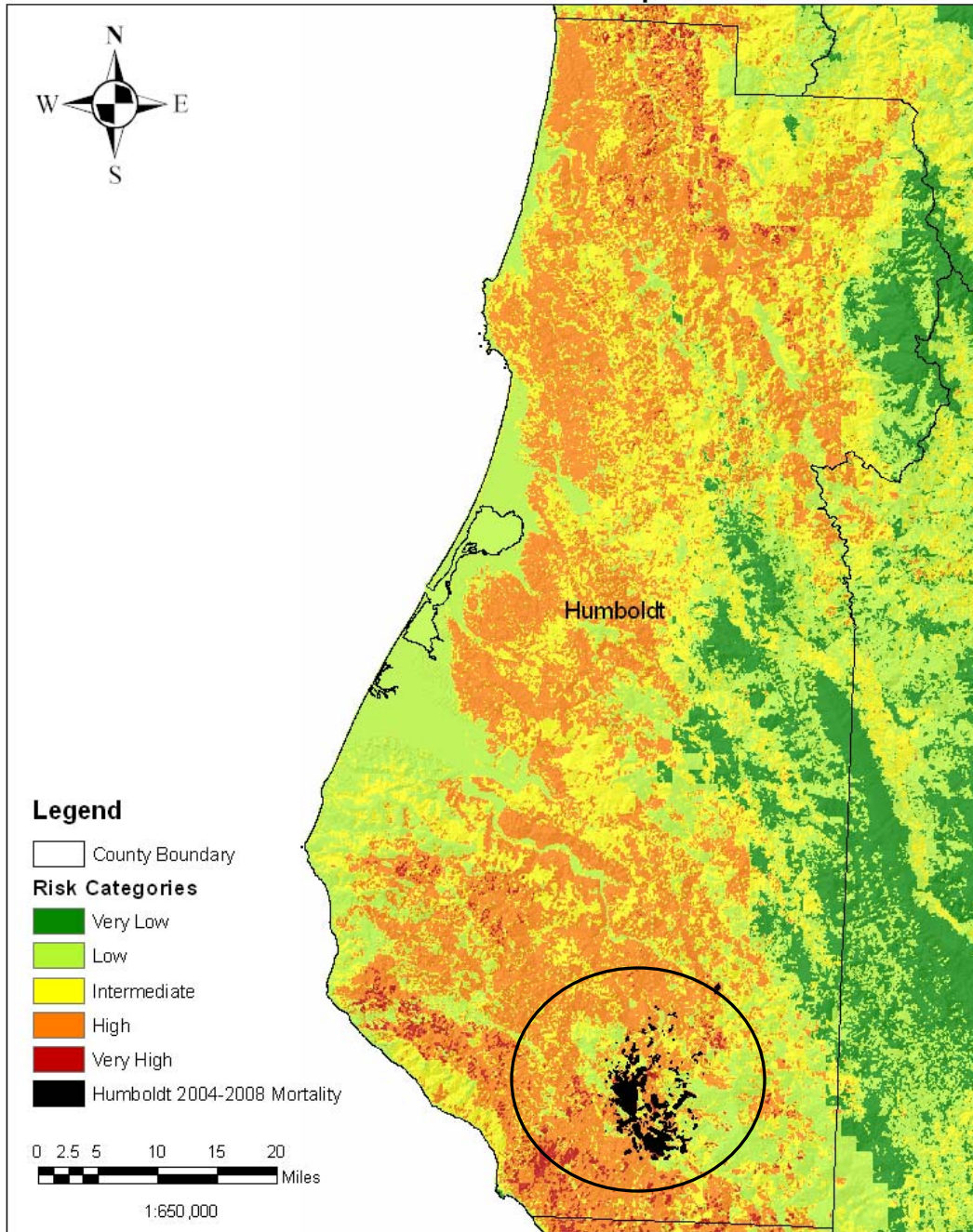
Management options depend on goals and stand conditions

- Remove infected hosts to stop spore production
- Change species composition
- Change stand structure and canopy cover, to dry a location
- Use Agri-fos to protect high value tanoak or oak trees
- Remove bay adjacent to high value tanoak or oak trees, stump treat bay to prevent sprouting
- Install a reduced or no-host zone to prevent tree to tree spread
- Integrate SOD management into other activities

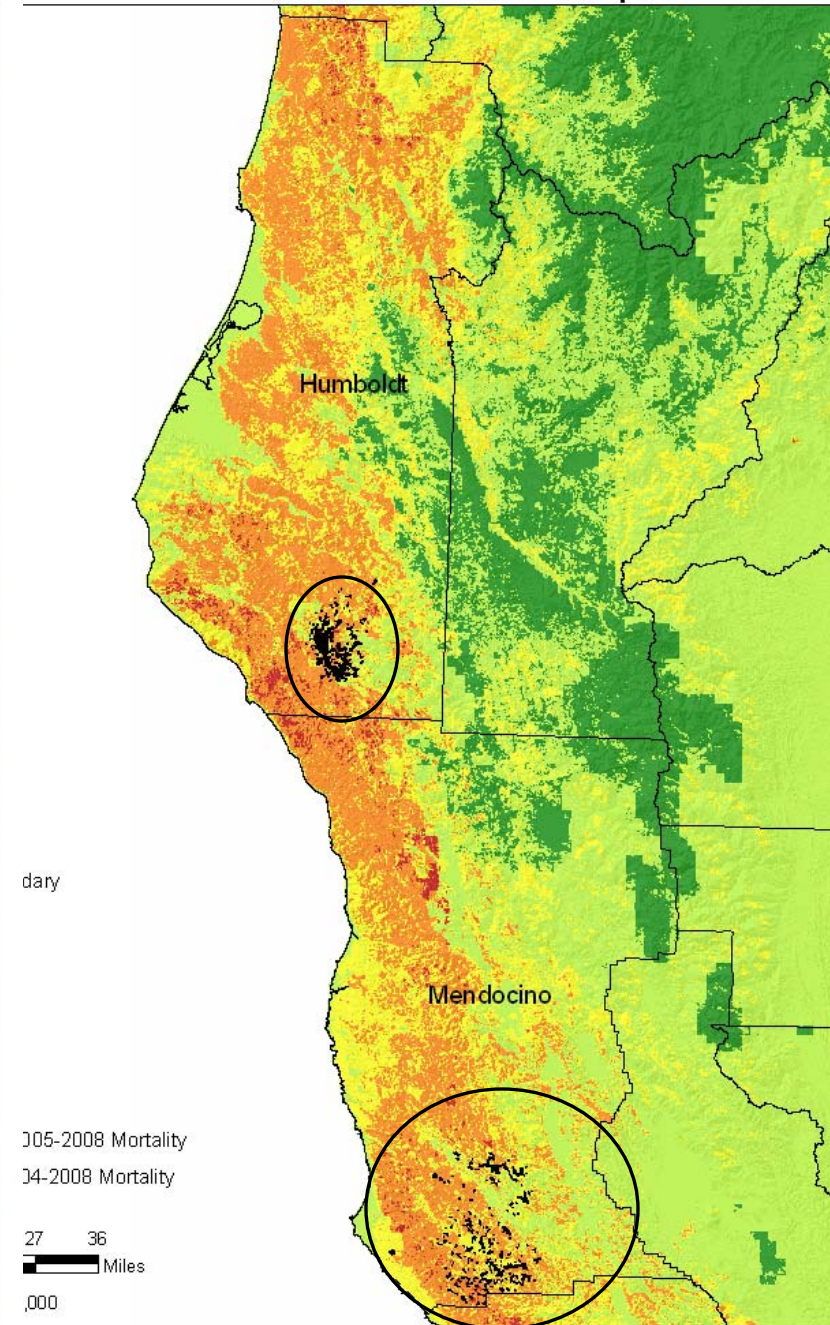
Outline

- What you can expect from this disease
- Is there hope
- Results of management trials
- Stand scale management approaches
- Landscape management approaches
 - Hot-spot approach?
 - Coordinated action?
 - Regulatory support for action?

Humboldt Risk Map



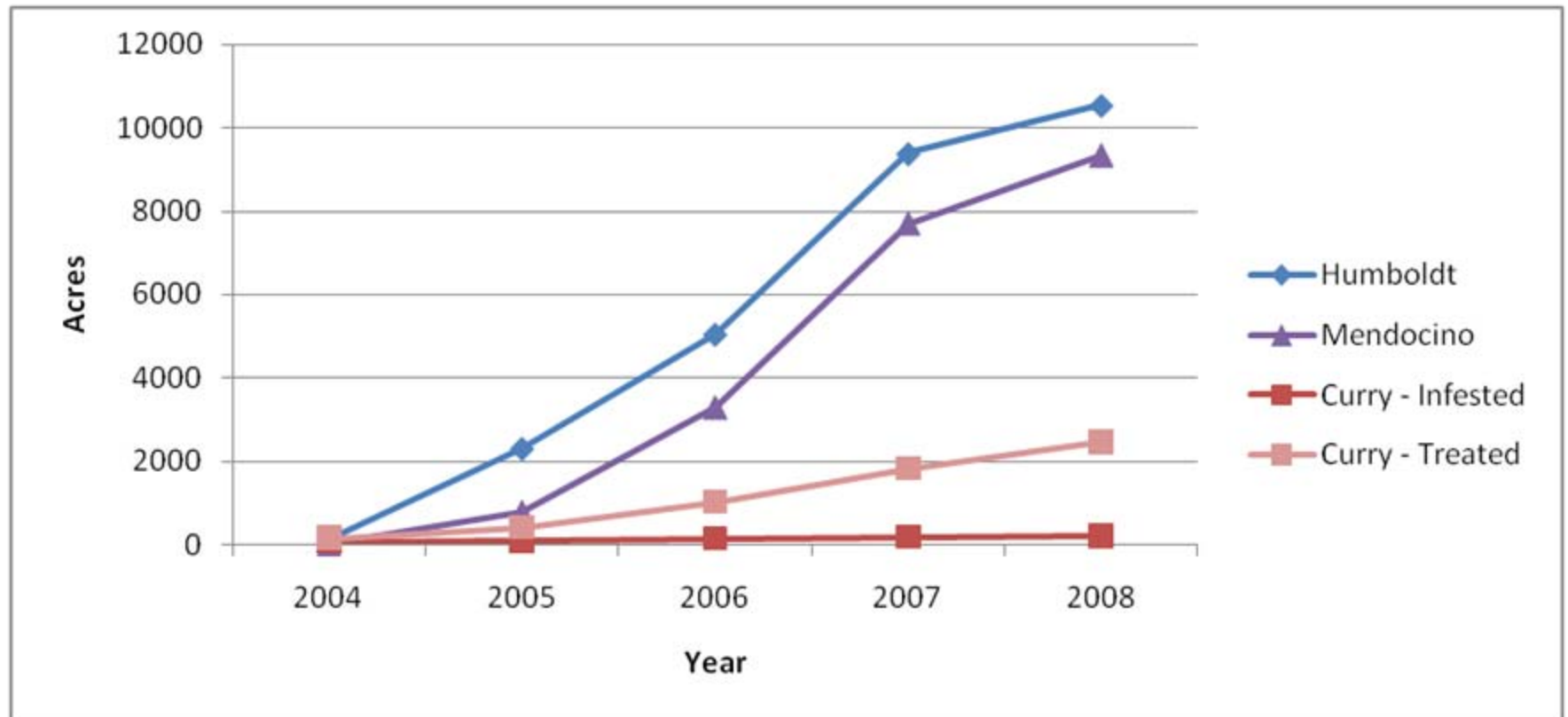
Humboldt - Mendocino Risk Map



Tools in the Tool Box

<i>Goal</i>	Minimizing Property Impacts from Sudden Oak Death	Strategic Protection of Tanoak Islands, Old-Growth Tanoak, or Particular Geographic Areas	Suppression of <i>Phytophthora ramorum</i> and Limitation of Spread
<i>Treatment</i>	Dead tree removal	Manual removal of bay laurel only	Manual removal of bay laurel and tanoak (+/- prescribed underburning)
	Reforestation	Agri-fos® application	Modified fuel hazard reduction removal (+/- bay girdling)
	Maintain some tanoak with thinning (manual or by Agri-fos®)	Combination of manual removal of bay laurel and Agri-fos® application	Herbicide host removal (bay laurel and tanoak)
	Combination treatments to address site specific goals		

Curry, Humboldt and Mendocino Acres



P. ramorum management

- Remember that it has mostly been a disease of the unmanaged landscape!
- Coordinated action in Oregon is producing results
- We still have options in the north coast
- We need to work together to have regulatory support to take action
- Monitoring disease development is essential
- We are developing forecasting tools, to help managers make decisions

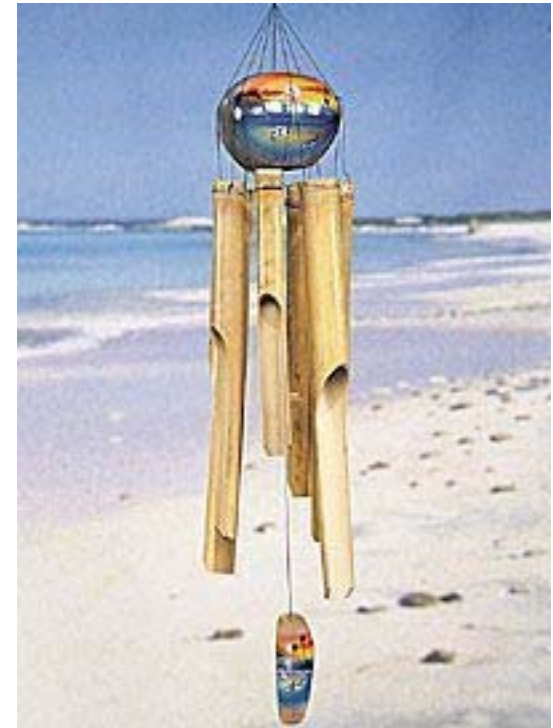
Acknowledgements

- Funding
 - USDA FS State and Private Forestry; PSW Research Station
 - CDF's California Forest Improvement Program and Vegetation Management Program
 - Bureau of Land Management
- In kind donations of labor and technical expertise
 - Cal Fire, UCCE, DPR, Cal Trans, and several small landowners.
- Organizations Involved
 - Treatment- Southern Humboldt Fire Safe Council/Institute for Sustainable Forestry, USFS State and Private aerial detection staff, DFG, USFWS, several private vegetation management contractors, and consulting foresters
 - Monitoring- Hoopa and Yurok Tribes, Redwood National Park, Bureau of Land Management, Mattole Restoration Council, UC Davis, UC Berkeley, Sonoma State

Ways that are being investigated to control *P. ramorum*

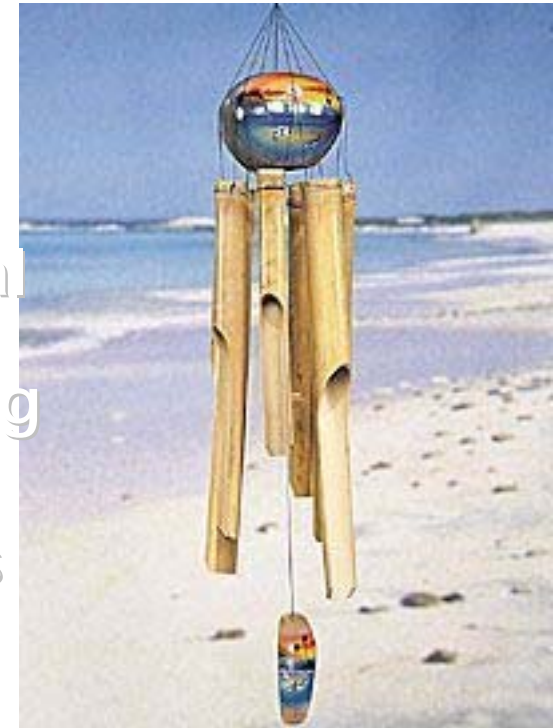
- Wind Chimes
- Holy Water
- Banning cell phone use
- Using dark energy from outer space
- Hugging trees

- Chemicals
- Natural resistance



Ways that were suggested to us to control *P. ramorum*

- Diagnosis
- Predictions/modeling
- Sanitation: soil/water/plant material
- Reducing risk: pruning and wounding
- Stand composition: removal of bays
- Preventive chemical treatment
- Natural resistance



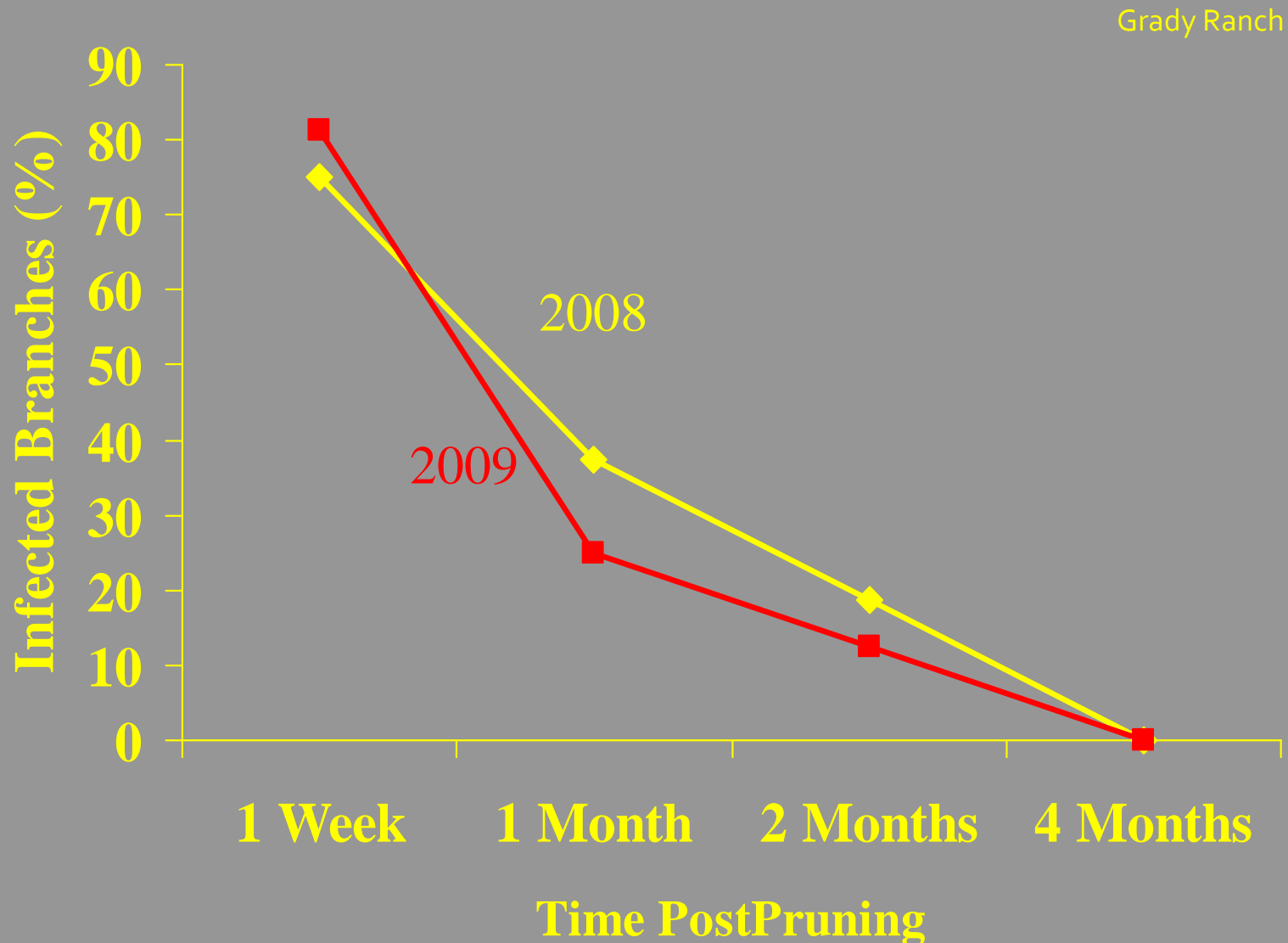
Transmission of SOD through pruning:

- ▶ Does tree pruning introduce SOD infection?
- ▶ Is the timing of cutting a factor?



Skywalker / Grady Ranch Site

Transmission of SOD Through Pruning



AgriFos and PentraBark

Topical Application



+

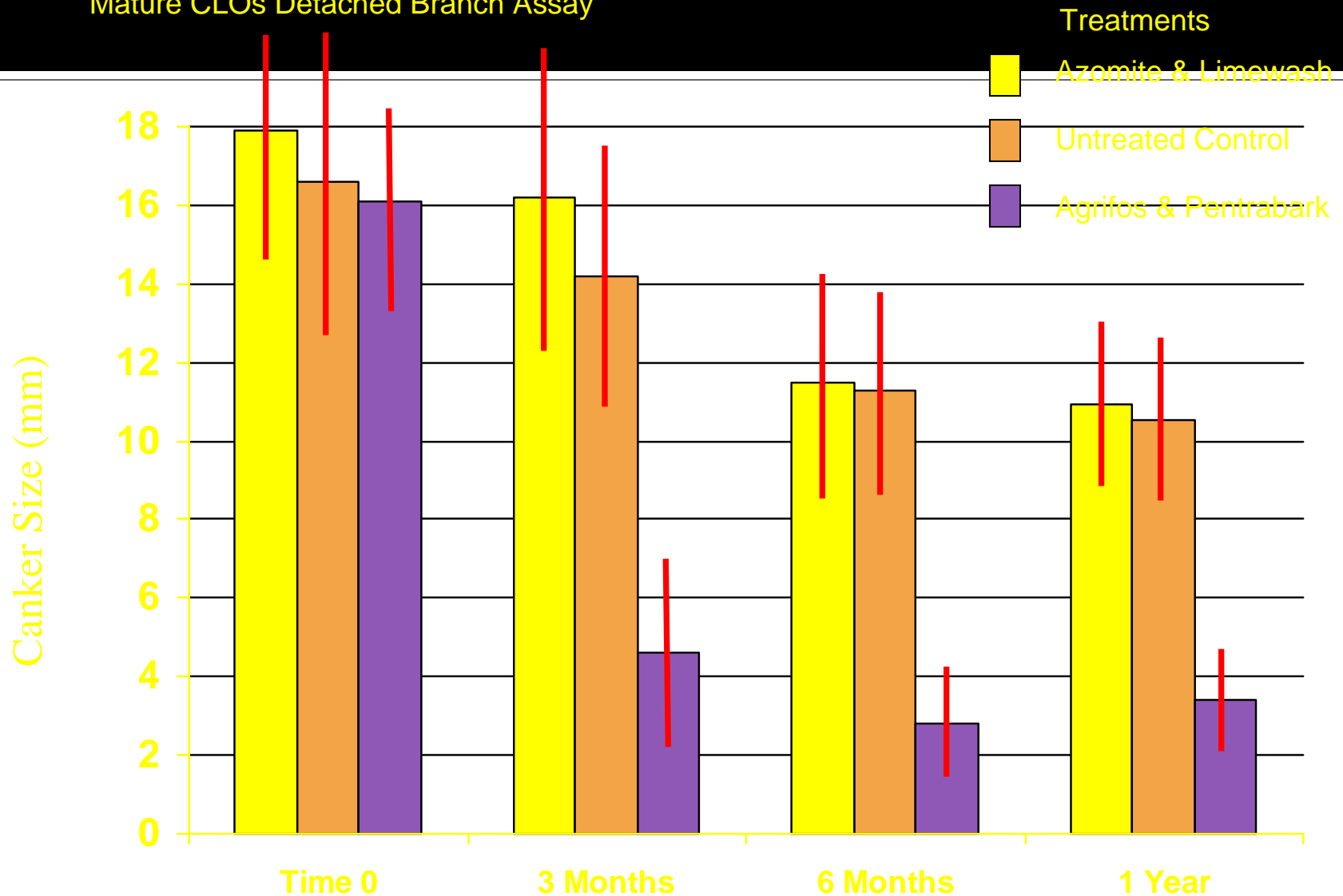


Azomite Soil Sweetener and Limewash Application



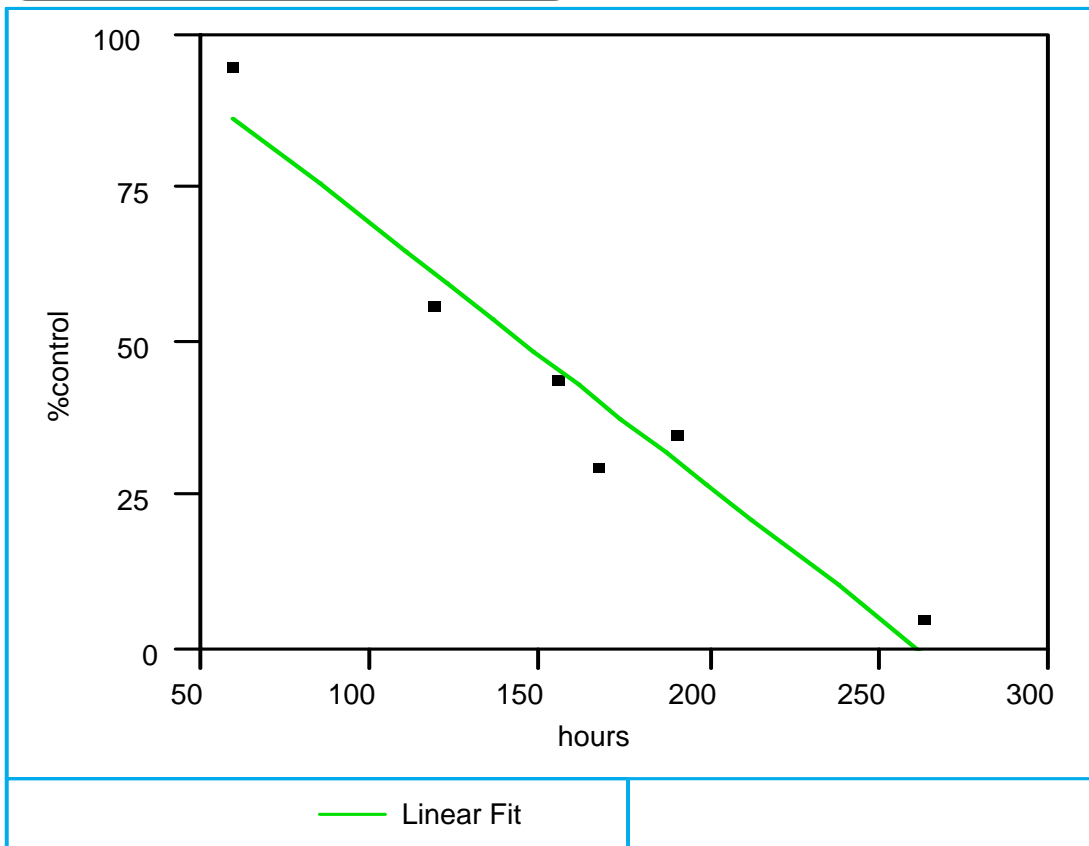
Agrifos vs. Azomite Treatments

Mature CLOs Detached Branch Assay



Efficacy of curative treatments (% control) plotted against time elapsed between inoculation and application of treatment

%control By hours



Linear Fit

$$\%control = 112.439 - 0.4267 \text{ hours}$$

Long Term Treatment of Tanoaks

- 32 Field Plots
- 6 Sites in 3 Counties
- 672 Tanoaks > 8cm DBH

Shell Beach

SDSF

Santa Lucia x2

Goetz

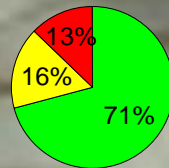
Mill Creek



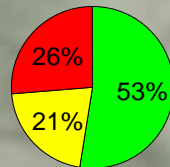
Long Term AF Treatment of Tanoaks

% Asymptomatic Control	56.5312	t-Ratio	-4.50771
% Asymptomatic treatment	74.9062	DF	15
Mean Difference	-18.375	Prob > t	0.0004
Std Error	4.07635	Prob > t	0.9998
Upper95%	-9.6865	Prob < t	0.0002
Lower95%	-27.064		
N	16		
Correlation	0.86547		

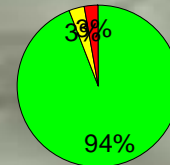
C1



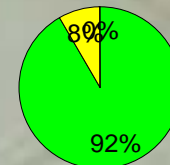
C2



T1



T2



What molecular mechanisms does treatment trigger? Are

Soquel Site A

Soquel Demonstration State Forest

11/2009

Bicycles for Cargo Transport

Matteo's Lab 2006



Viet Cong 1966



Sanitation

Drying plant woody debris fast

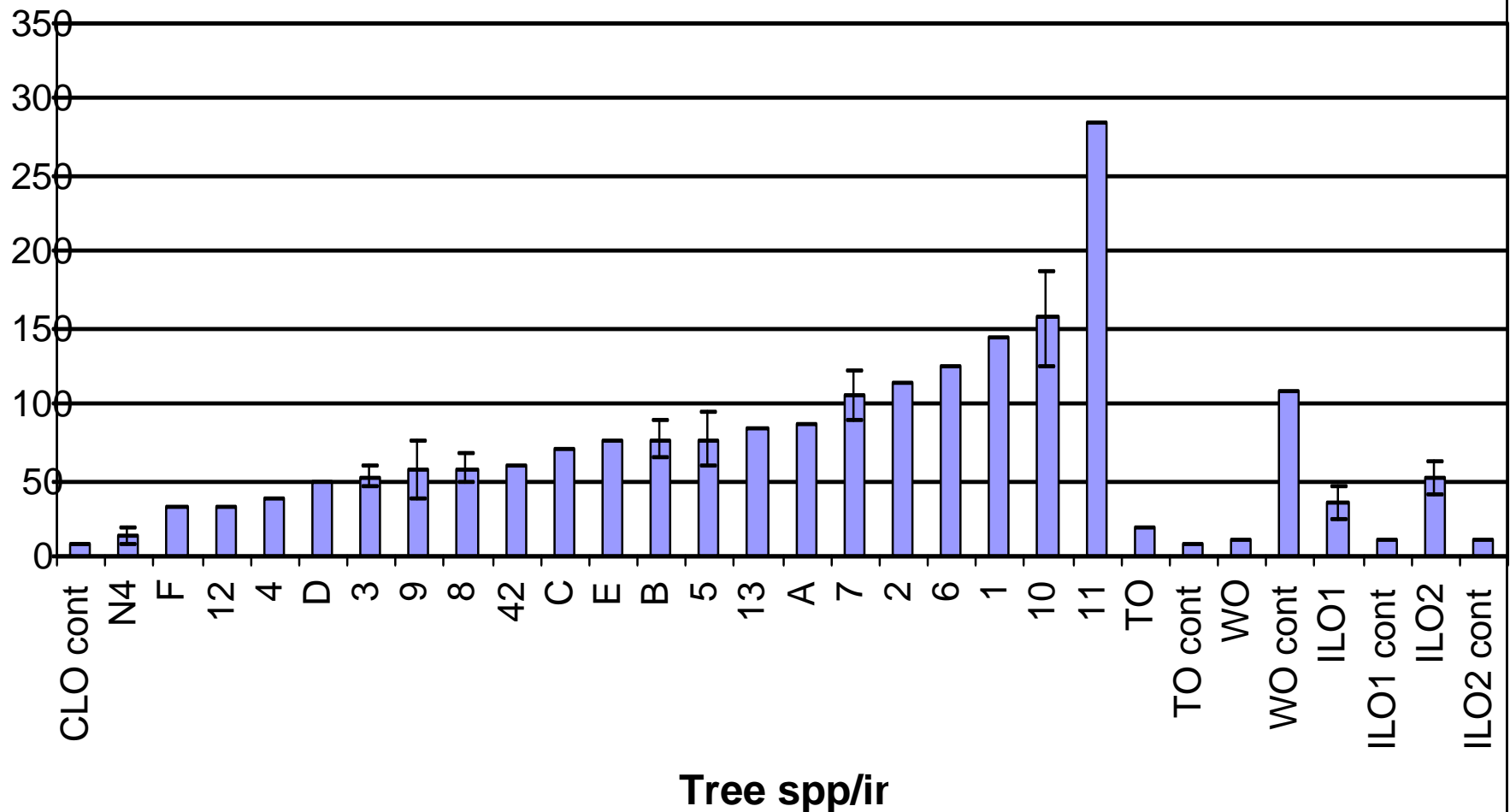
Composting

Chemical treatment of water

Diagnosis of wood decay agents
may be of use for so called
green tree failures

NATURAL RESISTANCE?

Figure 1: Longitudinal lesion lengths of coa



Population genetics of host can help determine nature, frequency, and distribution of possible resistance

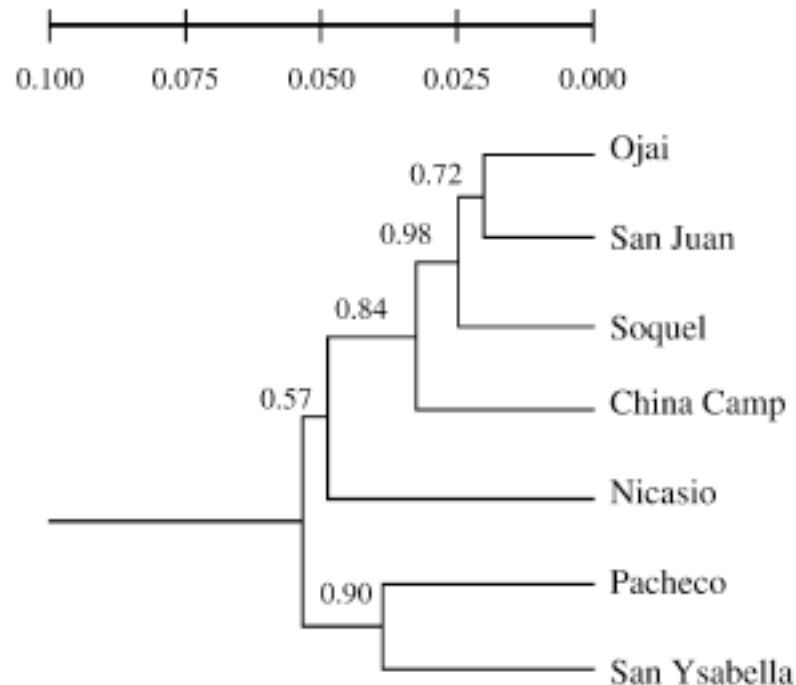


Fig. 5 UPGMA tree based on distances between populations estimated from Nei's unbiased genetic distance of amplified fragment length polymorphism (AFLP) fragments. Support for branches is given by bootstrap percentages for recalculated genetic distances from 1000 bootstrap replicates.

Table 5 Mantel R correlation coefficients between amplified fragment length polymorphism molecular similarity classes of *Quercus agrifolia* and Euclidean distances in lesion size of *Q. agrifolia* after inoculation with *Phytophthora ramorum* for each of four trials

Molecular similarity	Trial 1	Trial 2	Trial 3	Trial 4
Up to 99 %				
Lesion \times genotype	0.022	0.040	-0.019	-0.04
Up to 95 %				
Lesion \times genotype	-0.05	0.03	-0.102*	0.05
Up to 90 %				
Lesion \times genotype	-0.05	-0.01	-0.053	0.070
Up to 75 %				
Lesion \times genotype	0.04	-0.04	0.05	-0.04
Up to 50 %				
Lesion \times genotype	0.01	0.02	0.02	0.10*

*, $P < 0.05$.

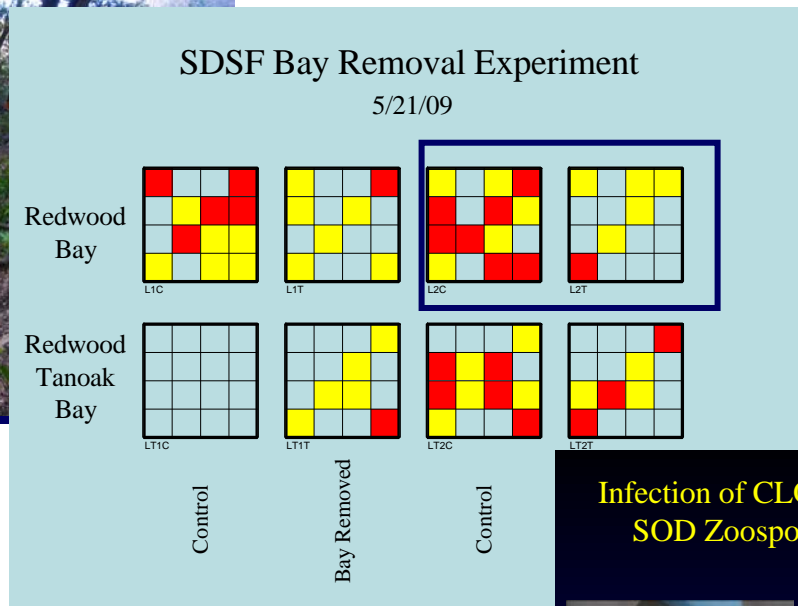
common population membership remained significant for the 25% small lesion size class for all trials.





Bay removal reduces but does not eliminate inoculum

California Bay Laurel Removal for SOD Control



Is this reduction
sufficient?



Infection of CLOs by
SOD Zoospores



Inoculator
&
Grafting
Wax

Pipetting
Zoospore
Suspension

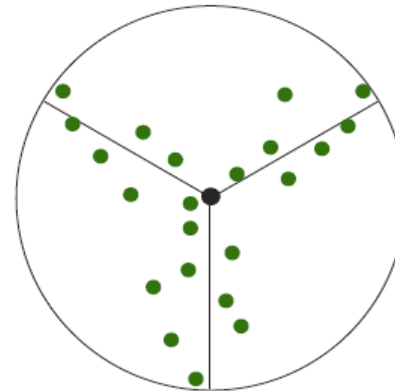


Elimination of all bays not feasible or desirable:

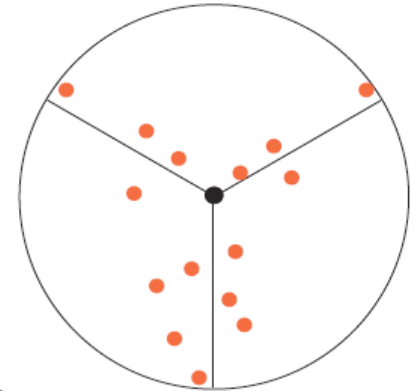
- Eliminate bays near oaks



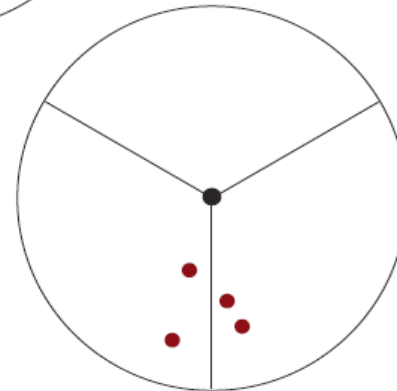
- Eliminate bays identified as hotspots



Sampled Bay Trees



P. ramorum isolated



Isolated at least 3 out of 4 sampling periods

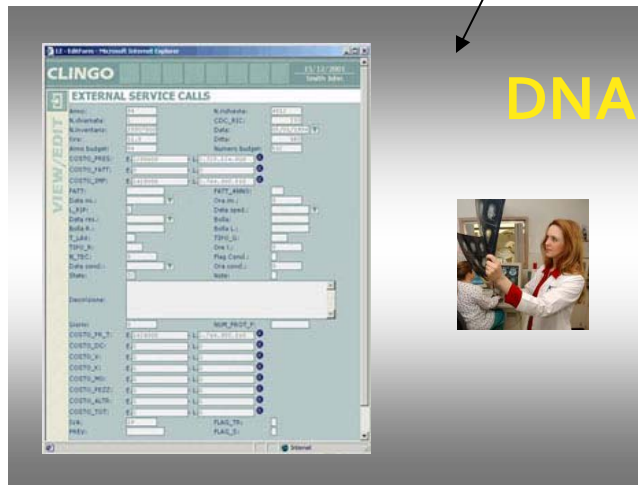
PREVENT: Diagnose



Symptoms relatively generic



DNA TESTS



ERADICATION

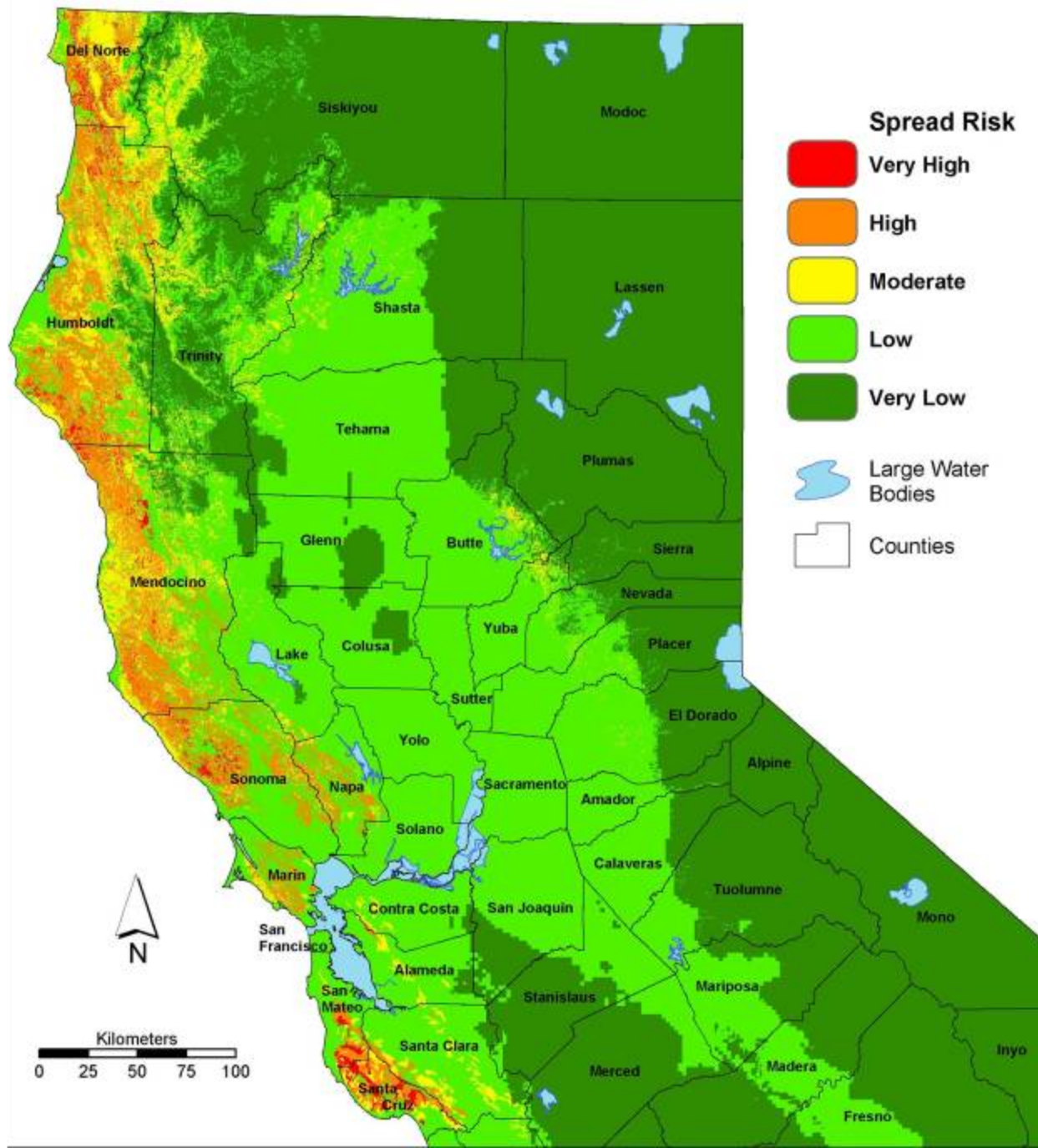


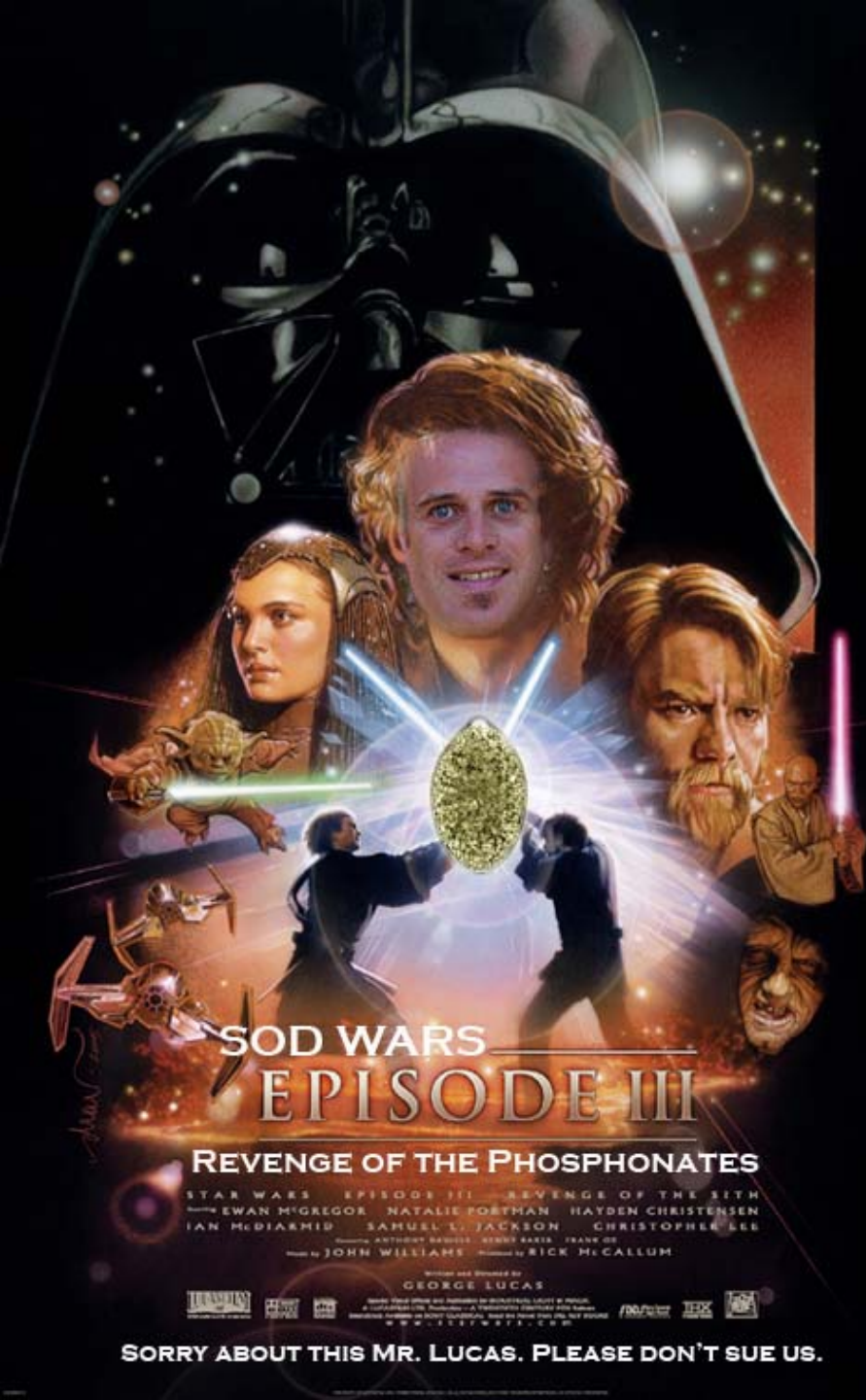


Nothofagus fusca = dead
end host

Fuchsia excorticata = super
spreader

Meentemeyer et al., 2004






USDA Competitive Grant Program

Pacific Southwest US Forest Service

The Betty & Gordon Moore Foundation

NSF: Ecology of Infectious Diseases



Sudden Oak Death Update for Foresters and Landowners

Ukiah & Eureka, California

May 12-13, 2010