- Why are elm beetles so successful in vectoring the Dutch Elm Disease pathogen

- How does Dutch Elm Disease affect a tree (hint: at least respond with two effects)

- Based on the lecture when did the two outbreaks of Dutch Elm Disease occur and were they caused by the same pathogen, if not please name the pathogens involvec

- Dutch Elm Disease is one of the best examples of interspecific introgression of adaptive genes. Explain which genes and why do we consider them adaptive

Oomycota

- Belong to a kingdom that includes kelp and diatomes
- Kingdom used to be called Chromista (brown algae), it is now the Straminopila
- It includes many important plant pathogens:
  - *Peronospora*: mostly aerial
  - *Pythium*: mostly soilborne organisms
  - *Phytophthora*: mixed biology

Blue mold of tobacco caused by *Peronospora tabacina*

- Ability to travel aerially for hundreds of kilometers from Caribbean to Southern US
- Ability to predict arrival of inoculum based on weather pattern
- Some species capable of over-wintering in buds

Oomycetes are <u>not</u> fungi

- Cellulose in cell wall
- Ploidy is 2n
- Result of sexual activity is oospore (2n)
- Meiosis, somatogamy, caryogamy all occur at the same time

- Water adapted biology, flagellate phase
- No septa, holocoenocytic hyphae

- Chitin in cell wall
- Ploidy is n, or n+n
- Result of sexual activity is a spore n
- Meiosis, somatogamy,caryogamy are usually interupted by vegetative (somatic phase)
- Better adapted for aerial transmission
- Septate hyphae

Phytophthora

- Some important plant pathogens, with very well known history
  - *Phytophthora infestans* and the Irish potato famine
  - *Phytopthora cinnamomi* and the Jarrah dieback in Australia

The Irish Potato Famine

- From 1845 to 1850
- *Phytophthora infestans*
- Resulted in the death of 750,000
- Emigration of over 2 million, mainly to the United States.

Phytophthora: "plant destructor"

- Best known pathogen whose long-distance transport linked to agriculture.
  - *Infected root-stocks*
  - *Infested soil*
  - *Infected plants*

90+ species of *Phytophthora*

- 60 until a few years ago, research accelerated, especially by molecular analyses
- Differentiated on basis of:
  - Type of sexual intercourse
  - Type of sexual activity
  - Number of hosts
  - Ideal temperature
  - Type of biology
  - Evolutionary history (Waterhouse-Cooke)

Type of sexual strategy

Homothallic species, will produce both oogonia and antheridia and mate by themselves (hermaphrodite), low genetic variability. Strong inbreeding.

Heterothallic species need two individuals with different MATING TYPES. Normally defined as A1 and A2. Out-crossing species.

Type of sexual strategy

In area of origin expectations are:

-Both mating types if heterothallic

-Sexual activity and large number of different genotypes

-If species is homothallic expectation is that populations in isolated areas should be different genetically because of lack of gene flow and genetic drift (basic Darwinian concept**)**

Type of sexual strategy

If species is exotic, expectations are:

-Often one mating type only, or mating types introduced at different times.

-Low genotypic diversity, prevalence of clonal lineages

-If species is homothallic expectation is that all individuals will be similar, because there has been no time for genetic differentiation

Type of sexual strategy

Why should we care about sex ?

-Ability to create new alleles, better potential of adaptation to new conditions

-Ability to exchange genes with other individuals, if gene pool is large, it can be a great advantage

-For instance ability to overcome the fungicide metalaxyl happened when A1 and A2 of *P. infestans* got together and reproduced.

Number of hosts

- Single hosts, specialized: *P. sojae*, *P. lateralis*
- Multiple hosts, generalists: *P. cinnamomi* (3000 hosts!), *P. ramorum* (> 60)

Temperature

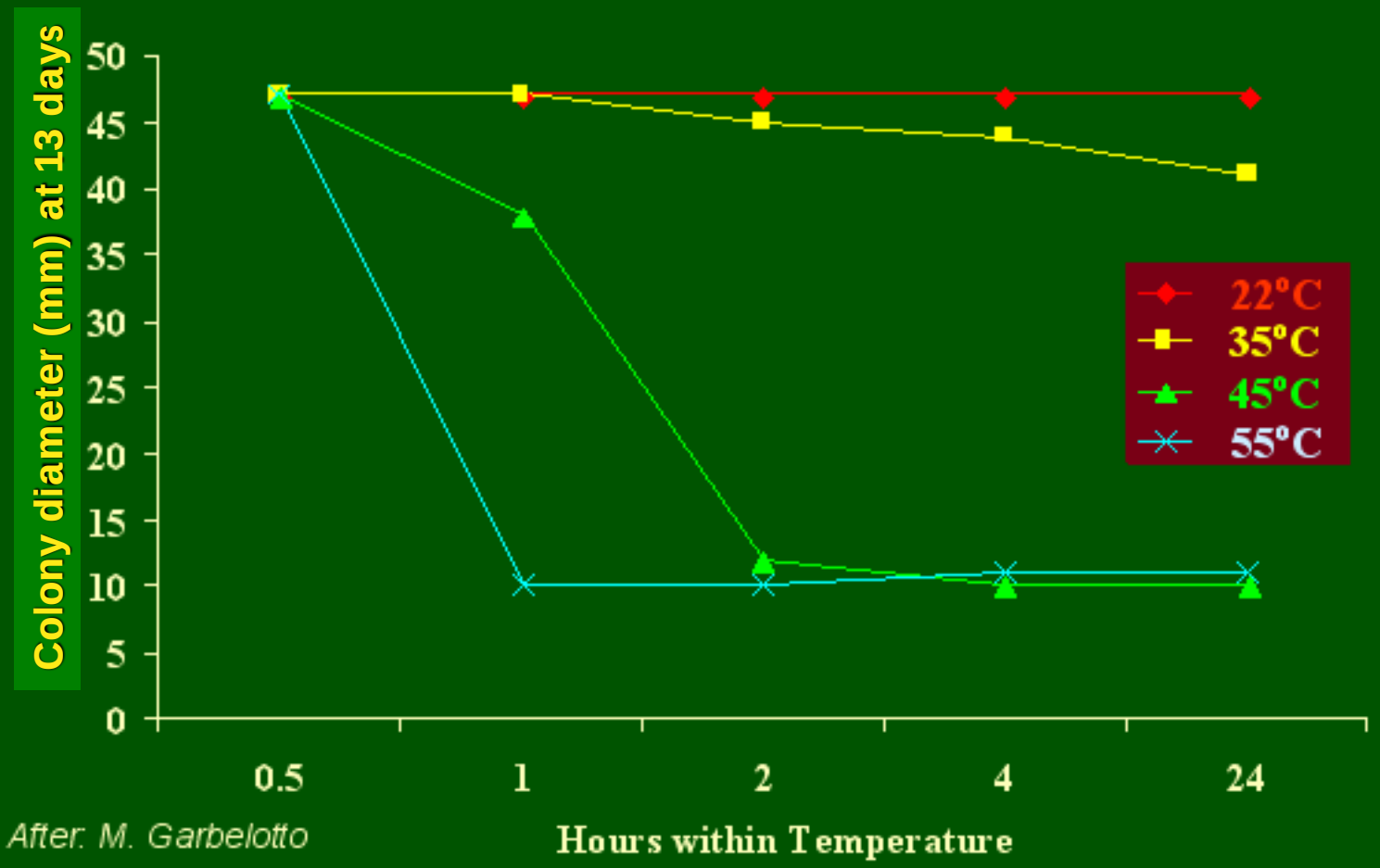
- **Optimal temperatures**: explain why many species are extremely seasonal, also correlated to area of original evolution of species.
  - Thermofilic spp. (summer species): *P. palmivora, P cinnamomi, P. citricola*
  - Psychrofilic spp. (winter species): *P syringae, P lateralis, P. hybernalis, P. ramorum*

- **Extreme temperatures**: ability to withstand extreme cold or heat. Normally depends on resting structures, and where they are produced.

  Species producing abundant chlamydospores within plant tissue will be more resilient

EFFECT OF TEMPERATURE ON MEAN PATHOGEN GROWTH

(starting colony size=10mm)



After: M. Garbelotto

Heat treatment results

- Pre-treatment - baseline (isolation success)
  - Wood Chips = 96% (n=87)
  - Wood Logs = 44% (n=48)
  - Bay Leaves = 100% (n=50)
- 1 week of heat
  - Wood Chips = 0% (n=87)
  - Wood Logs = 0% (n=48)
  - Bay Leaves = 30% (n=50) WHY?
- 2 weeks of heat
  - Wood Chips = 0% (n=87)
  - Wood Logs = 0% (n=48)
  - Bay Leaves = 0% (n=50)


Chlamydospores produced on and
in bay leaves

Type of biology

- **Waterborne, soilborne**
- Mostly root-infecting
- Can move from roots into root collar and stem
- Moves using water accumulations (floods) or streams
- Infested soil source of easy infection

- **Aerial**
- Infect leaves, twigs and branches
- Can move onto stems
- Moves using wind
- Need rainy conditions to infect (free water needs to accumulate)

Soilborne, waterborne species

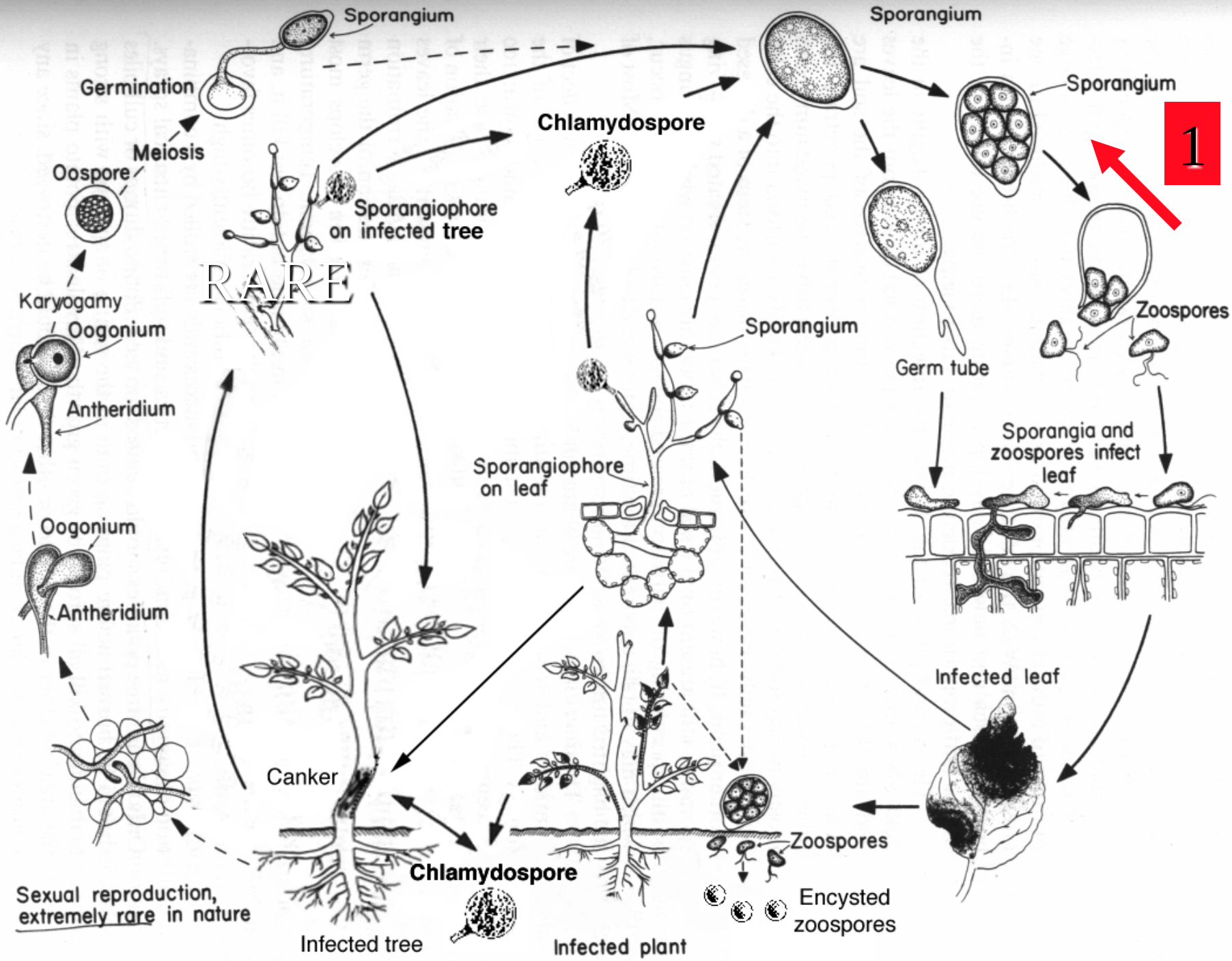
- Clear association with water: along streams, in areas that are temporarily flooded
- Ability to rest in soil with resting structures such as chlamydospores, oospores, but also encysted zoospores
- Chemotaxis: ability to move towards susceptible host, even in darkness

Aerial species

- Recently discovered for forests: all characterized by deciduous sporangia
- Distance of spread depends on how dry sporangia are. If sporangia are dried they can go far
- True aerial will naturally infect aerial parts without need for root infections or tranmssion by tools
- Ability to rest in soil with resting structures is not lost!!, but epidemiological relevance not clear in nature
- Chemotaxis: ability to move towards susceptible host, may be lost in darkness

Soil-Waterborne vs.Aerial

- *P. cinnamomi*
- *P. citricola*
- *P. cactorum*
- *P. quercina*
- *P. cambivora*
- *P. palmivora*
- *P. ramorum*
- *P. nemorosa*
- *P. pseudosyringae*
- *P syringae* (because of pruning)


Sporangium
Germination
Meiosis
Oospore
Karyogamy
Oogonium
Antheridium
Oogonium
Antheridium
Sporangiophore on infected tree
RARE
Chlamydospore
Sporangium
Sporangium
Sporangium
Germ tube
Zoospores
Sporangia and zoospores infect leaf
Sporangiophore on leaf
Sporangium
Infected leaf
Canker
Zoospores
Chlamydospore
Encysted zoospores
Sexual reproduction, extremely rare in nature
Infected tree
Infected plant
1

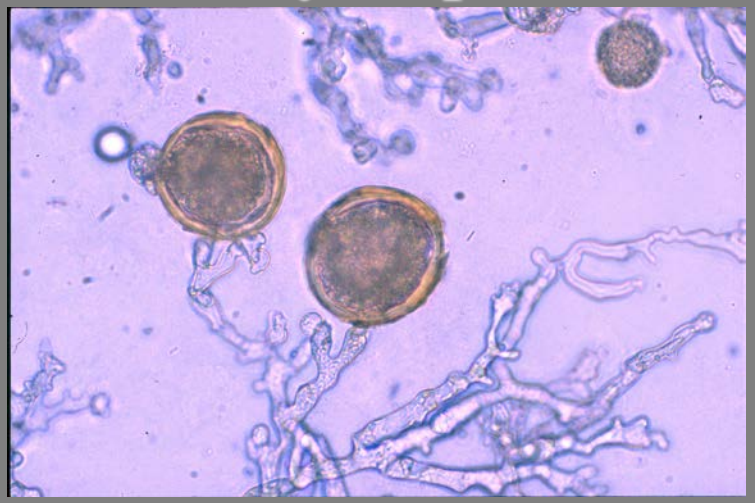
Important structures

- **Sporangia**: size, shape, L:B, papillate or not, deciduous or not
- **Stalks**: length
- **Zoospores**. **Encysted zoopsores**
- **Chlamydospores**: how are they carried (lateral vs. terminal), size, color, ornamentation
- **Oospores**
- **Hyphae:** swellings present or absent, linear or tormented
- **Colony morphology:** appressed vs aerial, fast-growing vs. slow-growing

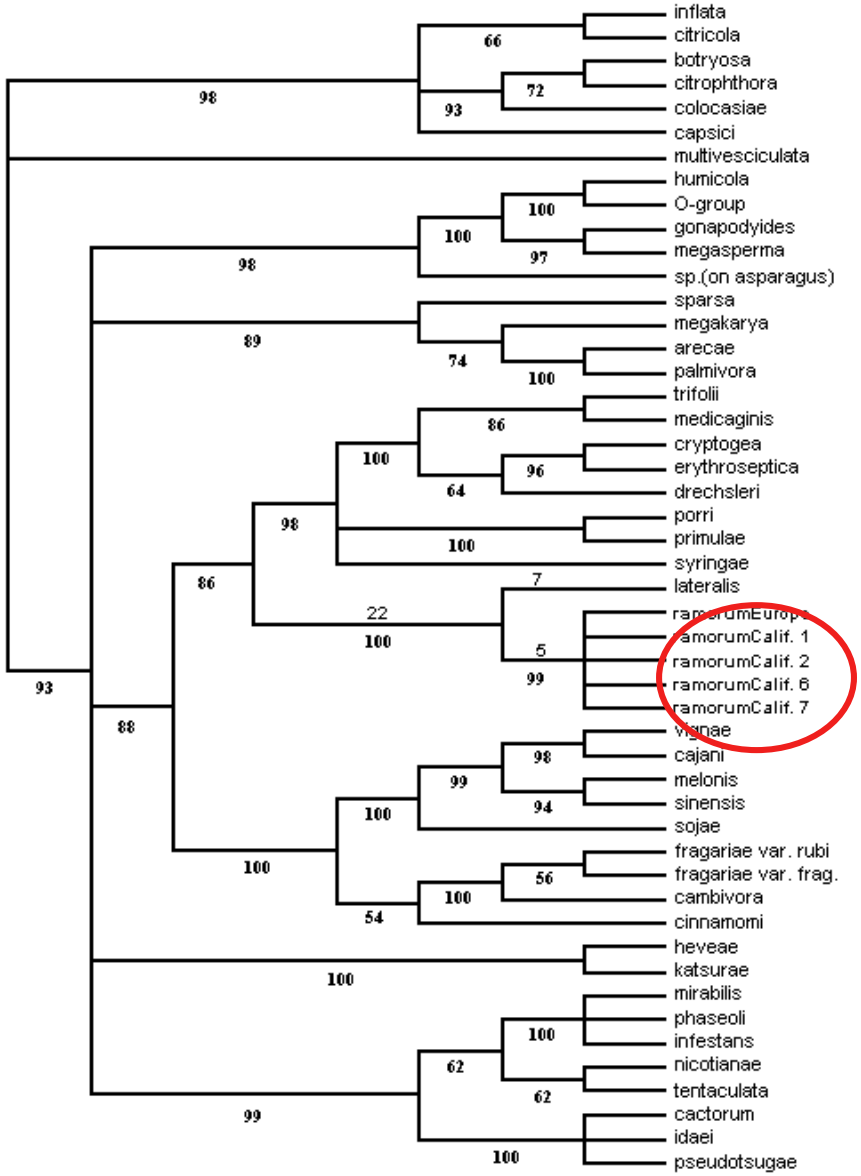
Phytophthora ramorum

Sporangia

Chlamydospores



Phytophthora - ITS



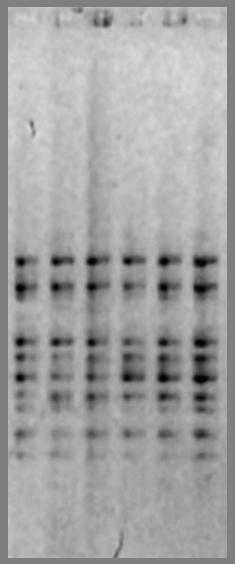
Use of molecular data

- Understanding of evolutionary relationships
- Definition of species, especially important if there are few morphological features to differentiate taxa
- Definition of genetic and genotypic diversity. Understanding of biology (sex vs. no sex, etc.)
- Diagnostics

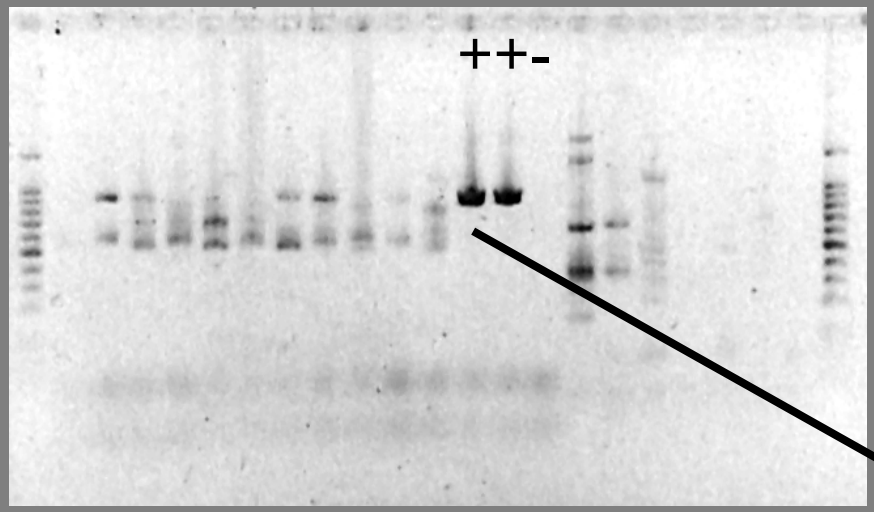
Environmental sample

DNA extraction

DNA fingerprints



Dna probes (plus/minus)



Complete sequencing

DIAGNOSTICS

- **Traditional isolation**, baiting, and morphological description
- **DNA-direct**: look for *P. ramorum* DNA itself
- **DNA-indirect:** proteins, elicitins
- **Quantitative PCR**: its applications and importance
- **Testing the process**, the procedure
- **Comparative testing** of reliability and sensitivity of different methods

Plating vs. Baiting

Method depends on substrate

Culturing, baiting, etc.

- **Knowledge is increasing** on tricks necessary to grow *P. ram* from plants and other substrates. Morphology is easy to recognize
- **False negatives** are frequent: mycelium is ephemeral, strong seasonality, huge effect of substrate (inhibitors), sensitivity to antibiotics, limited time window to isolate,

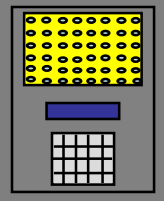
Overview of methods

collect symptomatic material

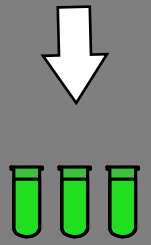
dry material, then grind

extract bulk DNA

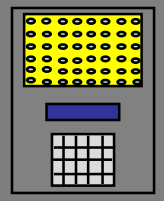
1st round PCR reaction (using phyto1,4)



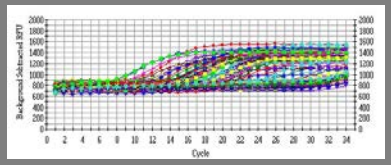
Amplify in thermocycler



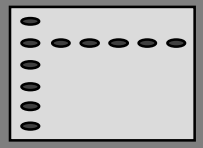
2nd round PCR reaction (using phyto2,3) with first round products (1:500 dilution) Opt. SYBR green



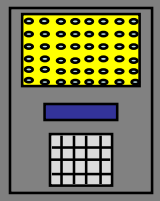
Amplify in light-cycler



light-cycler detects fluorescent products and determines melt temperatures



Electrophoresis of 2nd round PCR products



Amplify in thermocycler

Why molecular diagnostic

- Some microbes are hard or impossible to culture
- Morphology of microbes can be extremely variable (interpretation will be subjective)
- Cryptic species: sometimes associated with critical traits, e.g. host range

DNA-based diagnosis: major principles

- Diagnosis relies on one or a few loci, not on the whole genome

- DNA sequence database (e.g. GENBANK) will determine potential *locus* of choice
- *Locus* needs to be informative at the right taxonomic level: e.g. conserved within species and variable between species
- Approach needs to capitalize on sequence differences among related species (probably a good idea to know nature of the sequence one is using): generally this is done by designing taxon-specific primers
- Method sensitivity will depend on nature of selected locus: single- vs. multiple loci, nuclear vs. mitochondrial
- Method needs to include variable independent ways to verify results in order to avoid false positives

Two conceptually different approaches:

- DNA probes that will detect an entire group of pathogens
- As a second step, refine the diagnosis for a specific pathogen
- Good sensitivity, results will be very informative regarding a variety of microbes

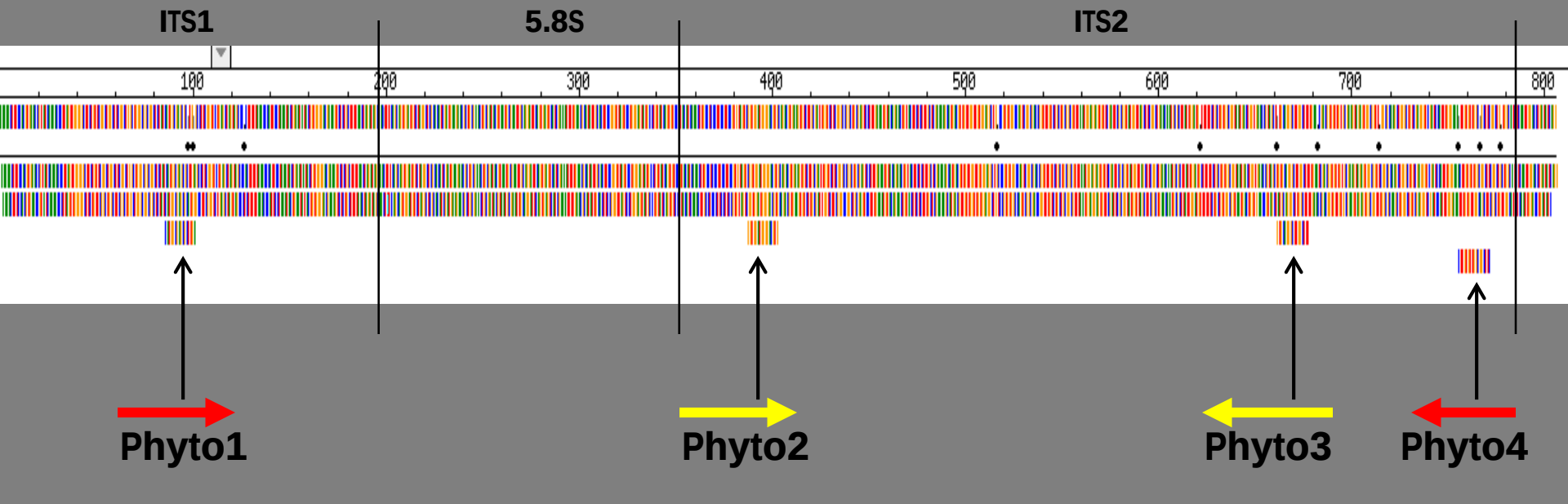
- DNA probe that are very specific for one pathogen
- Most sensitive technique, but obviously will only provide info one 1 microbe

Methods

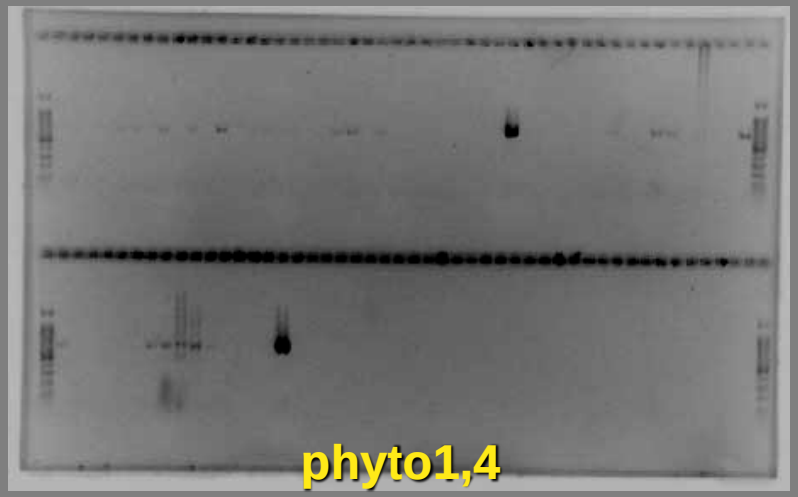
Designed 2 sets of *P. ramorum* specific primers (www primer3 software)

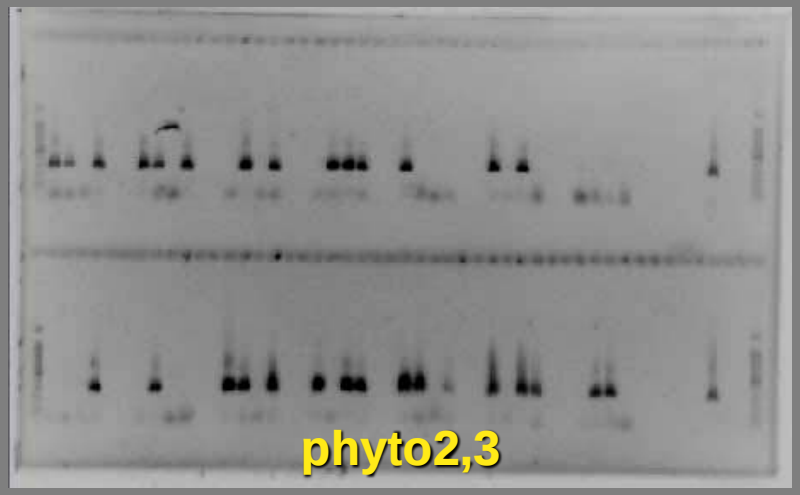
- **phyto1-phyto4 (1st round PCR)**
  - **highly specific for *P. ramorum***
    - **687 bp fragment (in between red arrows)**

- **phyto2-phyto3 (2nd round PCR)**
  - **nested in phyto1-4 amplicon; specific for *Phytophthora* spp.**
    - **291 bp fragment (in between yellow arrows)**



Why NESTED approach
(using more specific primers on first round)





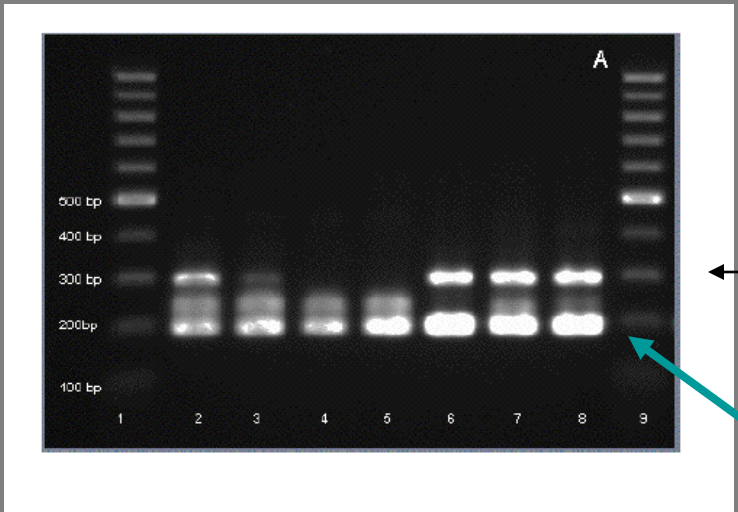
Increase sensitivity while maintaining specificity

Some infected wood / leaf extracts need both rounds before a positive result

- Amount of pathogen DNA low in some tissue

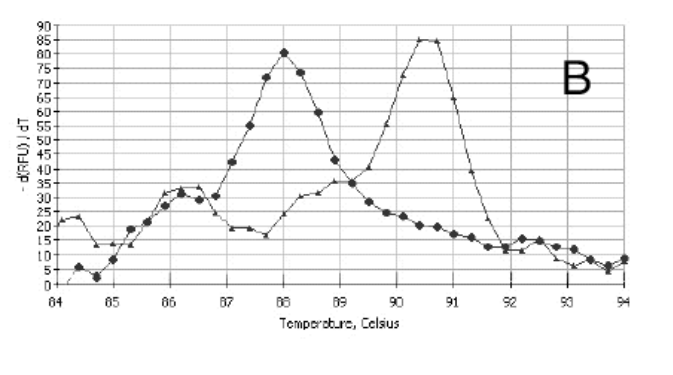
Run	Round	# positive	% positive
run1	phyto1,4	8/60	13%
	phyto2,3	20/60	33%
run2	phyto1,4	12/56	21%
	phyto2,3	20/60	54%
run3	phyto1,4	15/55	27%
	phyto2,3	39/55	71%

Quantitative PCR



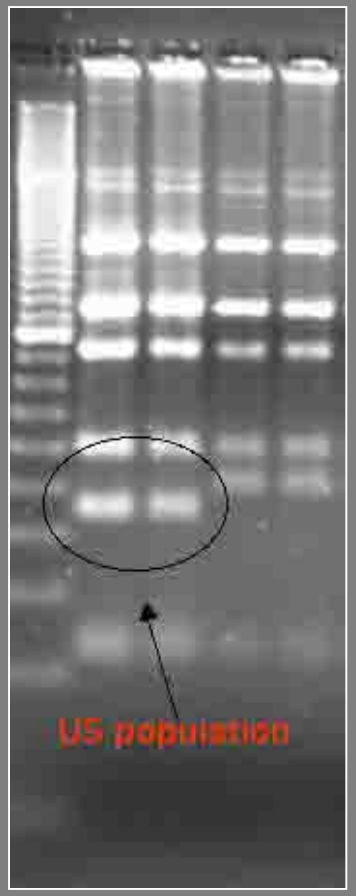
← P. ram

Universal

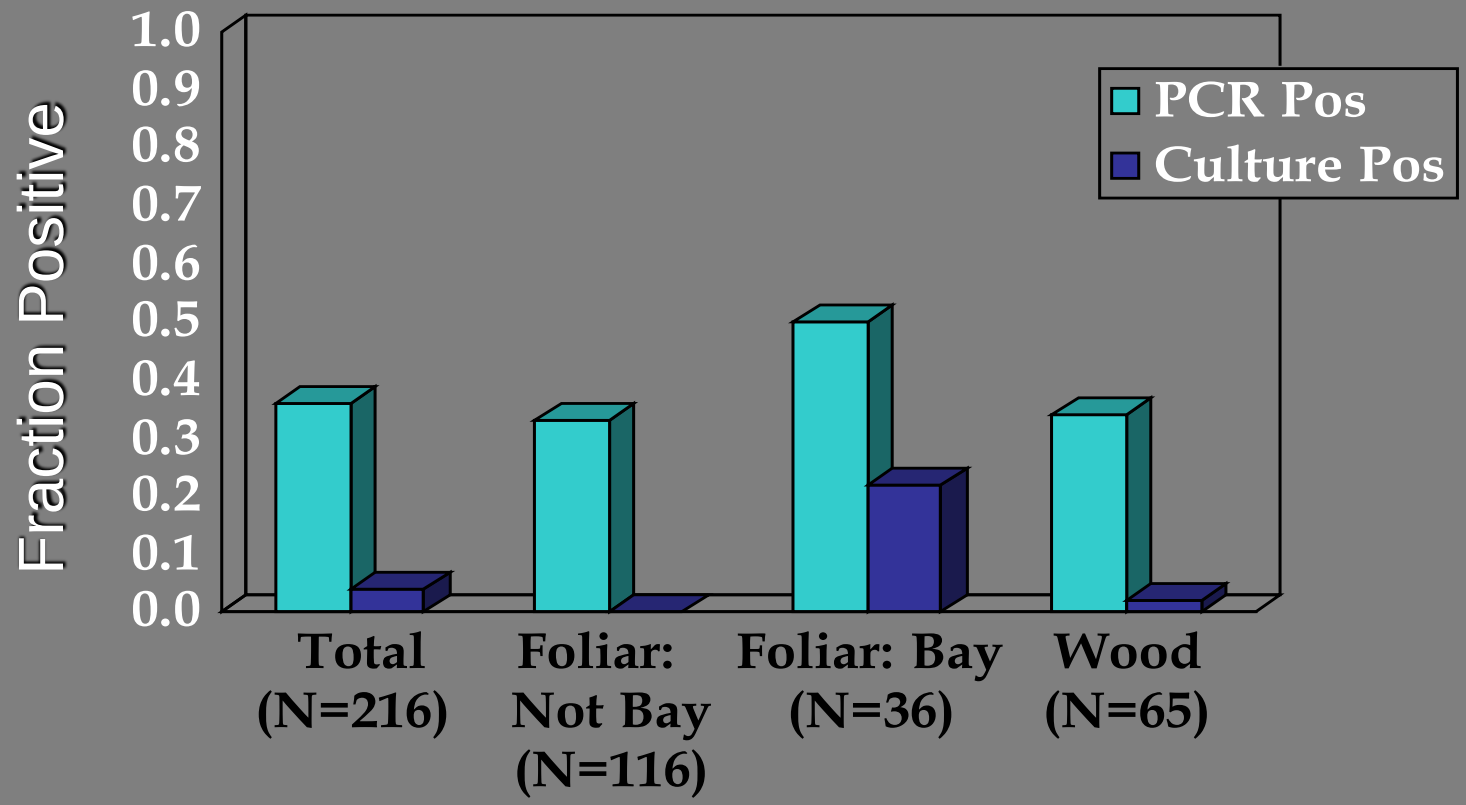


Melt curve

RFLP

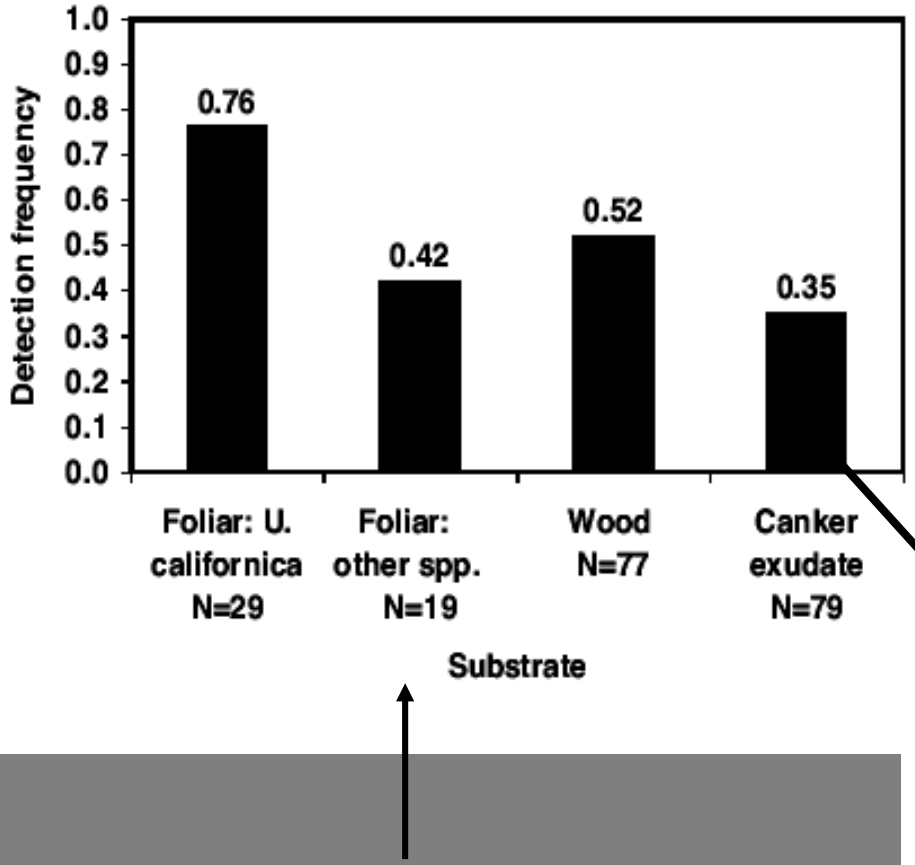


PCR success is much higher than isolation success on "difficult" samples.

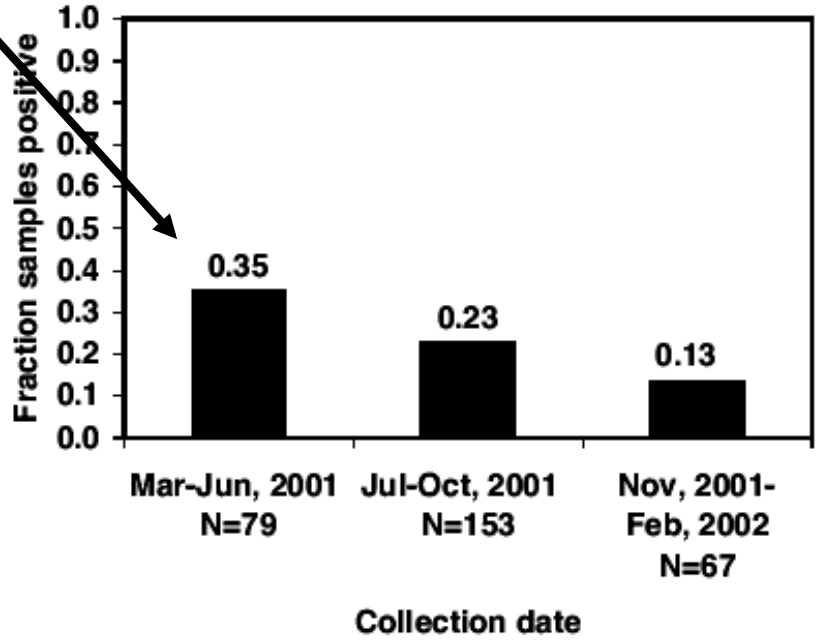


Significant effect of diagnostic type (P <0.001) and sample type (P=0.0036)

FIELD DATA (from Hayden et al. 2004)



Effects of substrate and season →



Generic assays

- Martin's group protocol, mitochondrial locus, quantitative
- Hong's group SSCP, ITS based

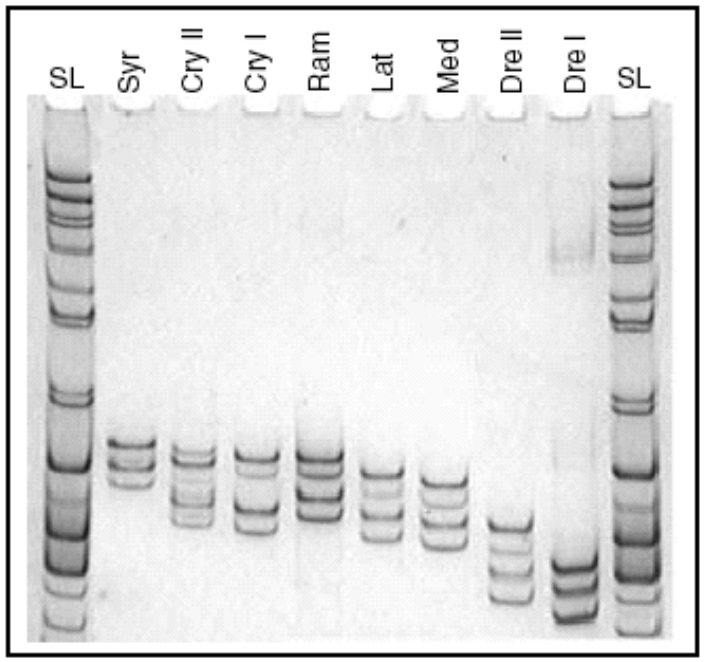
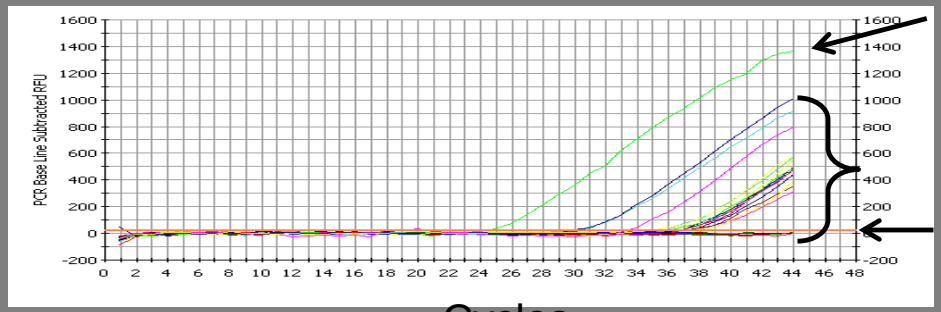


Fig. 2 Single-strand-conformation polymorphism profiles of ITS-1 for *Phytophthora ramorum* (Ram) and its genetically close relatives. Cry I and Cry II, *P. cryptogea* subgroups I and II; Dre I and Dre II, *P. drechsleri* subgroups I and II; Lat, *P. lateralis*; Med, *P. medicaginis* and Syr = *P. syringae*. SL represents a single strand DNA ladder

Quantitative PCR

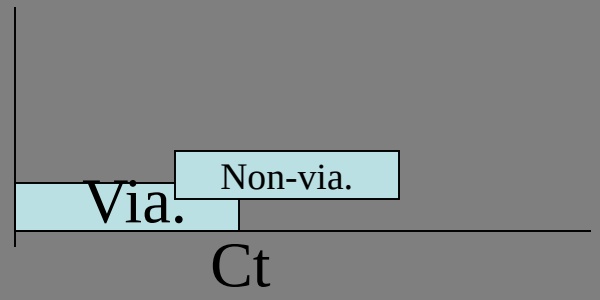
- Allows to check for specificity of PCR product
- Allows to quantify amount of target DNA
- Allows to quantify effects of different substrate/extraction



More DNA---------Less DNA

Applications of Q-PCR

- **VIABLE vs. NON V**.
- Bay laurel leaves were separated in two groups based on success of isolations
- Mean Ct were significantly different and predicted distributions were not totally overlapping



- **ASSESS IF SENSITIVITY OF ASSAY WAS SUFFICIENT**
- Use PCR to determine whether *P. ram.* DNA present in finished compost
- PROBLEM: substrate strongly affects PCR results.
- Approach: amount of DNA detectable in all samples was determined and then used Q-PCR to find out whether sensitivity was sufficient

- Assay capable of detecting 240 g/ml ^10-5. Samples had at least one order of magnitude more DNA

MANAGEMENT

- **Single tree level**: excise cankers/eliminate inoculum sources/ make conditions unfavorable to the pathogen
- **Landscape level**: stand management questions being researched: remove bays (Garbelotto) ? How to thin tanoaks (O'Hara) ? Effect of fire (Moritz) ? Inoculum identification (many groups) and reduction. Chemical treatments on oaks and on bays.

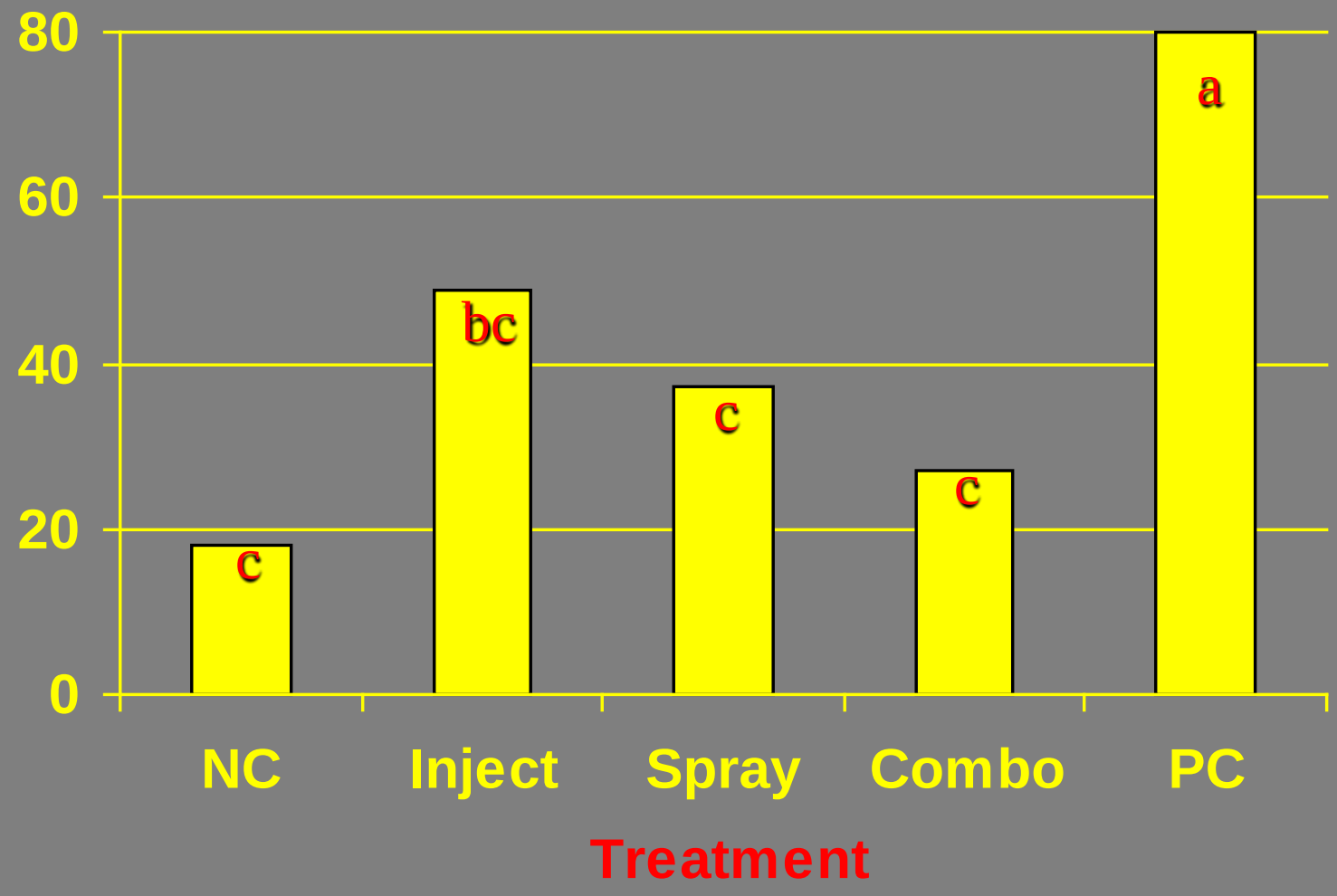
Treatments and inoculum reduction

- Preventive phosphonate treatment work and are registered for oaks tanoak
- A completely novel application method was developed for these compounds: mix with Pentrabark and apply on stem

- Developed and tested some heat and heat+ vacuum treatments
- Proven that in absence of oospores composting eliminates pathogen
- Wood and other green waste eliminates sporulatiuon

Felton
Shreve Oaks

Treatment vs. Canker Size

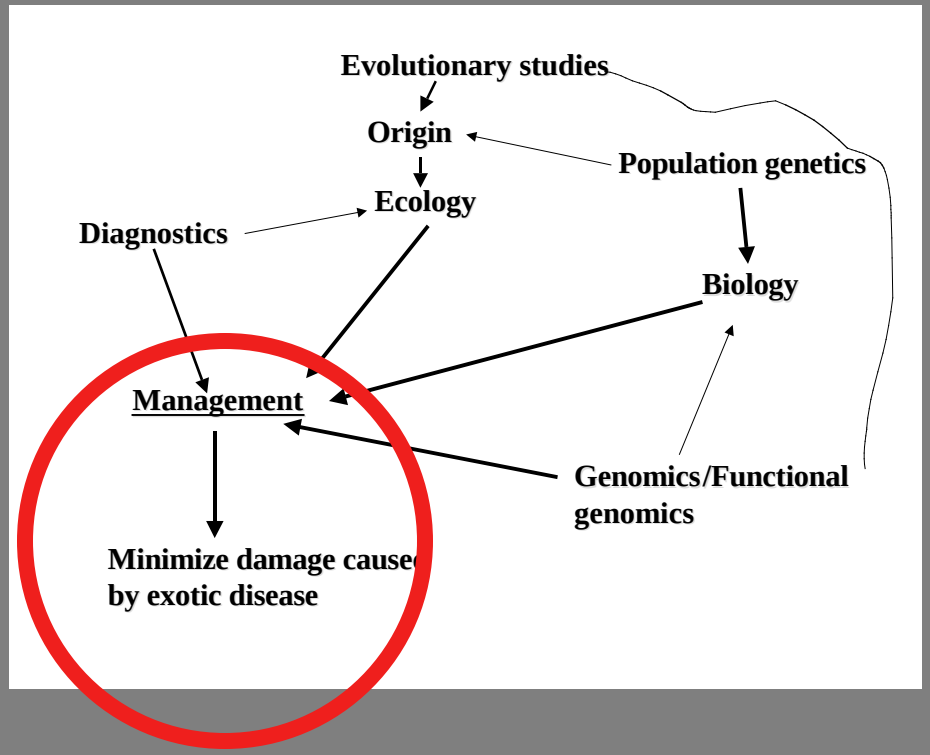


Forest-Regional level

Avoid introduction, early detection, quarantine system is important as no real solution to problem exists once pathogen is introduced

P. ramorum is not *P. ramorum* is not *P. ramorum*
US lineage (A2) EU lineage (A1) New strain in WA
Avoid even regional movement of infected plant material is important

Remember this slide ?



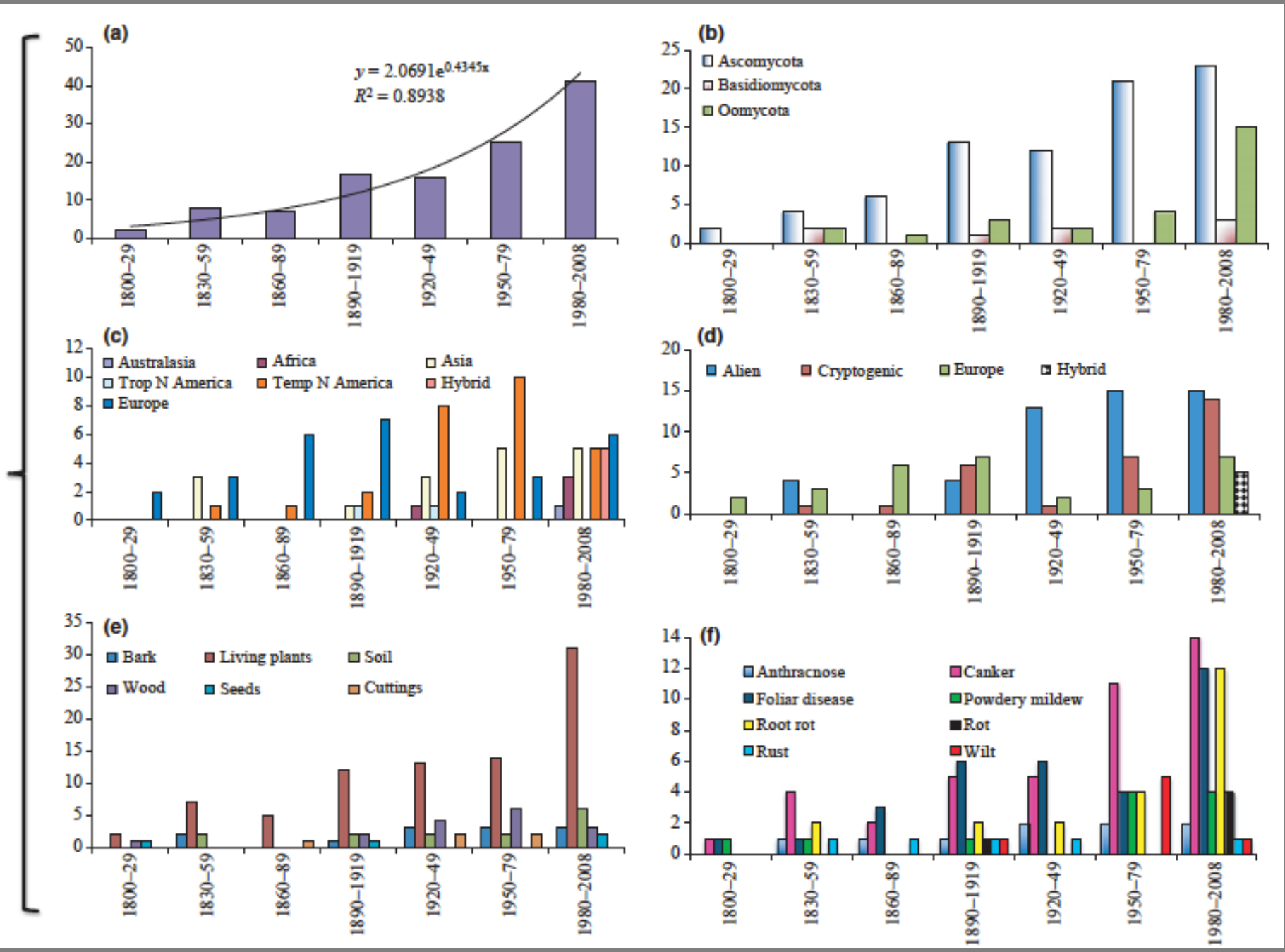
"**The state of our knowledge**" is growing but still too limited to achieve final goal

New Phytologist

Research

Biogeographical patterns and determinants of invasion by forest pathogens in Europe

A. Santini[1], L. Ghelardini[1], C.De Pace[2], M. L. Desprez-Loustau[3], P. Capretti[4], A. Chandelier[5], T. Cech[6], D. Chira[7], S. Diamandis[8], T. Gaitniekis[9], J. Hantula[10], O. Holdenrieder[11], L. Jankovsky[12], T. Jung[13], D. Jurc[14], T. Kirisits[15], A. Kunca[16], V. Lygis[17], M. Malecka[18], B. Marcais[19], S. Schmitz[5], J. Schumacher[20], H. Solheim[21], A. Solla[22], I. Szabò[23], P. Tsopelas[24], A. Vannini[25], A. M. Vettraino[25], J. Webber[26], S. Woodward[27] and J. Stenlid[28]


No. of IFPs
(a)
y = 2.0691e^0.4345x
R² = 0.8938
1800–29
1830–59
1860–89
1890–1919
1920–49
1950–79
1980–2008
(b)
Ascomycota
Basidiomycota
Oomycota
(c)
Australasia
Africa
Asia
Trop N America
Temp N America
Hybrid
Europe
(d)
Alien
Cryptogenic
Europe
Hybrid
(e)
Bark
Living plants
Soil
Wood
Seeds
Cuttings
(f)
Anthracnose
Canker
Foliar disease
Powdery mildew
Root rot
Rot
Rust
Wilt

Daddy, everybody knows preventing new infestations is the best thing to do!!!!!

Pathologists at the center of a circus

Matteo Garbelotto, Katherine Hayden, ShannonU.C. Berkeley

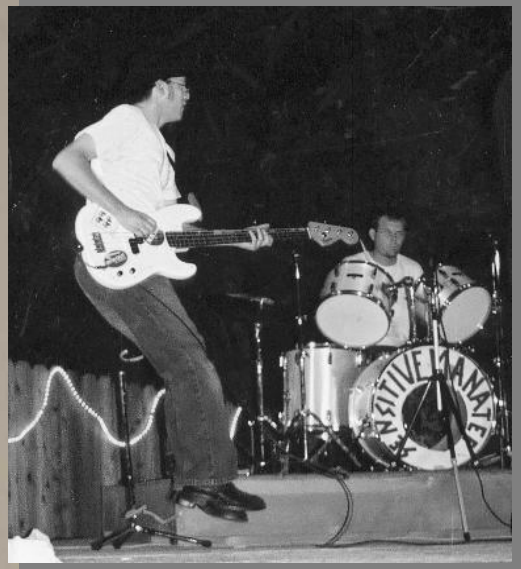
Takao Kasuga

ARE YOU PREPARED?

SUDDEN OAK DEATH

IS COMING…AND THE ONLY CURE IS EXPOSURE

SUDDENOAKDEATH99@HOTMAIL.COM



Tanoak mortality by “causes unknown” (1995-2001)

Girdling aerial ‘cankers’ removed from roots

Tanoak

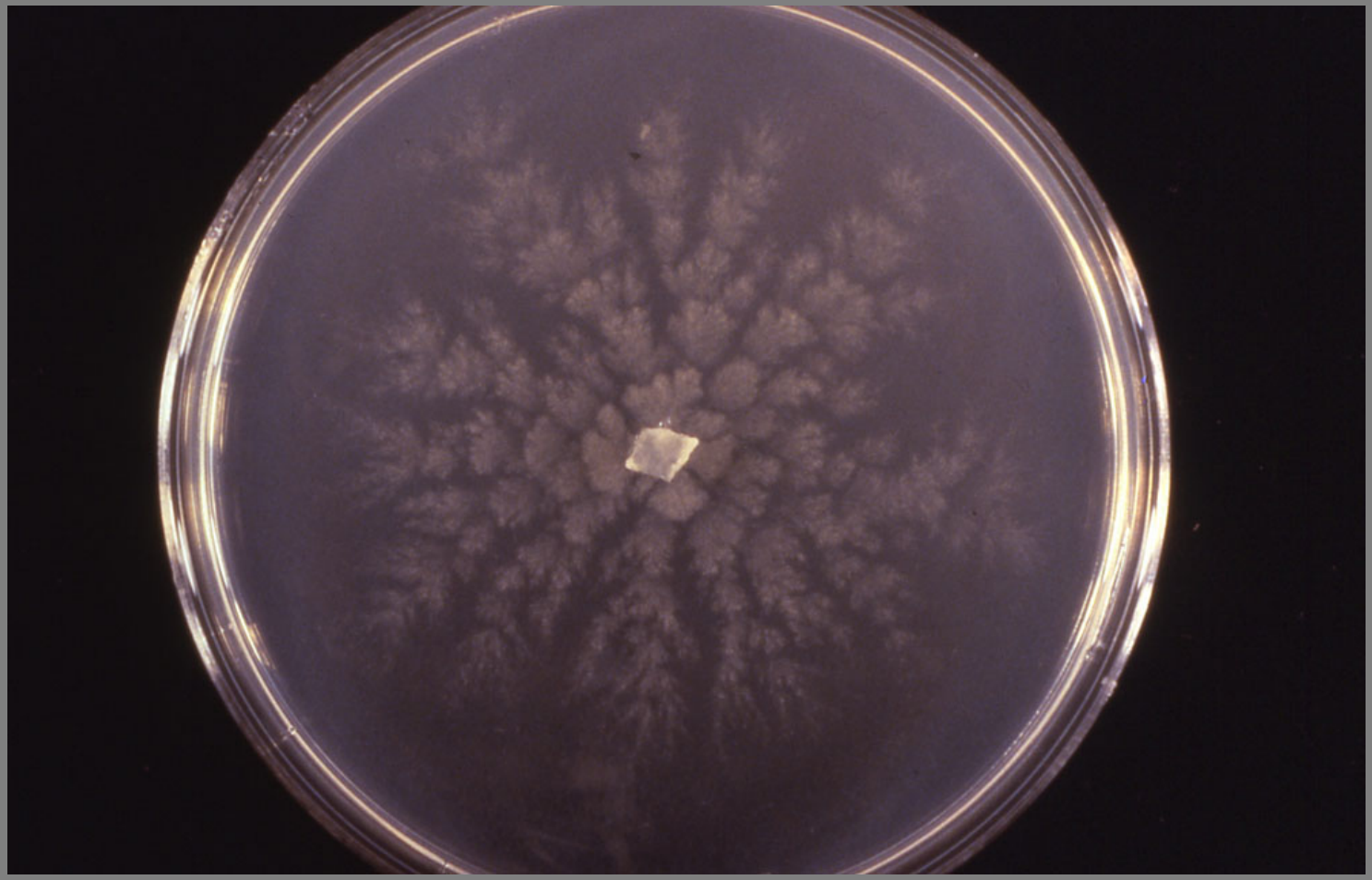
Notholithocarpus densiflorus

Big Sur
2006
K. Frangioso

Ecological Impacts

- There are about 110 species of birds which breed in California's oak woodlands. Another 60 or so species use oak woodlands outside the breeding season.
- 105 mammal species.
- 58 amphibians and reptiles.
- An estimated 5,000 species of insects.
- An unknown number of microbes.
- Wide variety of other trees, shrubs and flowering plants which co-exist with oak woodlands.

P. ramorum growing in a Petri dish



Organism new to science

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

***Phytophthora ramorum* as the Cause of Extensive Mortality of *Quercus* spp. and *Lithocarpus densiflorus* in California**

D. M. Rizzo, Department of Plant Pathology, University of California, Davis 95616; **M. Garbelotto,** Department of Environmental Science, Policy and Management, Ecosystem Science Division, University of California, Berkeley 94720; **J. M. Davidson** and **G. W. Slaughter,** Department of Plant Pathology, University of California, Davis; and **S. T. Koike,** University of California Cooperative Extension, 1432 Abbott Street, Salinas, CA 93901

Tanoak vs. Oak mortality

- Tanoak – Big Sur
  - 70%
- Marin – predicted
  - 15 years for 90%

All size classes

Infection yearly if rain

Leaves infectious

Small number of sporangia necessary

- Oak- Big Sur
  - 40%
- Marin – predicted
  - 35 years for 90%

Small trees not affected

Infection only when Spring rainfall high

Not infectious

Confirmed Susceptible Species

Andrew's clintonia bead lily
Ardisia
Bearberry
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
Chinese guger tree
Coast live oak
Coast redwood
Dogwood species
Douglas fir
Eastern Joy Lotus Tree
European ash
European turkey oak
European yew
Evergreen huckleberry
Evergreen maple
False Solomon's seal
Formosa firethorn
Fetterbush
Goat willow
Grand fir
Griselinia
Holly
Holly olive
Holm oak
Horse chestnut
Hybrid witchhazel
Japanese evergreen oak
Laurustinus
Leucothoe species
Lilac
Loropetalum species
Madrone
Magnolia varities
Manzanita
Michelia
Mountain laurel
Myrtle-leafed Distylium
Northern red oak
Oleander
Oregon ash
Oregon grape
Osmanthus
Pacific yew
Persian ironwood
Pieris varieties
Planetree maple
Poison oak
Prunus species
Red fir
Red lotus tree
Red tip photinia
Redwood ivy
Rhododendron species
Roble beech
Rosa species & hybrids
Rugosa rose
Salal
Salmonberry
Scotch heather
Scribbly gum
Sessile oak
Sheep laurel
Shreve's oak
Southern red oak
Spicebush
Spike witch hazel
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

Confirmed Susceptible Species

Andrew's clintonia bead lily
Ardisia
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Mountain laurel
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Pieris varieties
Planetree maple
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Prunus species
Red fir
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Roble beech
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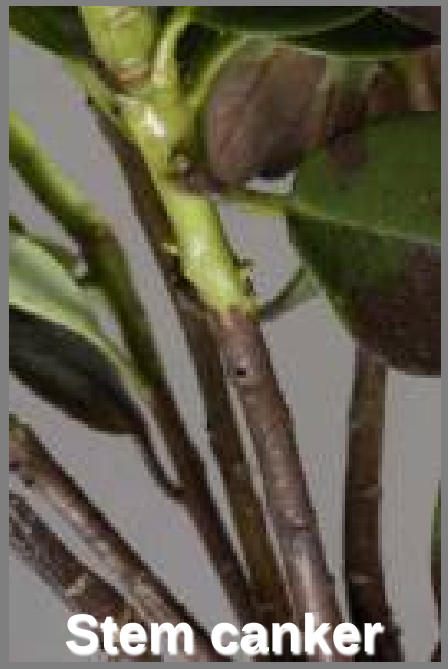


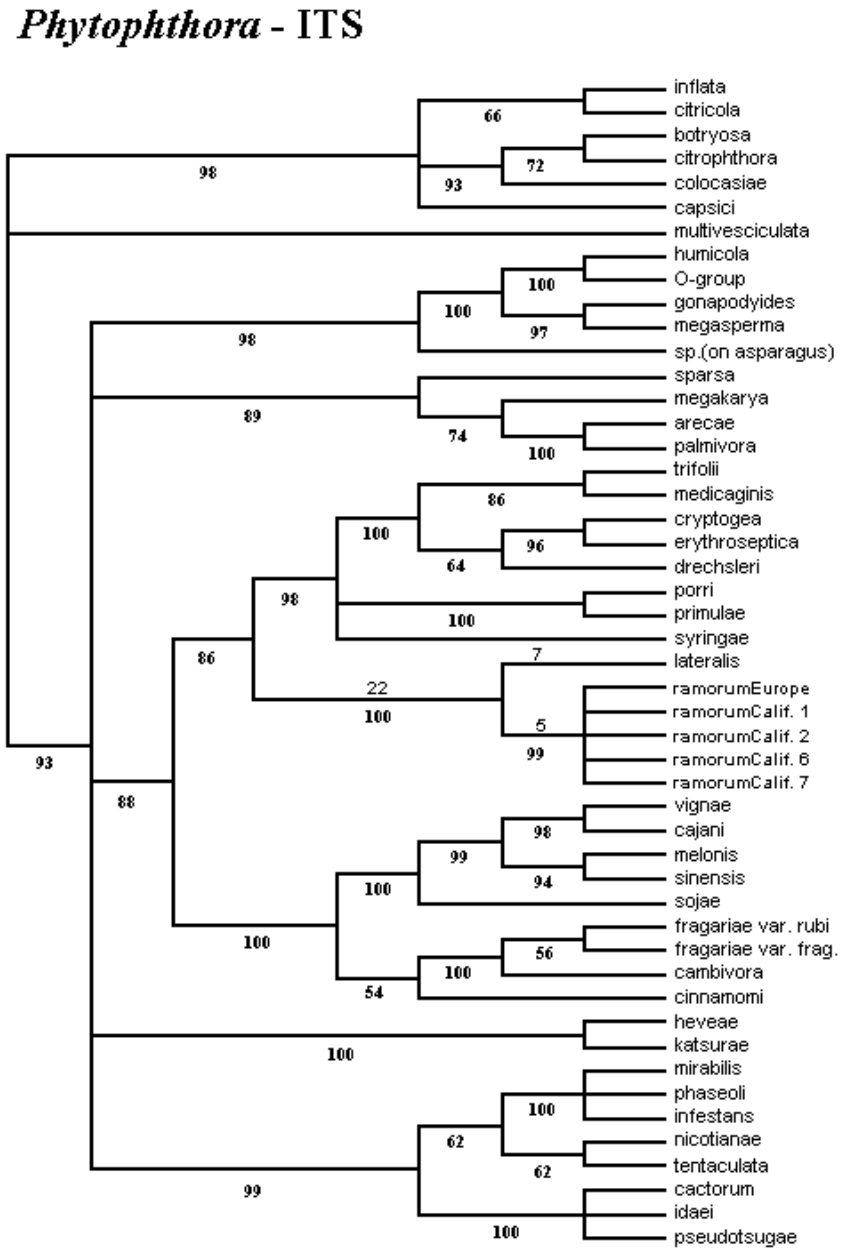
Rhododendron:

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

Use of DNA and genome information to:

- Identify organism, even directly from plants and the environment
- Determine pathways of spread and introduction
- Determine mechanisms and rates of transmission and type of reproduction
- Determine evolution and how to fight it

Phytophthora ramorum



More problems

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

Douglas-fir

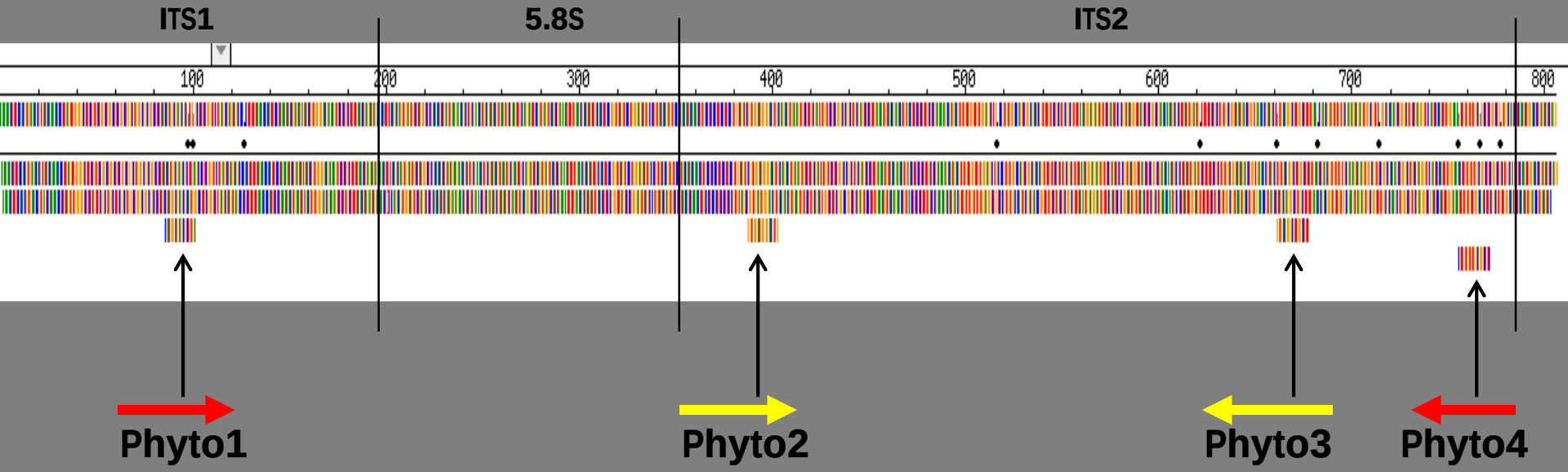
Buckeye

Redwood

DNA-based diagnostics

Designed 2 sets of *P. ramorum* specific primers

However assays used for regulatory purposes need to fulfill some stricter requirements than those used solely for research




ViewSonic

Host list expanding vertiginously rapidly

- Each confirmed host immediately regulated to avoid further spread
- Regulated hosts and the industries that depend on them highly impacted economically
- Peer reviewed publication process too slow

- Using a combination of classical and modern approaches, host infection was confirmed in 2 independent labs (UCB and UCD)

- Information was released to office of UC president, to CDFA, and to APHIS 2 days prior the ad hoc press release about new host

- Nonetheless, this caused host-ility in the industries affected by discovery

CALIFORNIA OAK MORTALITY TASK FORCE

What is Sudden Oak Death?

Sudden Oak Death is a tree disease caused by the plant pathogen *Phytophthora ramorum*. The disease kills some oak species and has had devastating effects on forests in California and Oregon.
Read more about Sudden Oak Death.

Search

Quick Links

- Current newsletter
- SODMAP
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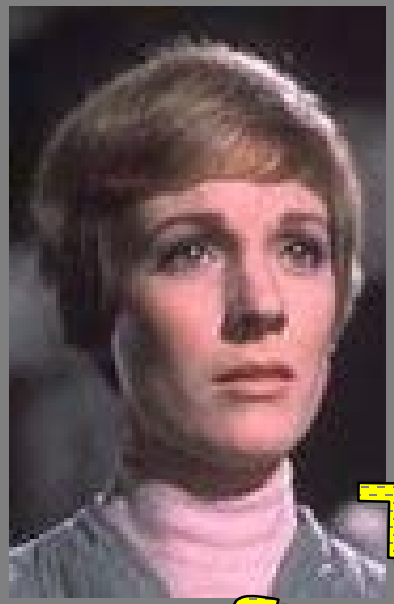
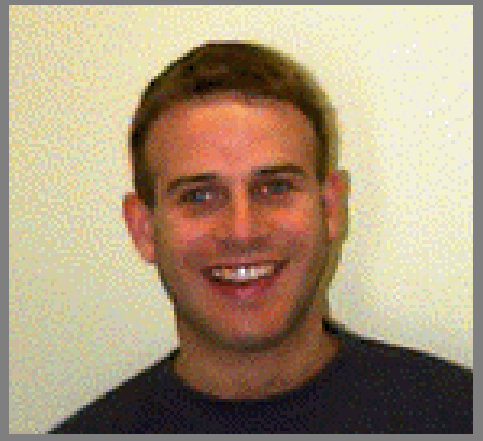
- Regulations
- Research
- Join the mailing list
- Media inquiries

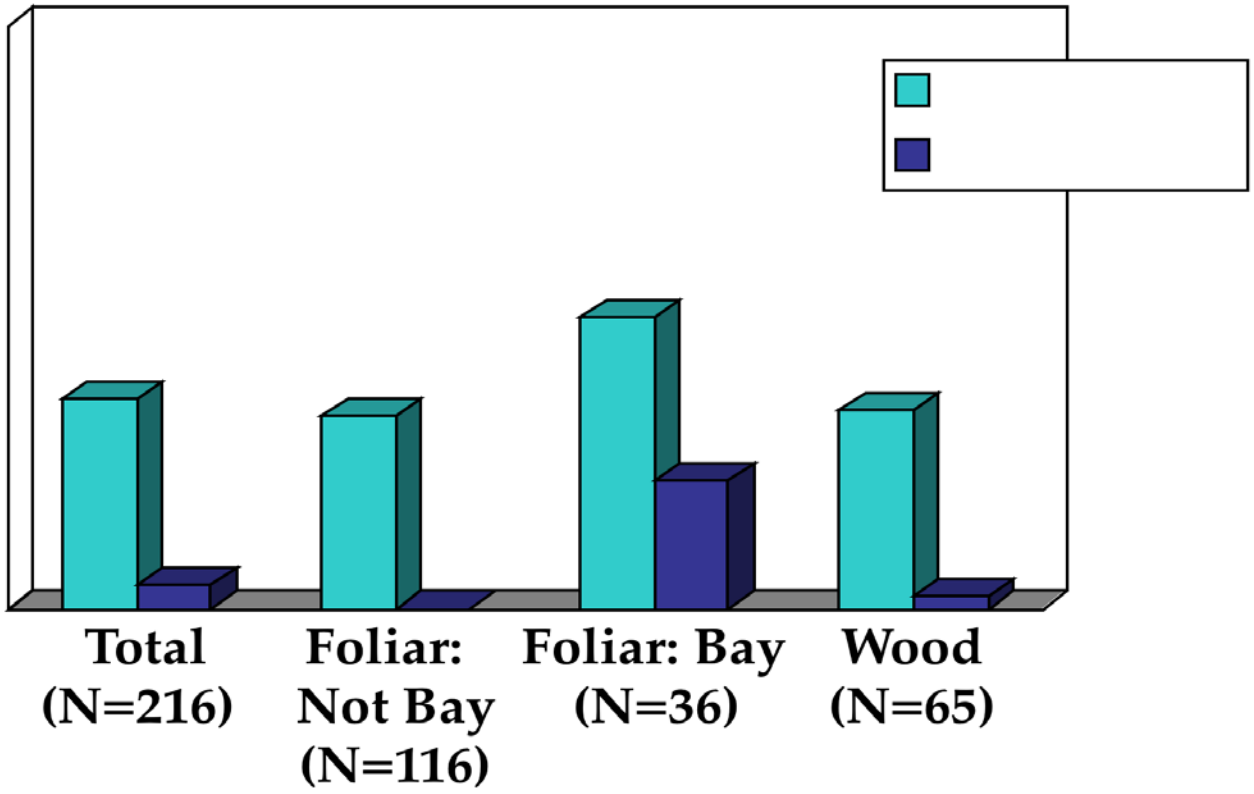
Symptom Gallery



These are some of our favorite hosts....



Culture versus nested PCR



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

Validation of a real-time PCR method for the detection of *Phytophthora ramorum*[1]

A. Chandelier[1], K. Ivors[2], M. Garbelotto[3], J. Zini[1], F. Laurent[1] and M. Cavelier[1]

[1]*Walloon Agricultural Research Centre, Department of Biological Control and Plant Genetic Resources, rue de Liroux, 4, B-5030 Gembloux (Belgium); e-mail: chandelier@cra.wallonie.be*

[2]*Department of Plant Pathology, North Carolina State University, 455 Research Drive, Fletcher, North Carolina 28732 (USA)*

[3]*Ecosystem Sciences Division, ESPM, University of California, 151 Hilgard Hall #3110, Berkeley, California 94720 (USA)*

To validate a real-time PCR method for the detection of *Phytophthora ramorum*, an intra-laboratory

Techniques

TaqMan Chemistry for *Phytophthora ramorum* Detection and Quantification, with a Comparison of Diagnostic Methods

Katherine Hayden, Kelly Ivors, Carla Wilkinson, and Matteo Garbelotto

Plant Pathology (2009) Doi: 10.1111/j.1365-3059.2009.02209.x

Diagnostic sensitivity and specificity of different methods used by two laboratories for the detection of *Phytophthora ramorum* on multiple natural hosts

A. M. Vettraino[a], S. Sukno[bc], A. Vannini[a] and M. Garbelotto[d*]

[a]*University of Tuscia, S. Camillo de Lellis snc, 01100 Viterbo, Italy;* [b]*Department of Plant Pathology and Microbiology, Texas A&M University, 2132 College Station, TX 77845, USA;* [c]*Centro Hispano-Luso de Investigaciones Agrarias (CIALE), Departamento de Microbiología y Genética, Universidad de Salamanca, Calle Del Duero 12, 37185 Villamayor, Salamanca, Spain; and* [d]*Department of Environmental Science, Policy and Management, University of California, Berkeley, 137 Mulford Hall, Berkeley, CA 94720-3114, USA*

In spite of data submitted to CDFA…

- Lobbying power of industry maintained diagnosis in a “business as usual” status

- Then, in the spring of 2004 hundreds of thousands of plants that had been misdiagnosed as healthy were shipped from California and Oregon to 22 States, causing the largest trace-back/trace-forward operation in the history of the country

In the Fall of 2004….

The assay we developed became the first DNA assay to diagnose non viral plant pathogens. Now diagnosis of most invasive regulated microbes is DNA-based in North America, Australia, European Union, China, etc.

efsa
European Food Safety Authority
Committed to ensuring that Europe's food is safe

Make a difference to European food safety – join EFSA's Scientific Panels

Are you a scientist with experience of chemical risk assessment? Are you interested in making a difference to European food safety? Then apply to join two of EFSA's Scientific Panels dealing with food ingredients and packaging and be a part of Europe's network of top food safety scientists. EFSA's experts help to protect European consumers by delivering high-calibre independent scientific advice to European decision-makers on food and feed safety. Successful applicants will be appointed as a Panel member for a three-year term starting in July 2014. Scientists are invited to apply by 17 June 2013.

Press release: EFSA seeks new experts to join two Scientific Panels

Call for expressions of interest for membership of EFSA's Scientific Panels

EFSA's Scientific Panels

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Featured Topics

- Zoonotic diseases
- Antimicrobial Resistance
- Bisphenol A
- Animal Welfare
- Aspartame
- Independence

All topics

Ornamentals as spread-routes?

- Is *P. ramorum* a previously unnoticed native species?
- Is it an exotic organism?
  - -High level of mortality
  - -Limited range
  - -Suggest exotic, but need to prove that point

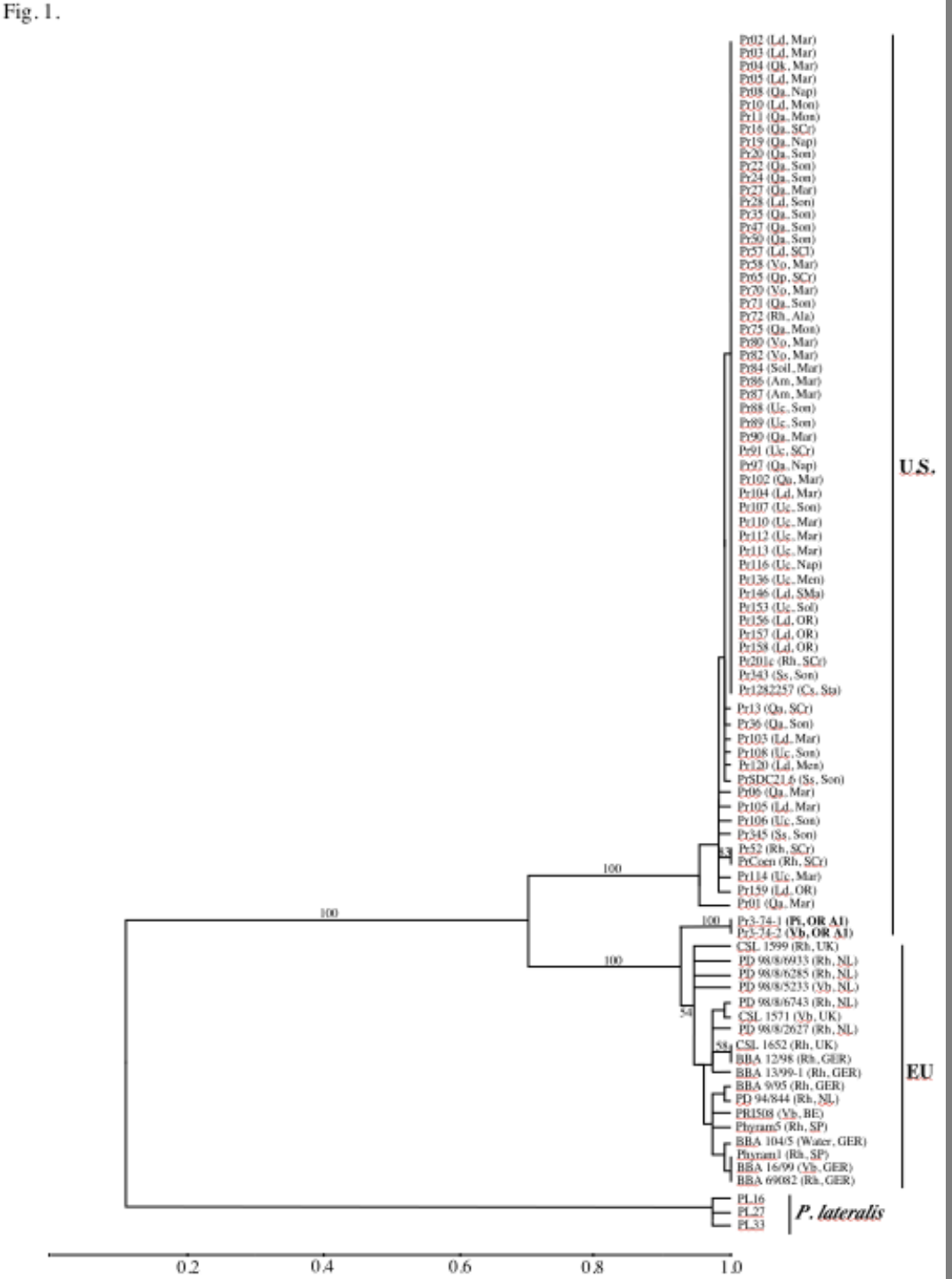
Genetic structure

- Need a number of independent and neutral DNA markers
- Are our isolates the same as the European ones?
- Is the genetic structure suggestive of an introduced or native species?

Phytophthora Genome Sequences Uncover Evolutionary Origins and Mechanisms of Pathogenesis

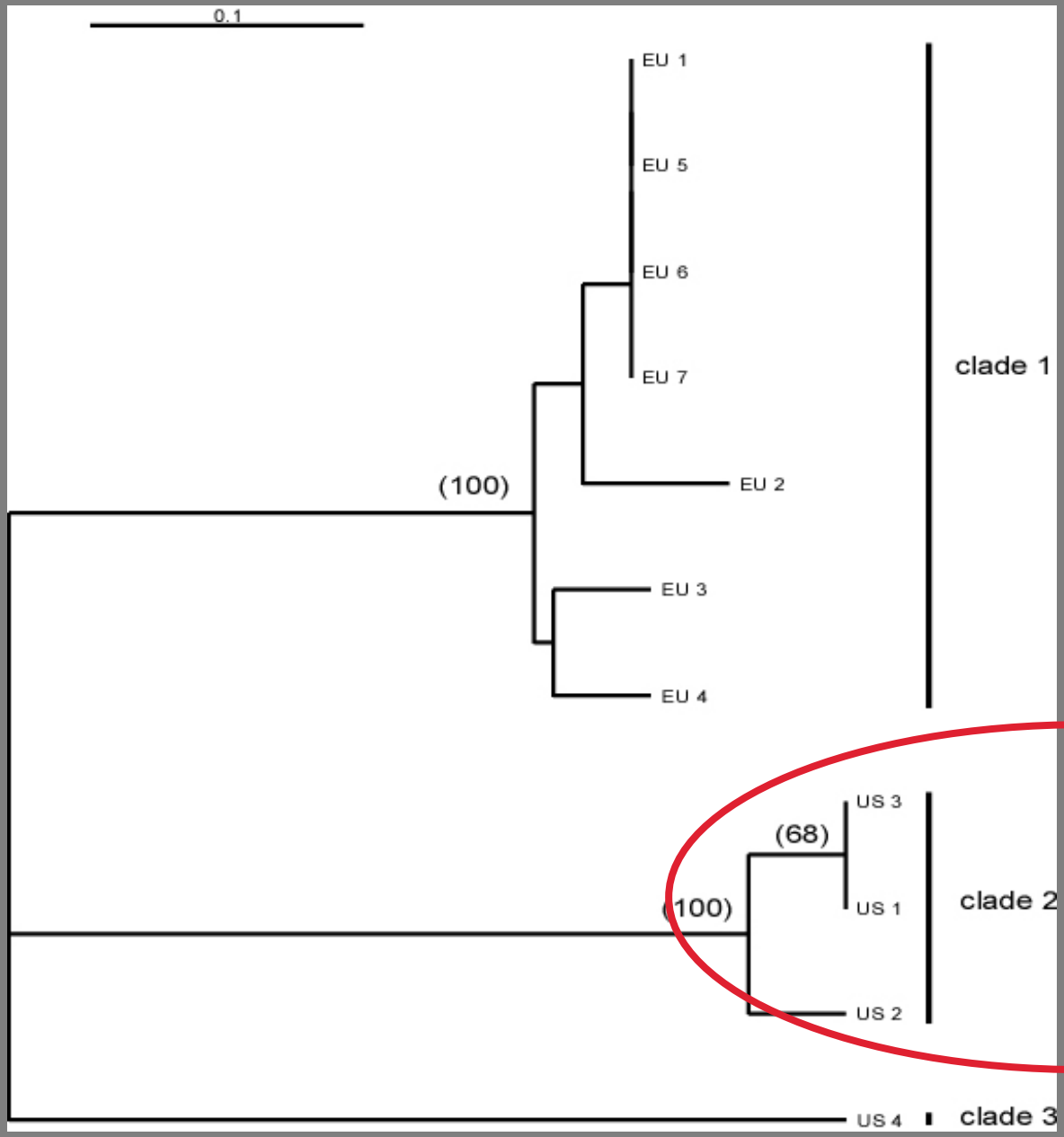
Brett M. Tyler,[1*] Sucheta Tripathy,[1] Xuemin Zhang,[1] Paramvir Dehal,[2,3] Rays H. Y. Jiang,[1,4] Andrea Aerts,[2,3] Felipe D. Arredondo,[1] Laura Baxter,[5] Douda Bensasson,[2,3,6] Jim L. Beynon,[5] Jarrod Chapman,[2,3,7] Cynthia M. B. Damasceno,[8] Anne E. Dorrance,[9] Daolong Dou,[1] Allan W. Dickerman,[1] Inna L. Dubchak,[2,3] Matteo Garbelotto,[10] Mark Gijzen,[11] Stuart G. Gordon,[9] Francine Govers,[4] Niklaus J. Grunwald,[12] Wayne Huang,[2,14] Kelly L. Ivors,[10,15] Richard W. Jones,[16] Sophien Kamoun,[9] Konstantinos Krampis,[1] Kurt H. Lamour,[17] Mi-Kyung Lee,[18] W. Hayes McDonald,[19] Mónica Medina,[20] Harold J. G. Meijer,[4] Eric K. Nordberg,[1] Donald J. Maclean,[21] Manuel D. Ospina-Giraldo,[22] Paul F. Morris,[23] Vipaporn Phuntumart,[23] Nicholas H. Putnam,[2,3] Sam Rash,[2,13] Jocelyn K. C. Rose,[24] Yasuko Sakihama,[25] Asaf A. Salamov,[2,3] Alon Savidor,[17] Chantel F. Scheuring,[18] Brian M. Smith,[1] Bruno W. S. Sobral,[1] Astrid Terry,[2,13] Trudy A. Torto-Alalibo,[1] Joe Win,[9] Zhanyou Xu,[18] Hongbin Zhang,[18] Igor V. Grigoriev,[2,3] Daniel S. Rokhsar,[2,7] Jeffrey L. Boore[2,3,26,27]

Human genome just completed when *P. ramorum* was discovered: one of the first species to be fully sequenced, and the eukaryotic species with the shortest time between discovery and sequencing (3 years)



- 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 sex and recombination in US nurseries
- US forest population is genetically very homogeneous, trademark of an introduced species

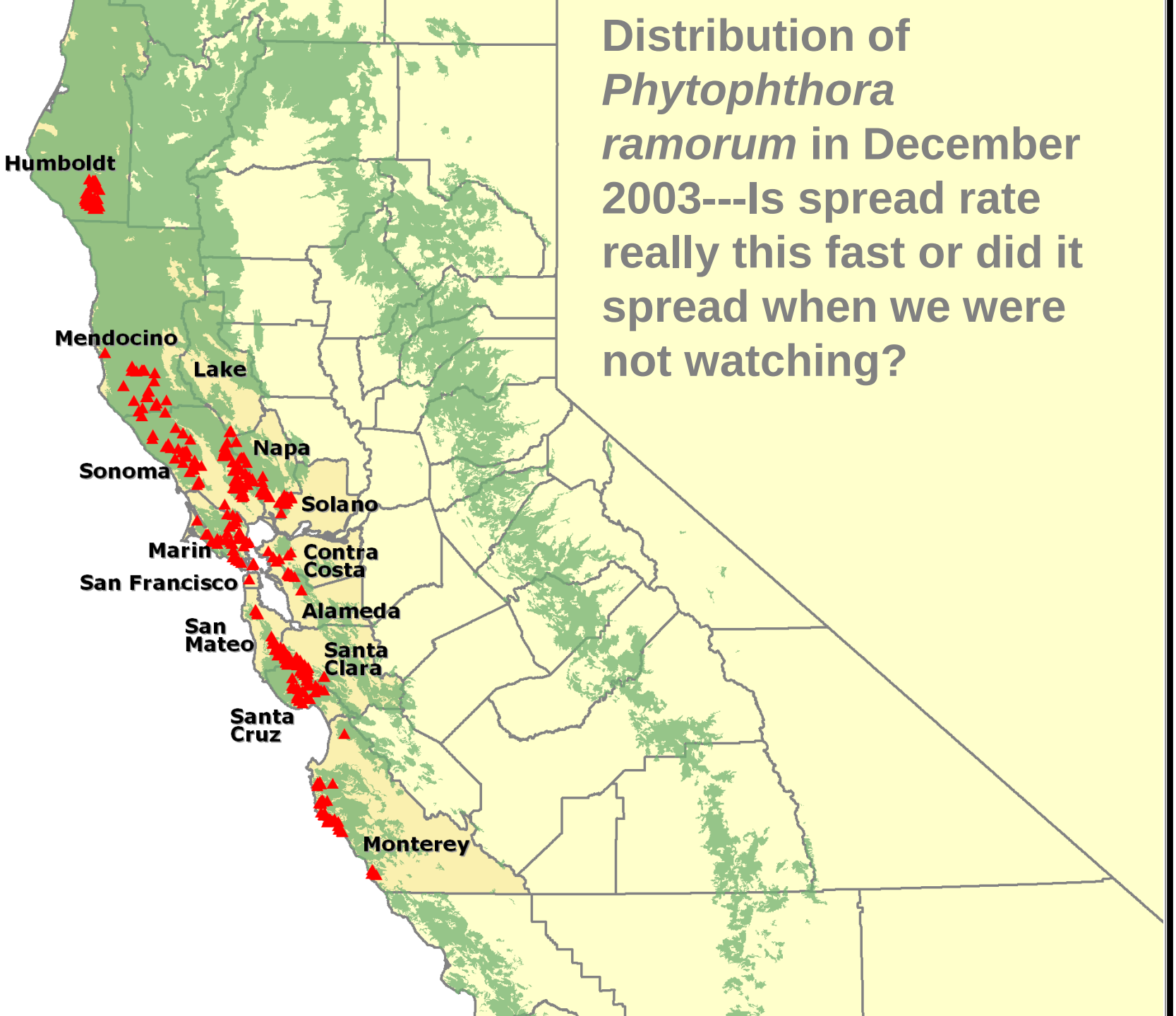
There are actually three different lineages



Mating Type	Growth Rate
EU1	Fast
NA1	Slow
NA2	Fast

Can these data be used to infer which came first…

- FORESTS
  - NA1
- NURSERIES
  - NA1
  - NA2
  - EU2


Distribution of *Phytophthora ramorum* in December 2003---Is spread rate really this fast or did it spread when we were not watching?
Humboldt
Mendocino
Lake
Napa
Sonoma
Solano
Marin
Contra Costa
San Francisco
Alameda
San Mateo
Santa Clara
Santa Cruz
Monterey

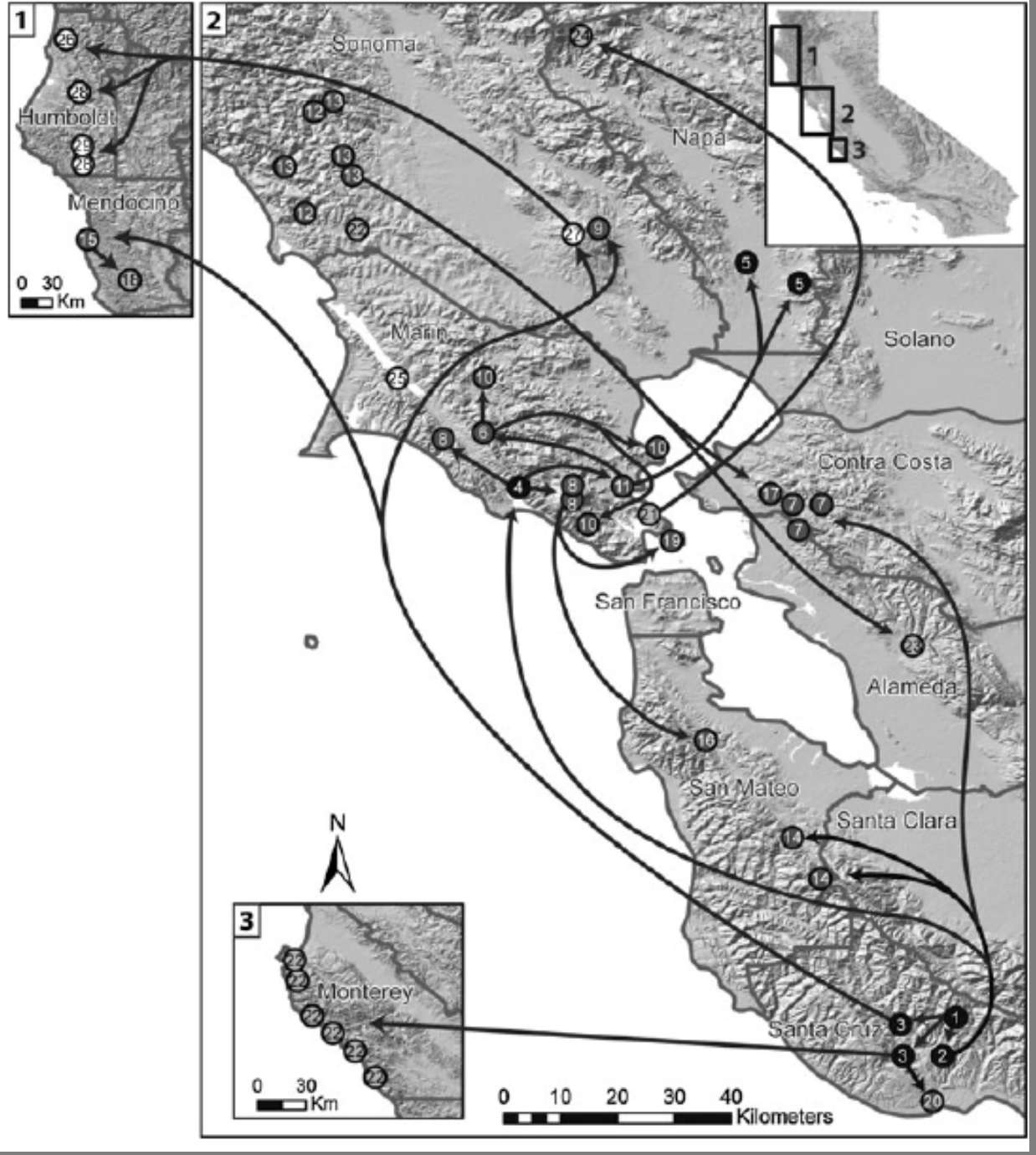
Genotyped approximately 1000 isolates from all known forest sites and from a number of nurseries

Biol Invasions
DOI 10.1007/s10530-013-0453-8

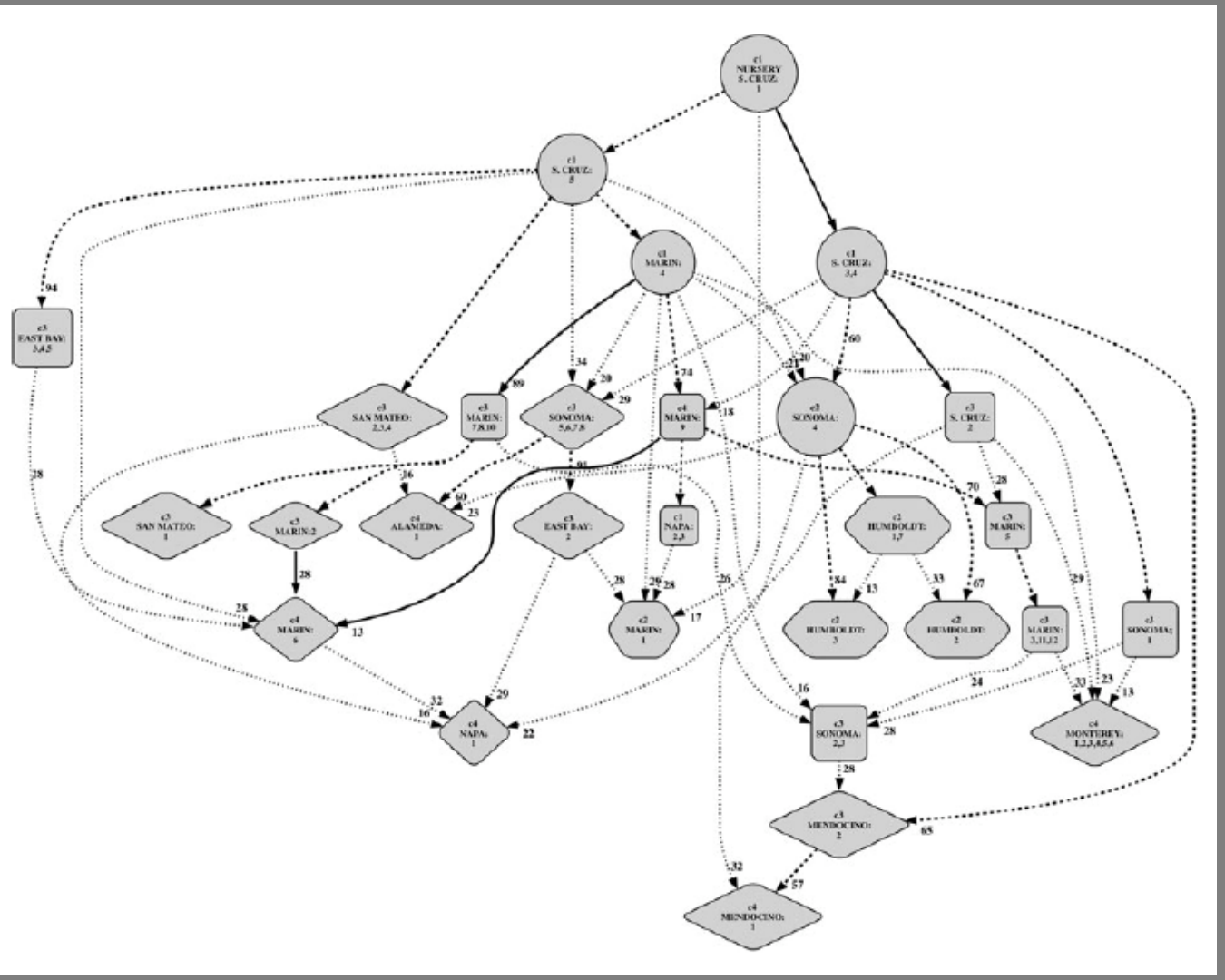
ORIGINAL PAPER

Combining field epidemiological information and genetic data to comprehensively reconstruct the invasion history and the microevolution of the sudden oak death agent *Phytophthora ramorum* (Stramenopila: Oomycetes) in California

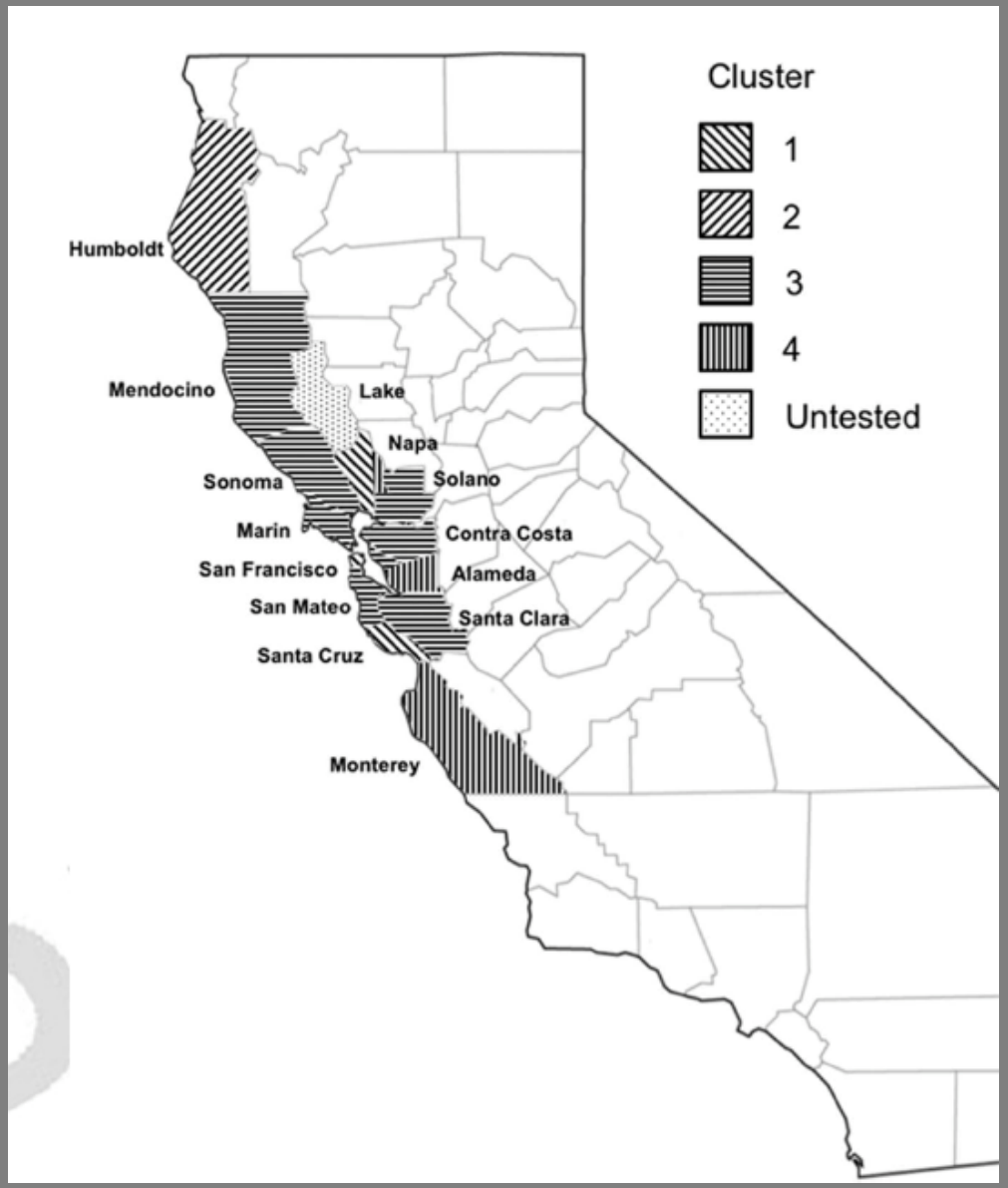
Peter J. P. Croucher · Silvia Mascheretti · Matteo Garbelotto



Long distance movement of identical strains proves human involvement: *P.ram* introduced multiple times



Using coalescence reconstructed most of the history of the disease



Cluster 1 of strains is the original introduced, but others are more widespread

Results

- Nurseries are basal, but where nurseries got it from still a mystery
- Introduced multiple times (at least 12 successful introductions throughout Northern California)
- It moves at relative short distances, but we show the greater the infestation, the more likely it will become a source

Disease Mitigation

- By reducing size of infestations we will prevent further spread (now that ornamental plants are regulated)

- We can base control on risk levels, risk level becomes high only when pathogen is **200 m** from oaks

- Different stakeholders, huge metropolitan area, rural areas, different interests, tribes vs. silicon valley..

Pathogen

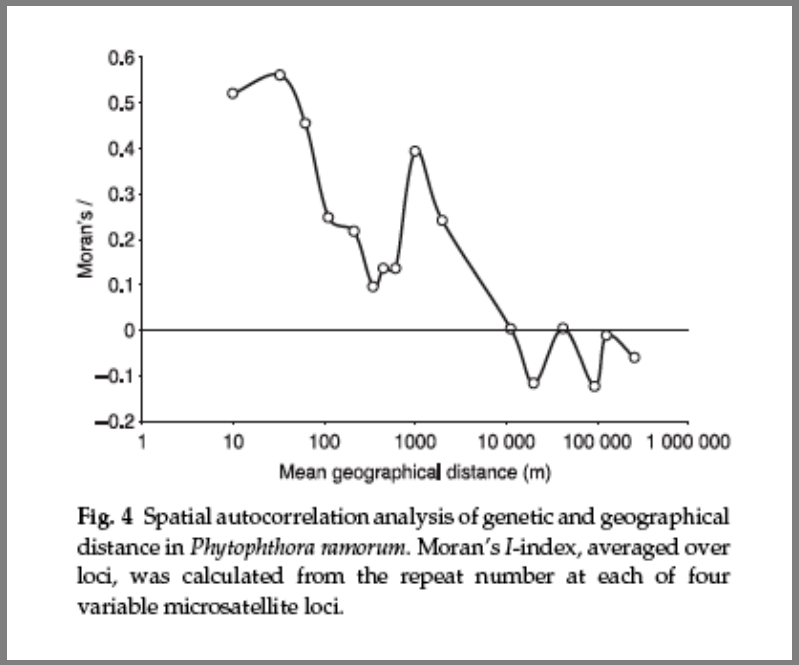


Fig. 4 Spatial autocorrelation analysis of genetic and geographical distance in *Phytophthora ramorum*. Moran's *I*-index, averaged over loci, was calculated from the repeat number at each of four variable microsatellite loci.

When multiple rain events occur sporangia are produced on leaves of infectious hosts such as bay laurel and tanoak and can be airborne mostly up to 100 m but when winds are strong up to 2 miles

EFFECTS OF SOD

- Ecological disaster: 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

Plant Pathologists at the Center of a Circus

A devastating oak disease has reshaped two scientists' careers

SOQUEL DEMONSTRATION FOREST, CALIFORNIA—Matteo Garbelotto stands under the redwoods and strips. The plant pathologist from the University of California (UC), Berkeley, is in the Santa Cruz Mountains to experiment on a tree-killing microbe and, as usual, things aren't going as planned. A fellow researcher forgot the cheesecloth used to cover inoculation wounds in trees, so Garbelotto tears up his undershirt as a substitute. He redresses and smiles sheepishly, as if to say, "This is just another day at the office."

Unexpected acts like this one—part performance, part sacrifice—have become the norm for Garbelotto ever since he became one of two lead scientists investigating sudden oak death. The disease is strangling oak trees in the hills of coastal Northern California and threatening to spread to other forest species; in one scenario, it could leap across the continent to eastern forests, potentially costing billions of dollars. Garbelotto's dramatic bent—inherited from his mother, an Italian actress—comes in handy. Sudden oak death has thrust him and research partner David Rizzo of UC Davis into the middle of a three-ring circus where sci-

The oak death duo. Matteo Garbelotto (left) and David Rizzo with a specimen of the pathogen that has strangled thousands of California oaks.

20 or 50 years [to understand] some diseases," says plant pathologist Clive Brasier of the U.K. Forestry Commission.

Budding disaster

In the spring of 2000, before Rizzo and Garbelotto had ever worked together, they visited a sunny hillside north of San Francisco that bore a dark omen—a stain on the trunk of a coast live oak oozing black sap. Some researchers thought beetles were to blame, but by January, the two had identified the cause as a water mold called *Phytophthora ramorum*, a funguslike relative of brown algae previously known only in rhododendrons in Germany

a symbol of California.

The story of a scourge that could devastate the Golden State's beloved forests and manicured backyards grabbed the media's attention. The humble field of plant pathology became headline news, and Rizzo and Garbelotto have been roped into almost 1000 media interviews since the outbreak began.

Rizzo, 42, traces his start in the field to his college days, when he hiked frequently in the Blue Ridge Mountains of the Appalachians. There he saw the stumps of chestnuts, once one of the tallest trees in the eastern United States. Between 1900 and 1940, a blight eliminated 3.5 billion of them. From this grew Rizzo's fascination with the power of microscopic organisms to change whole forests.

Garbelotto, 37, first became acquainted with fungi of the edible type: He grew up in Venice and the Italian Alps, where he and his siblings frequently gathered mushrooms for the dinner table. Later he studied pathogenic fungi.

Media reports often typecast the two personalities. Garbelotto has been called "dashing" and "colorful," while Rizzo is chronically referred to as "bookish" or even "balding." "He's the cool guy from Berkeley, and I'm the boring guy from Davis with two kids," Rizzo says.

Whatever their quirks, the two seem doomed to being cast as heroes chasing after a villain. "You have this Godzilla fungus, and the scientists are often portrayed as the Lone Ranger and Tonto," says plant pathologist Susan Frankel of the U.S. Forest Service in Vallejo, California.

When the disease hit, neither was in an ideal position to ride in on a white horse—Rizzo was focusing on pear trees and conifers, and Garbelotto was newly hired and just setting up a lab. Yet due to recent retirements, they were the only forest pathologists in the UC system, so they took the lead.

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

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

Enlist citizens to help generate a fine scale map of a forest disease

UC Berkeley a pioneer in citizen science

http://newscenter.berkeley.edu/2013/05/22/citizen-science-at-uc-berkeley/

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UC Berkeley a pioneer in citizen science

By Sarah Yang, Media Relations | May 22, 2013

BERKELEY — Calbug is just the latest example of UC Berkeley inviting the public to help with a large-scale science project. Before Calbug, there was SETI@home, one of the earliest and most successful volunteer-oriented research projects in the world. Launched in 1999, SETI@home continues to enlist citizens to download software onto their computers to analyze chunks of radio data from space in the search for extraterrestrial intelligence.

Notably, some of the earlier citizen science projects looked to the stars, including Stardust@home, built by UC Berkeley physicists. Volunteers helped find tiny particles of interstellar dust from stars many light years away from Earth with the aid of a virtual microscope that runs off of a Web browser.

Back on Earth, the public can help track sudden oak death disease through SOD Blitz, a program headed by Matteo Garbelotto, adjunct professor a Environmental Science, Policy and Management. samples for laboratory examination.

Garbelotto also relied upon citizen scientists for a Venice Museum of Natural History. Volunteers, m

http://baynature.org/articles/blitzers-search-for-sod/

Blitzers Search for SOD « Bay Nature

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Home > Articles > Blitzers Search for SOD

Blitzers Search for SOD
On the Front Lines of Oak Resea

by Sue Rosenthal on March 08, 2012 in Plants

No Comments

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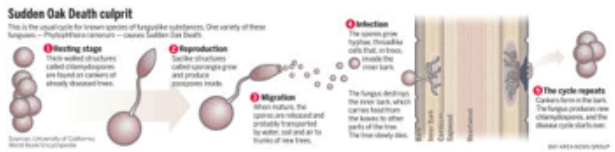
Citizen-scientists to help map Sudden Oak Death fungus

By Lisa M. Krieger

lkrieger@mercurynews.com

Posted: 04/12/2013 02:43:45 PM PDT



An army of citizen-scientists is being enlisted to help map outbreaks of the Sudden Oak Death fungus killing trees on the California coast and to perhaps control it.

The free community outreach campaign kicked off Friday in Santa Cruz. Other sessions will be held in Orinda, Berkeley, San Francisco, Saratoga, Burlingame, Woodside, Atherton and Los Altos Hills.

See more

- Find out what Sudden Oak Disease does to trees

Uncategorized

- Star Gazing and Night Hiking Resources 6 comments
- Photosynthesis in leaves that aren't green 8 comments
- Why do dragonflies swarm? 43 comments
- Stargazing in the SF Bay Area 0 comments
- Bird watching — there's an app for that! 0 comments

SOD Blitzes
16 locations in 2013
500 volunteers each year
Over20.000 samples
60.000 acres surveyed

Prepare collection packets
Train volunteers throughout State
Volunteers have 2 days to collect
Samples brought to Berkeley
Sampled Processed in Lab
Generate Map of disease Distribution
Map is available on internet
Data from map fed into Apps in real time