

Thousand Cankers Disease

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- **Thousand cankers disease (TCD) of *Juglans* species:
A little history with a California focus**
- **Research highlights**
- **Diagnostic issues and future directions**



T. Roubstova - UCD

Geosmithia morbida



Jim LaBonte – Oregon Dept Ag

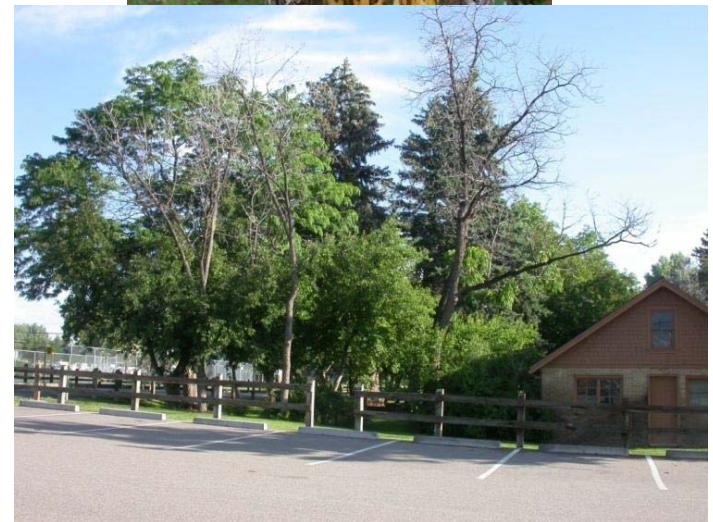
***Pityophthorus juglandis*
walnut twig beetle (WTB)**



Photo by Stacy Hishinuma

An epidemic in eastern black walnut, *Juglans nigra*, in western states

- Mortality of *J. nigra* was first observed in Utah & Oregon in the 1990's
- Mortality first observed in Colorado in 2001
- Disease widespread in Colorado Springs & Boulder by 2004
- By 2008 nearly all *J. nigra* trees in the Colorado Springs area were dead
- More than 2000 dead *J. nigra* in Boulder – Denver area
- Walnut twig beetle and new fungal pathogen associated with disease by Colorado State University scientists.



Photos by Ned Tisserat & Whitney Cranshaw, Colorado State University

Dying *Juglans hindsii* (northern CA black walnut) trees outside Davis, CA in 2008



Photos by Steve Seybold and Andy Graves



R. Bostock



R. Bostock

TCD in English walnut, *Juglans regia*

Photo by J. Hasey



Photo by C. Debuse



R. Bostock

Pityophthorus juglandis walnut twig beetle (WTB)

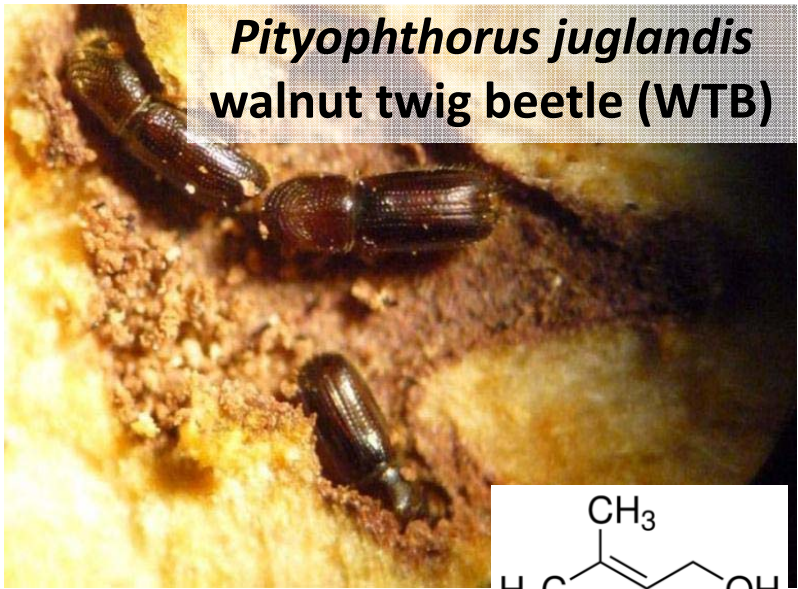
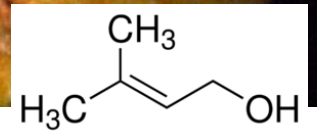
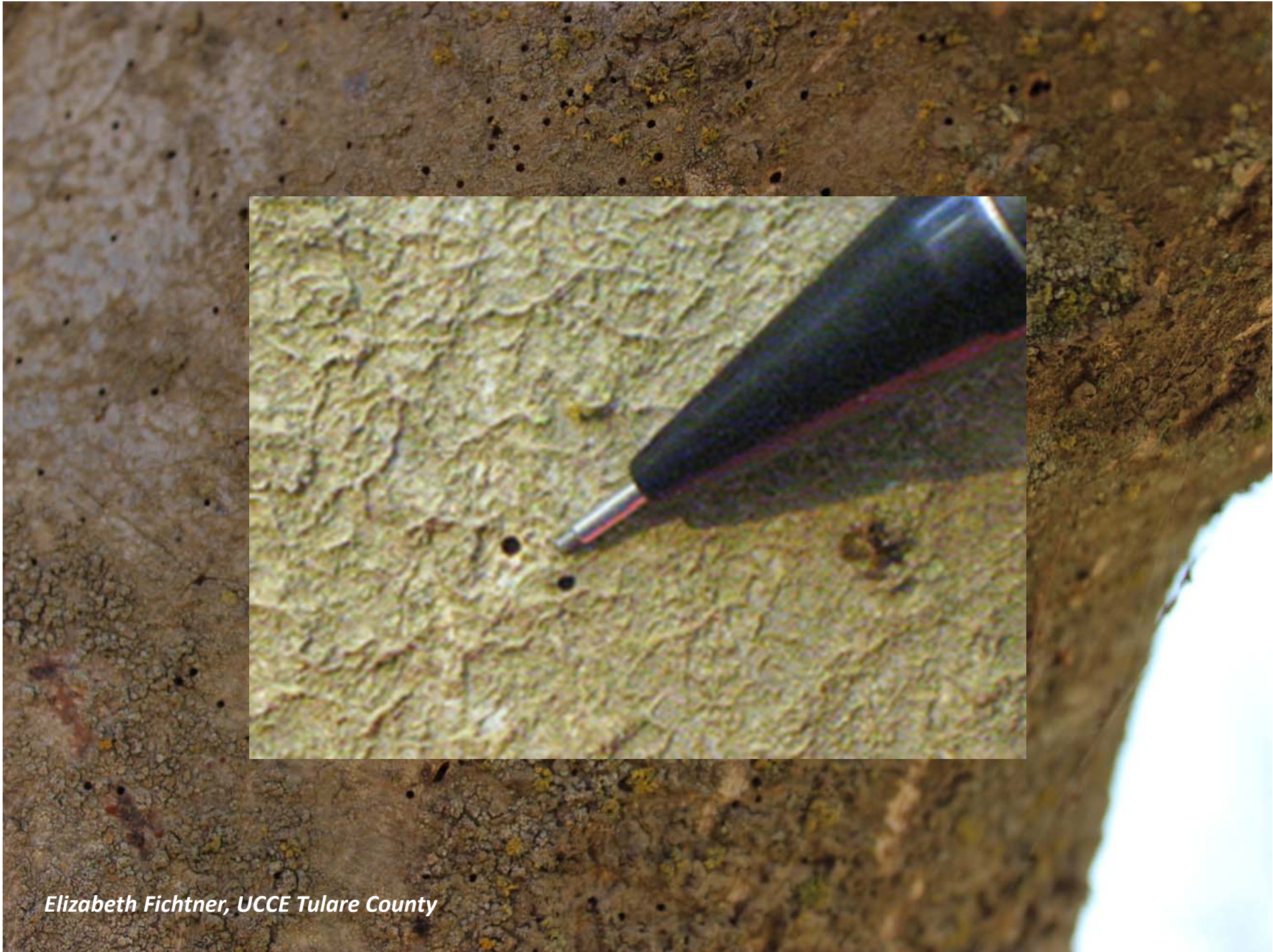


Photo by S. Seybold





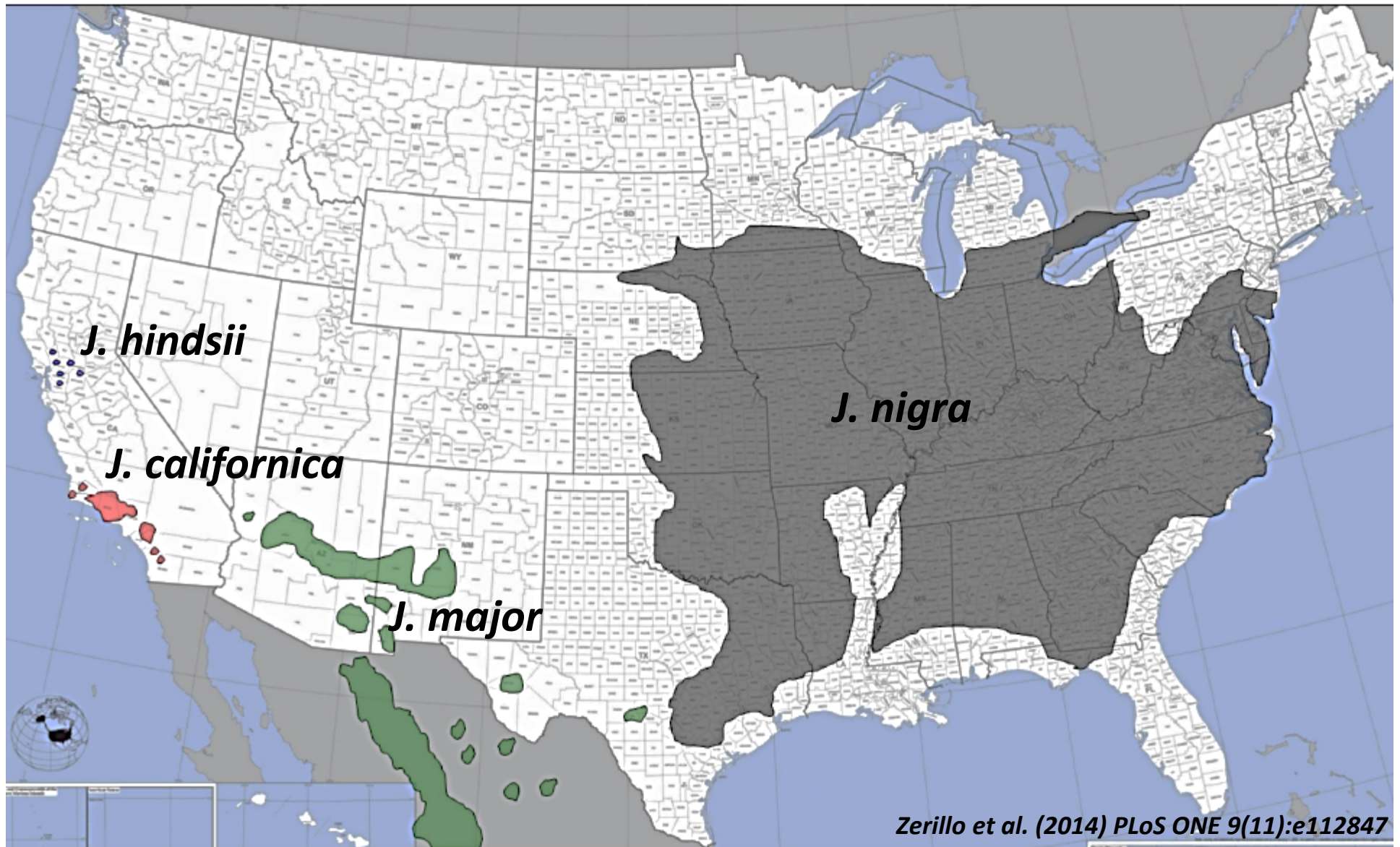
Elizabeth Fichtner, UCCE Tulare County

***J. regia* cv. Chandler**
Tulare County



Elizabeth Fichtner, UCCE Tulare County

Natural distribution of some native *Juglans* species in the USA



Comparative TCD Symptoms

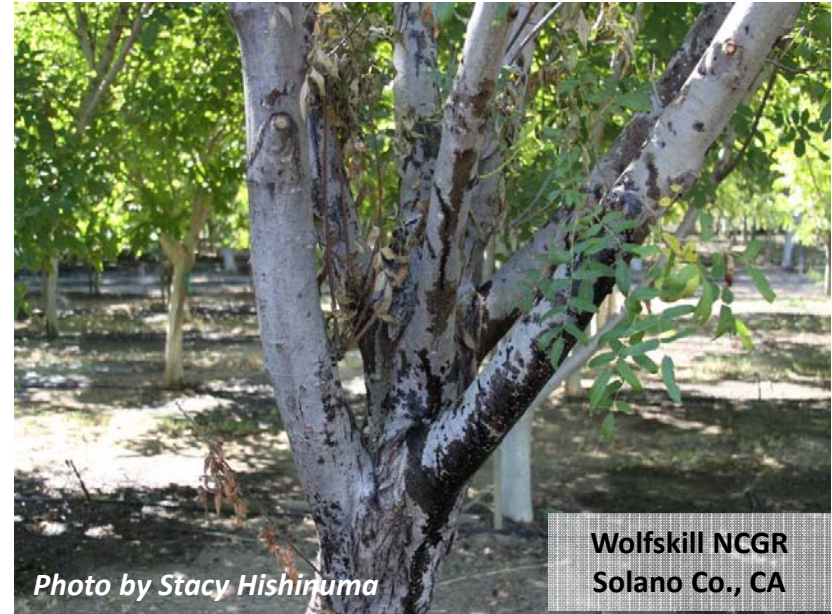


Photo by Stacy Hishinuma

Wolfskill NCGR
Solano Co., CA



Juglans major
(southern NM)

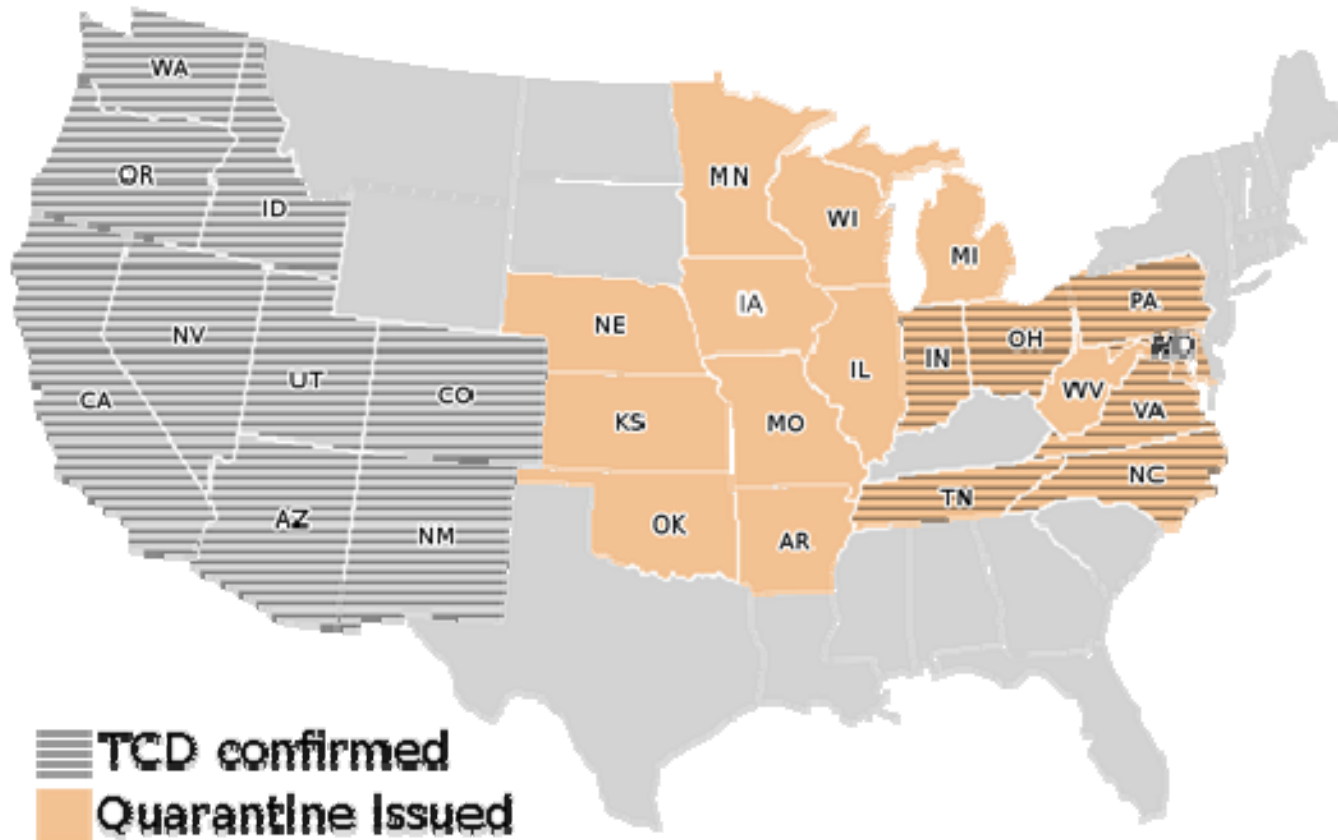
**Sporadic
cankers,
minimal staining
on bark surface**



Juglans californica
(in northern CA)

**Numerous cankers,
prolific staining on
bark surface**

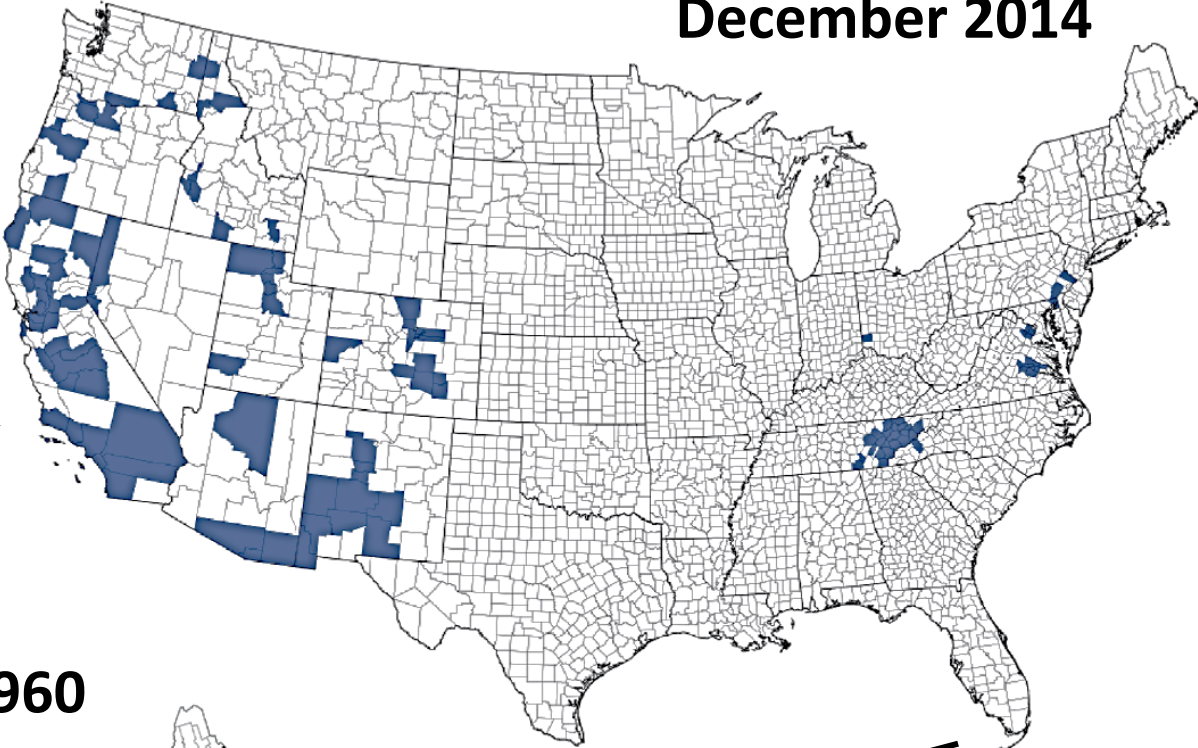
Distribution of TCD in the United States October, 2015



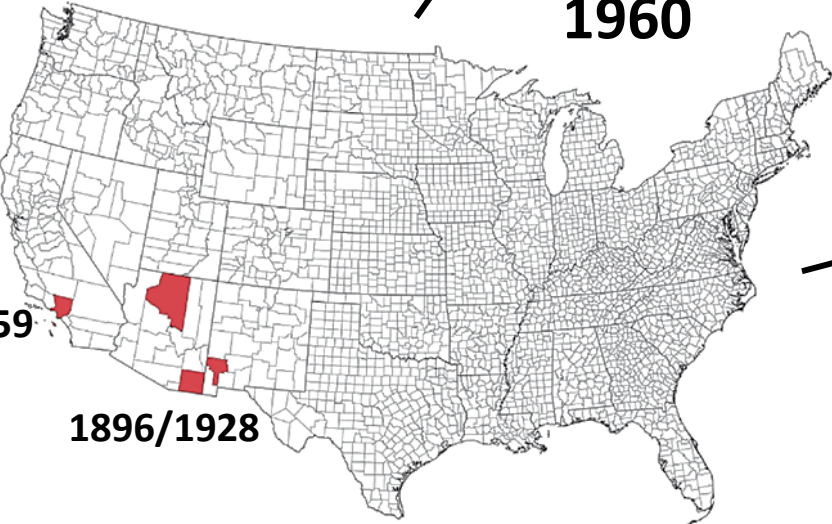
Also present in English walnut in Italy; interceptions in China!

WTB distribution in the USA

December 2014



1960

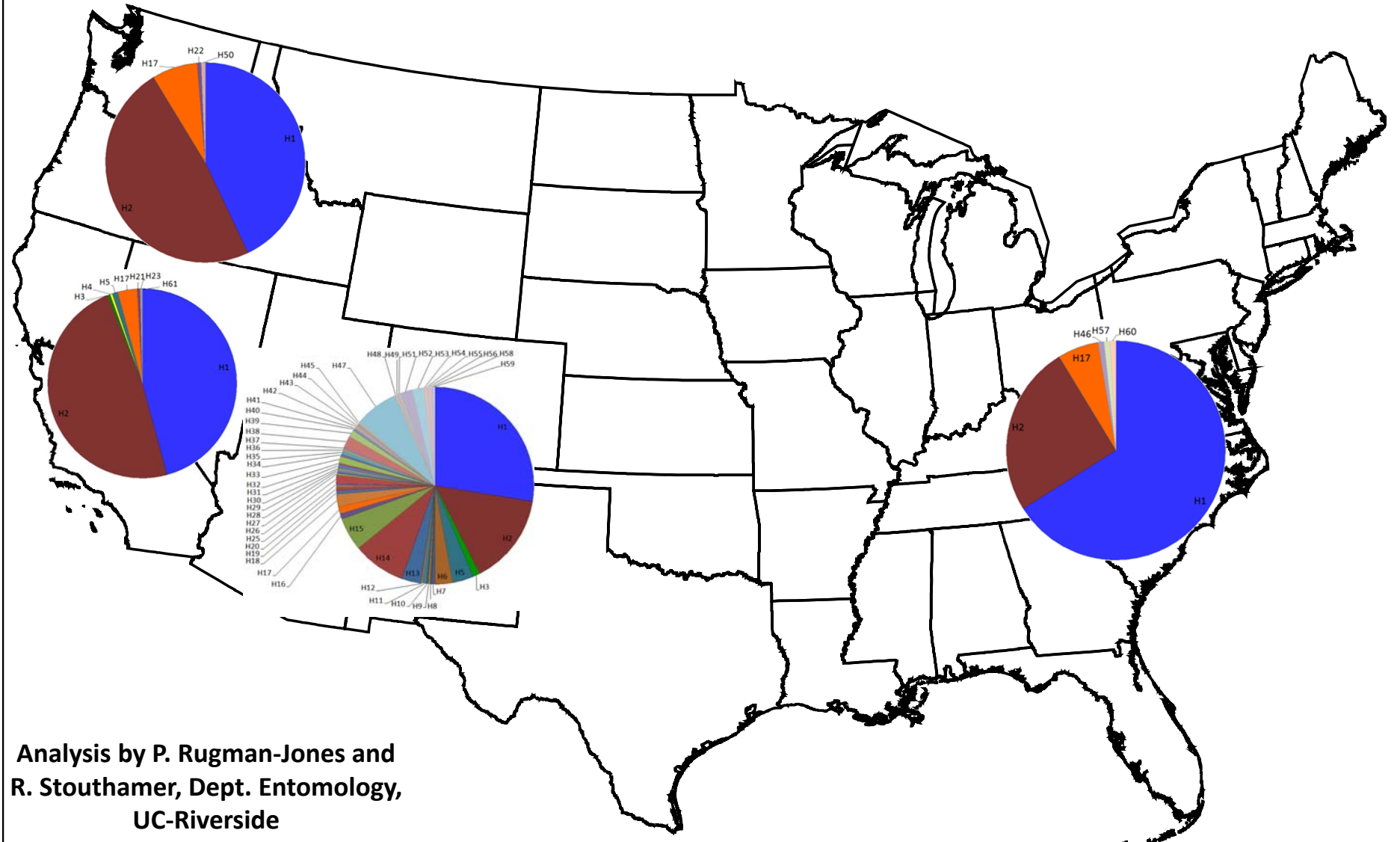


1959

1896/1928

from Rugman-Jones et al. (2015) PLoS ONE 10(2):e0118264

Genetic analyses of WTB mitochondrial haplotypes suggest migrations from southwest U.S. to other regions



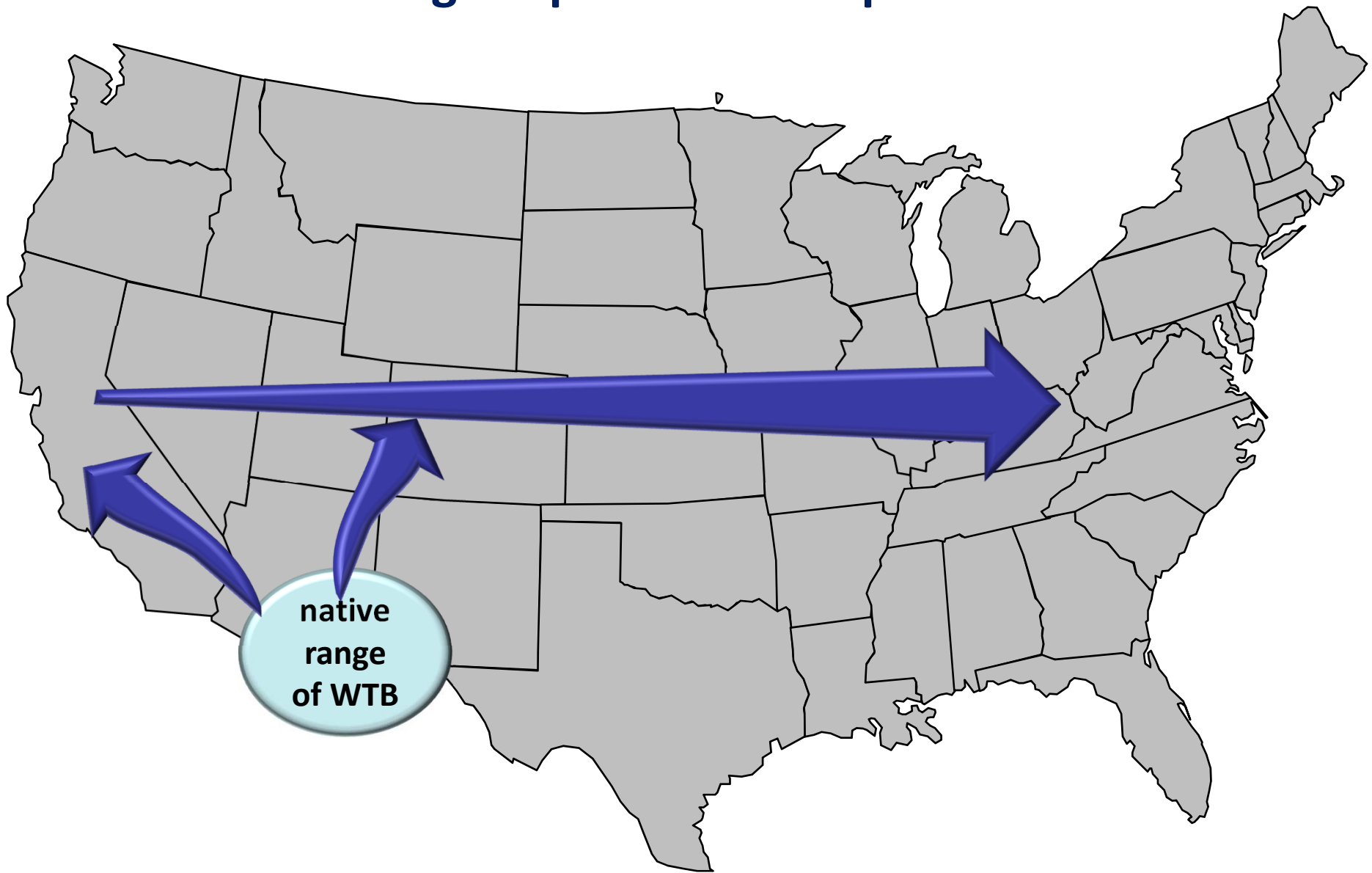
Analysis by P. Rugman-Jones and
R. Stouthamer, Dept. Entomology,
UC-Riverside

Graphic by A.D. Graves,
USDA FS FHP, Albuquerque

Rugman-Jones et al. (2015) PLoS ONE 10(2):e0118264

[Phylogram](#)

WTB range expansion and spread of TCD?



Zerillo et al. (2014) PLoS ONE 9(11):e112847

Rugman-Jones et al. (2015) PLoS ONE 10(2):e0118264



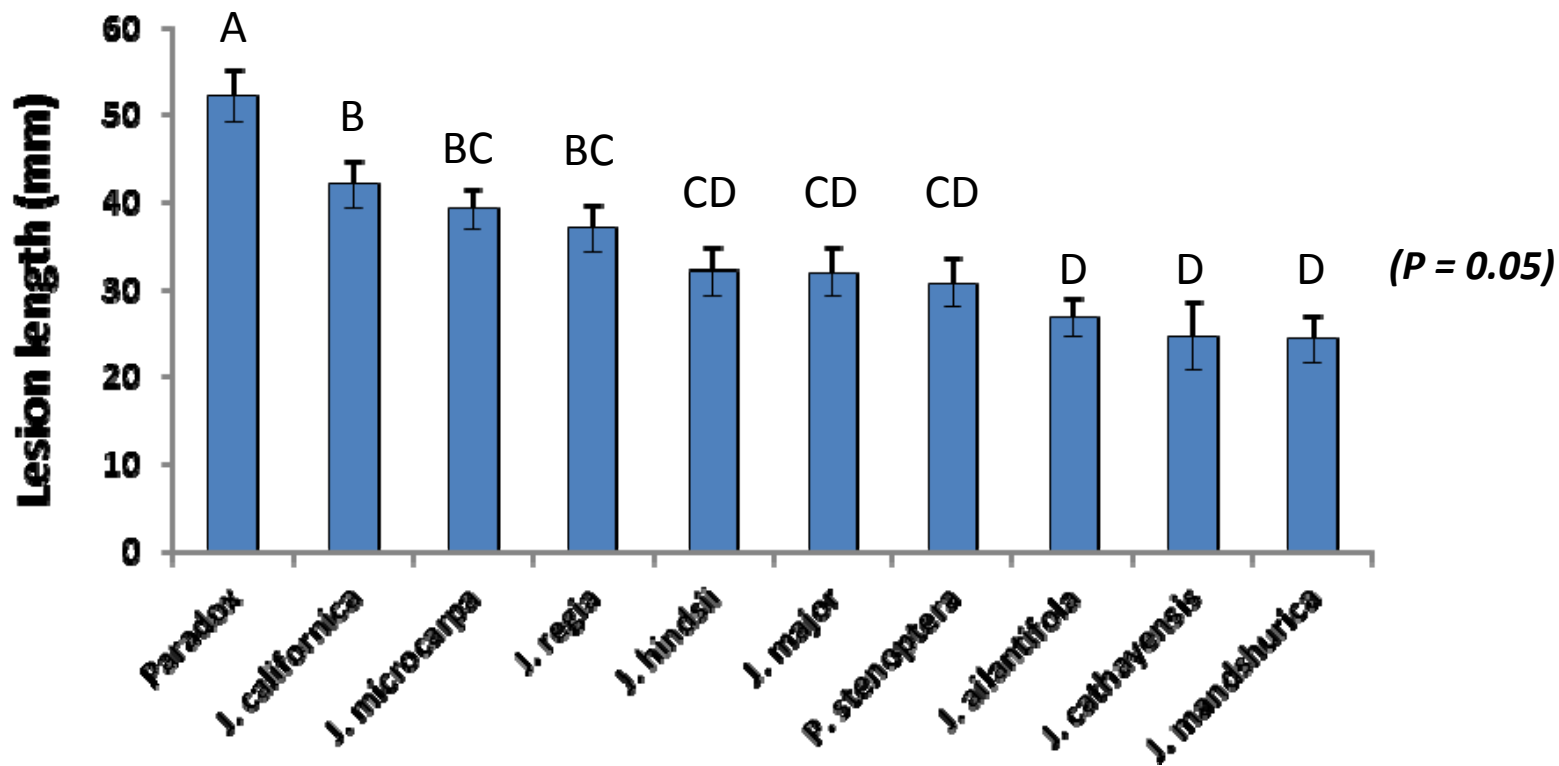
Juglans National Clonal Germplasm Collection USDA/ARS NCGR – Winters, CA

J. ailantifolia
J. australis
J. californica
J. cathayensis
J. hindsii
J. major
J. mandshurica

J. microcarpa
J. mollis
J. neotropica
J. nigra
J. olanchana
J. regia
J. sinensis
J. hindsii x *J. regia*
(*Paradox*)

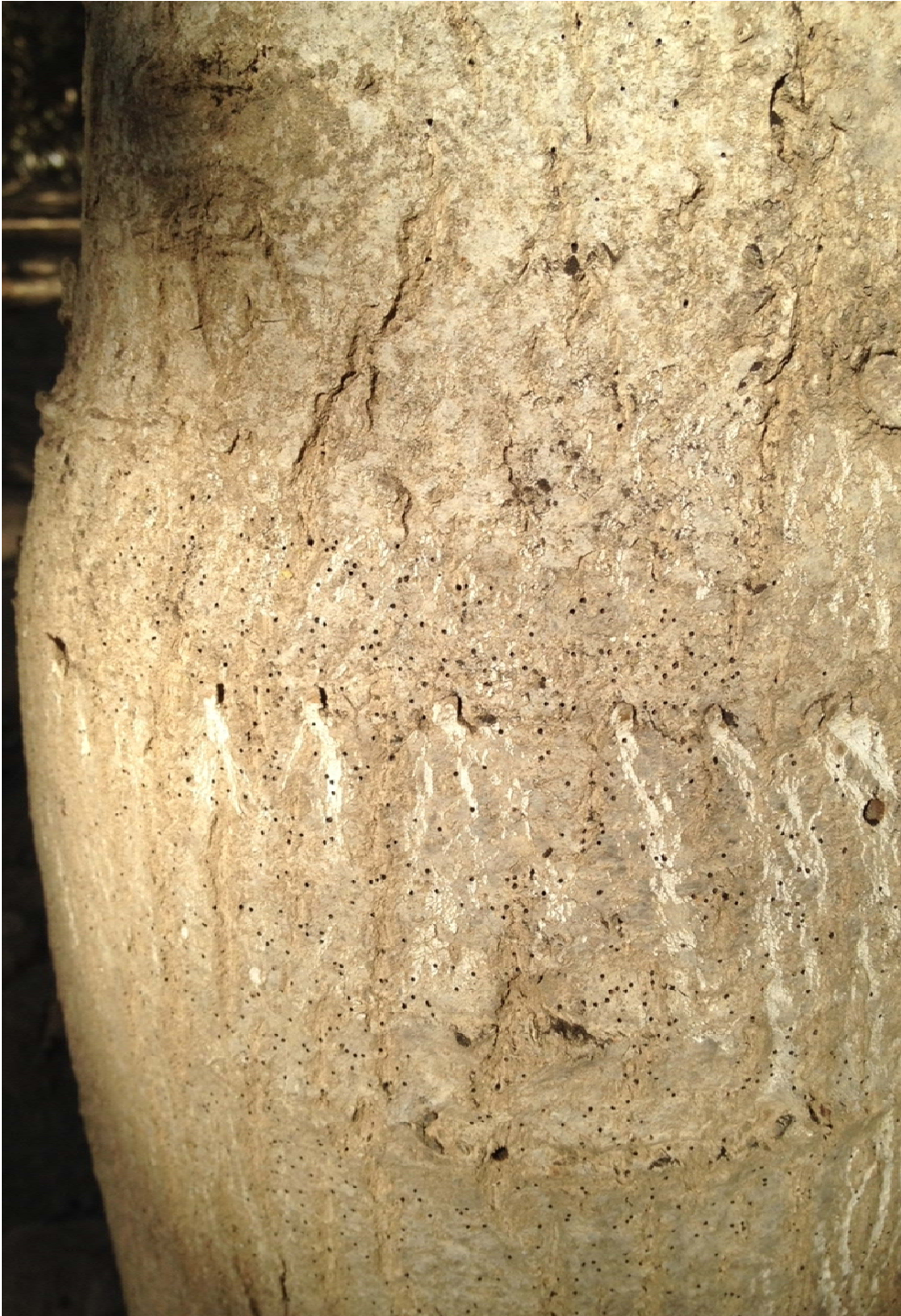
Most are developmental hosts for the WTB and can contract TCD

Juglans species show differences in susceptibility to *Geosmithia morbida*





TCD in Paradox rootstock, Tulare County
slide courtesy Elizabeth Fichtner

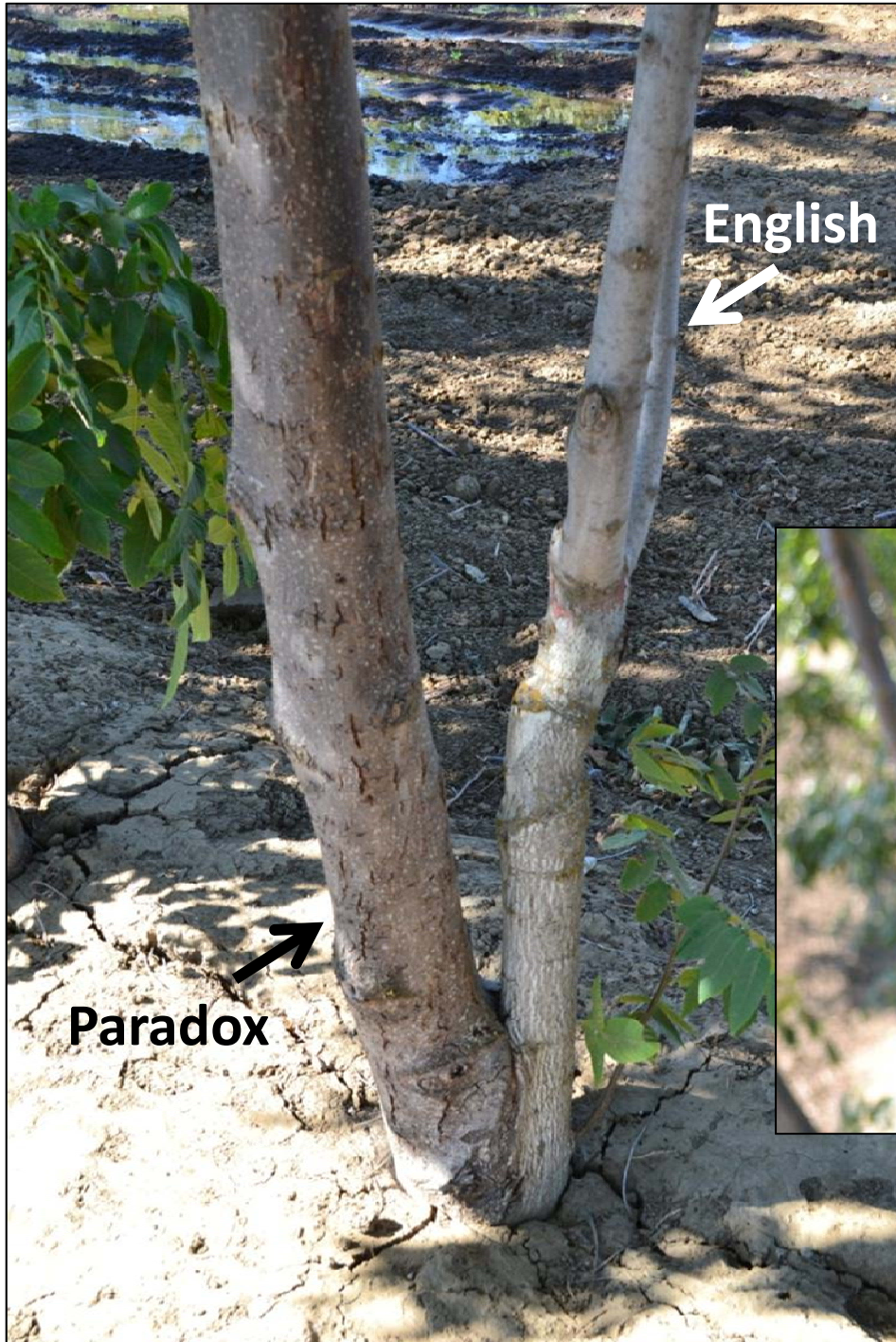


English scion

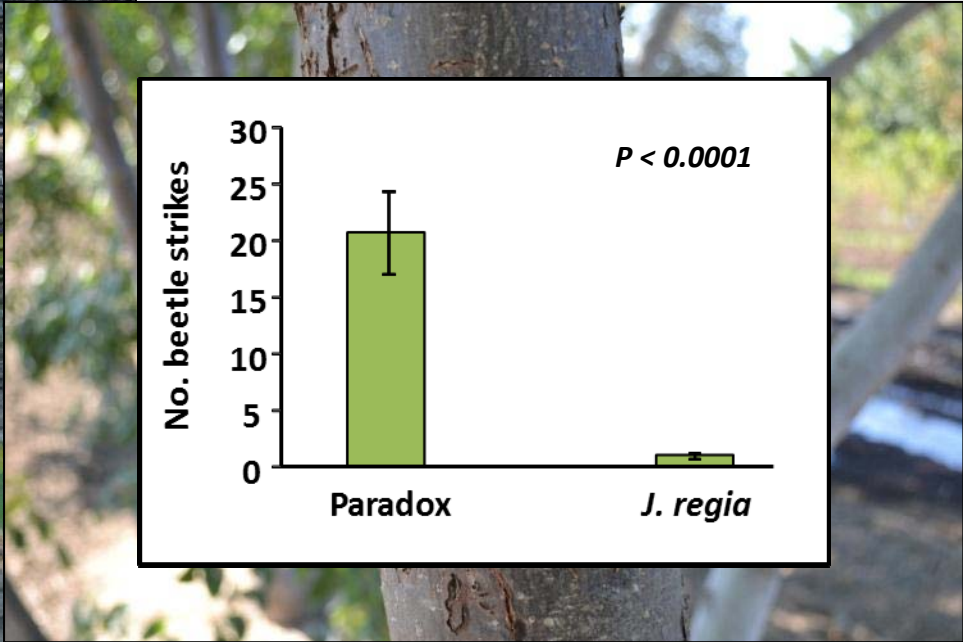
**WTB appears to select
Paradox over English**

Paradox rootstock

Tulare County, CA 2013



WTB appears to select Paradox over English



Armstrong Plant Pathology Research Station
UC Davis

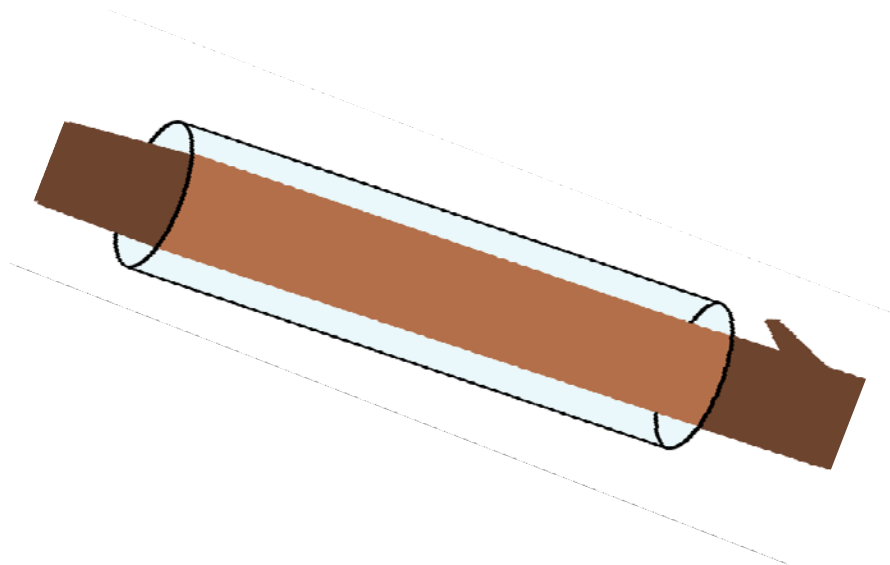
Unbaited live branch studies

NCGR – Winters, CA

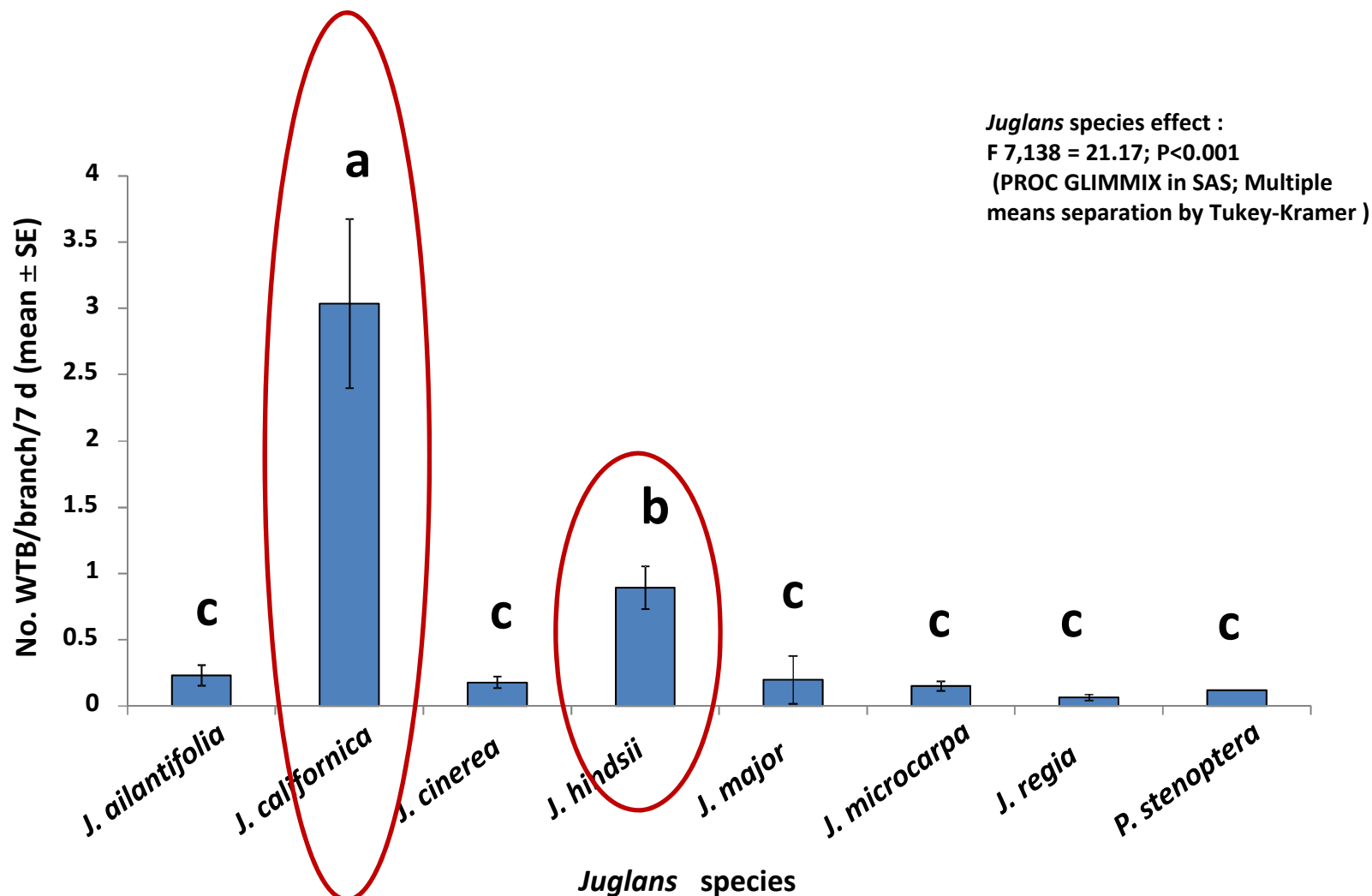
2012-2013

Juglans ailantifolia, *J. californica*, *J. cinerea*, *J. hindsii*, *J. major*, *J. microcarpa*, *J. regia*, and *Pterocarya stenoptera*

rees of each species, 1 uninfested branch/tree

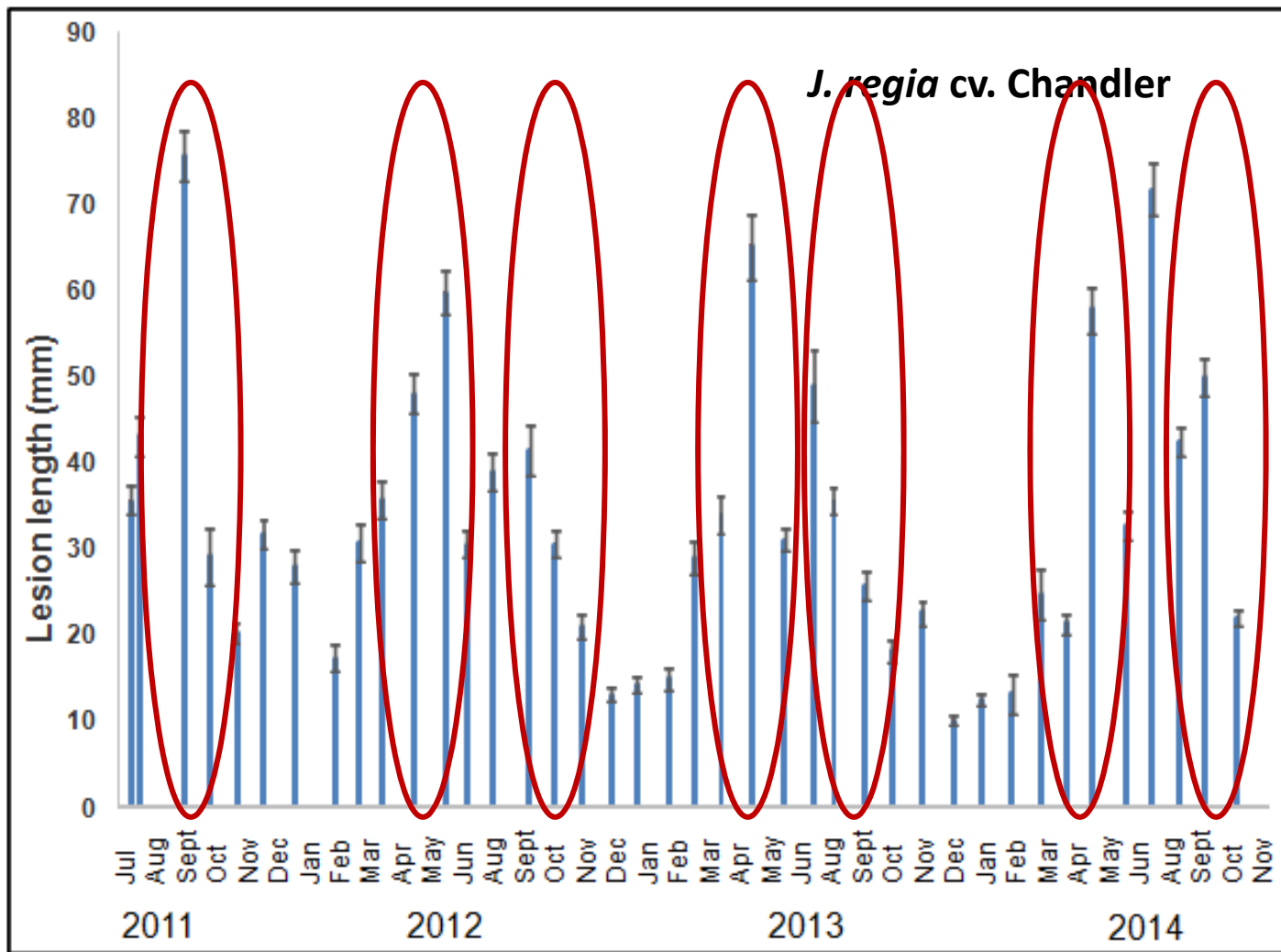


Landing rate of WTB on branches of six species of *Juglans* and one *Pterocarya* species (2013)

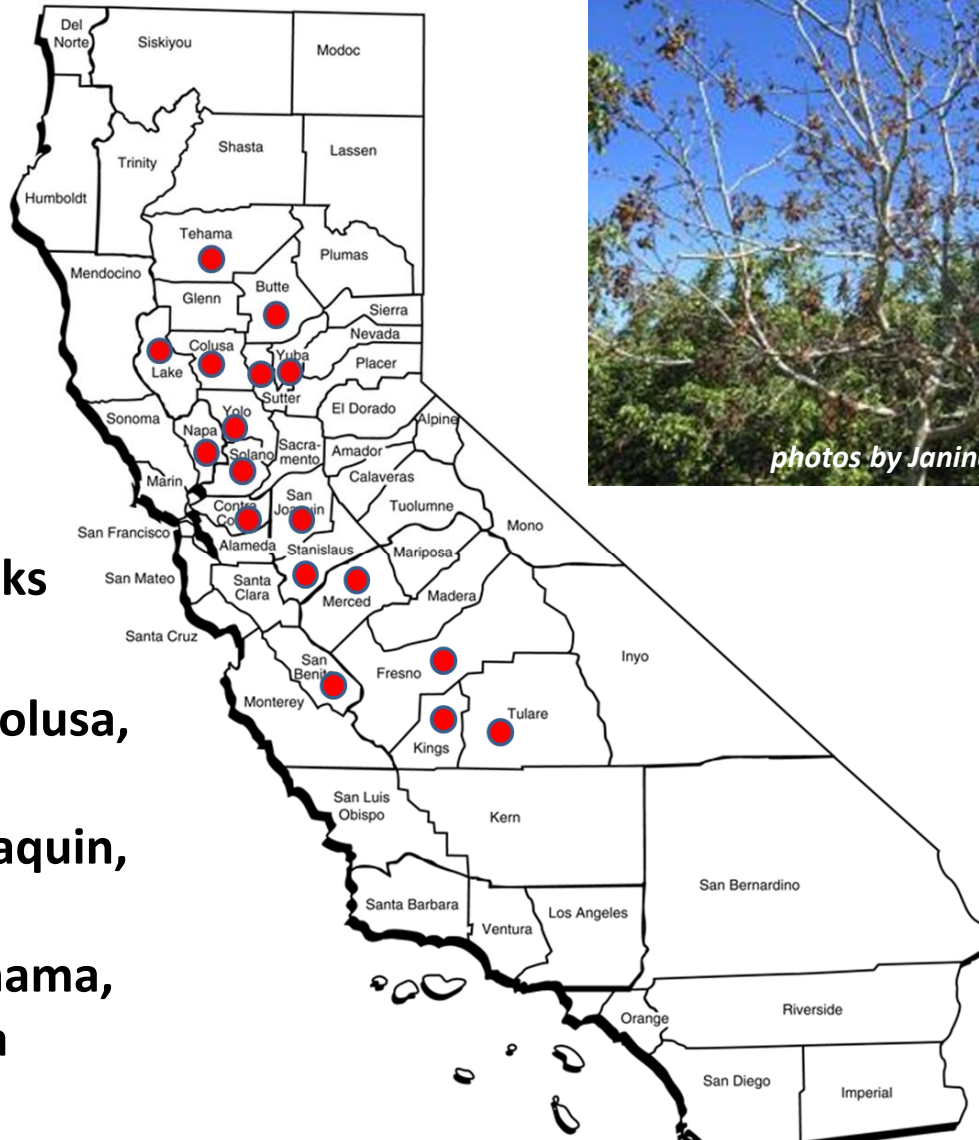


Seasonal variation in English walnut susceptibility to *G. morbida*

Key periods of beetle flights (May-July & Sept-Oct) and host susceptibility may overlap in spring and summer



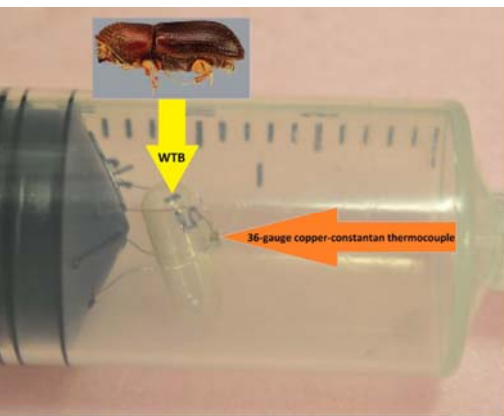
TCD Status in California English Walnuts



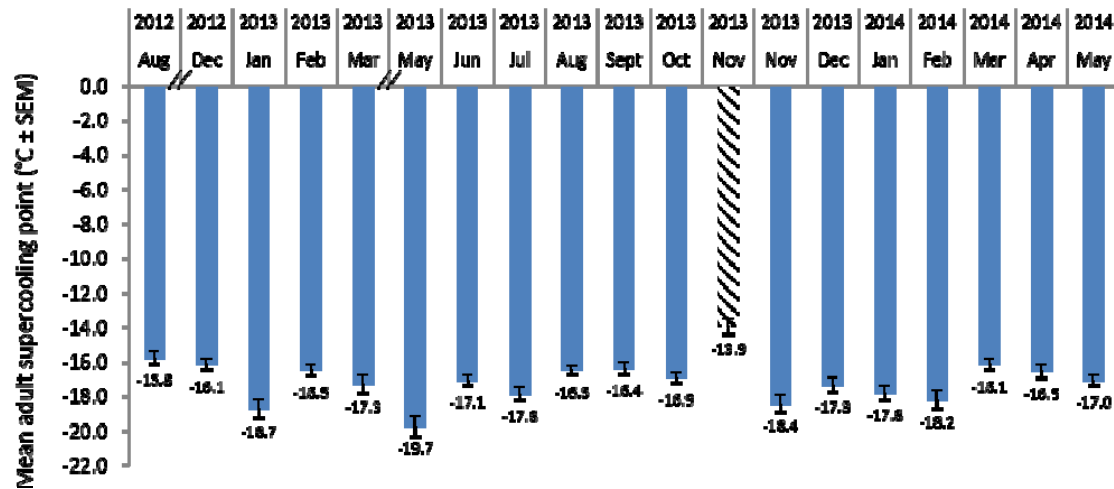
photos by Janine Hasey, UCCE

**Confirmed TCD in rootstocks
English scions in:
Butte, Contra Costa, Colusa,
Fresno, Kings, Lake,
Merced, Napa, San Joaquin,
San Benito, Solano,
Stanislaus, Sutter, Tehama,
Tulare, Yolo, and Yuba
counties**

WTB Cold Tolerance



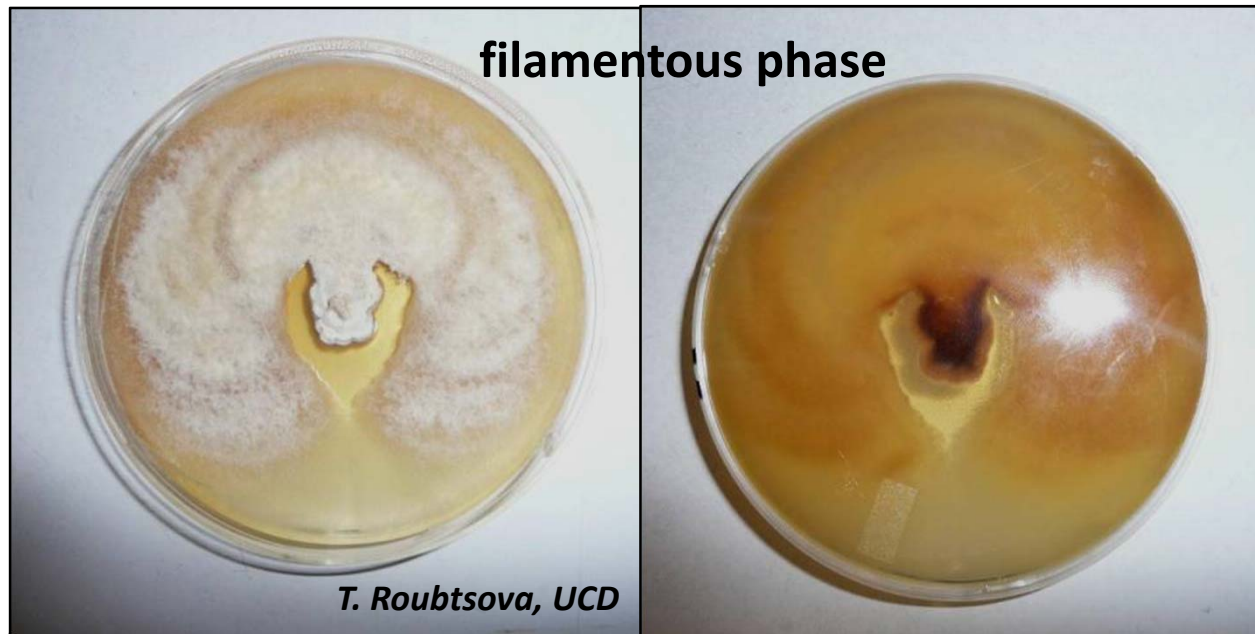
Contact thermocouple-thermometry



Changes in the mean supercooling point of WTB adults from Sutter Co., CA by month. Bars with crosshatching designate individuals from Orofino, ID (Andrea Hefty et al., unpublished data, Univ. of Minnesota, Ph.D. Thesis Data)

***Geosmithia morbida* – a new species 2011**

Kolarik M. et al. (2011) Mycologia 103:325





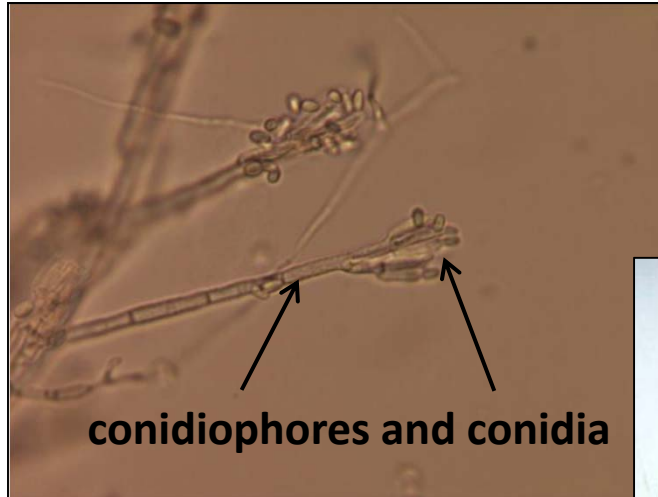
Removal of outer bark tissue reveals galleries and canker



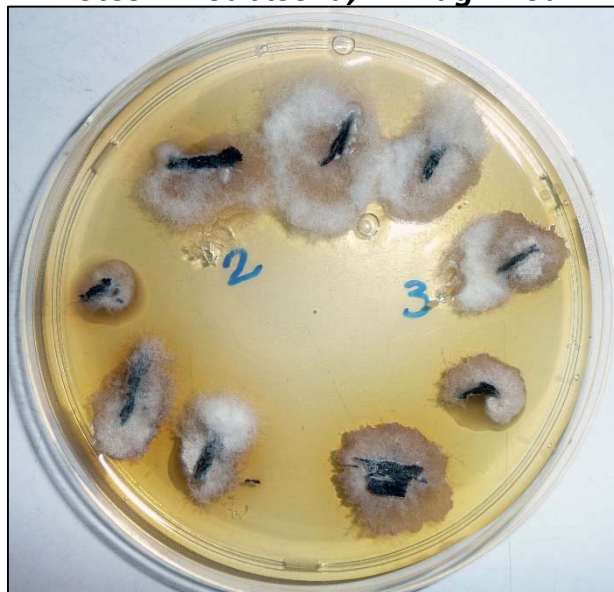
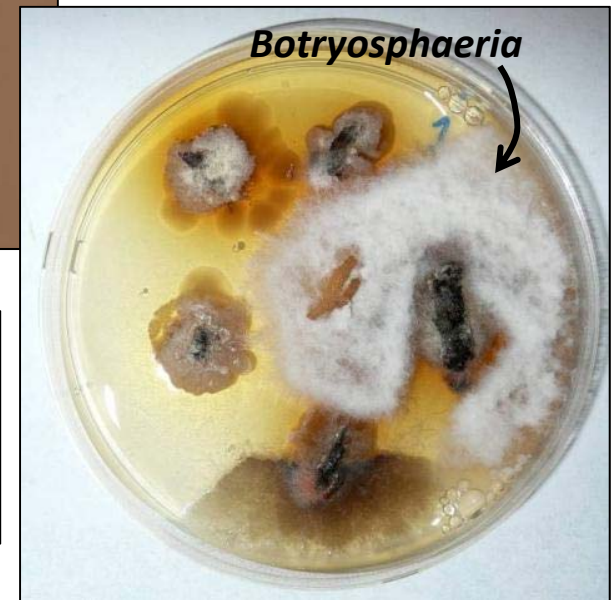
Geosmithia morbida – isolation on A-PDA



Photos T. Roubtsova, M. Yagmour



Sometimes find other canker fungi in disease samples also – can be tricky.



Several canker diseases impact CA walnuts

Challenge for field surveys and diagnostics



deep bark canker
(Brennaria rubifaciens)



lethal paradox disease

?

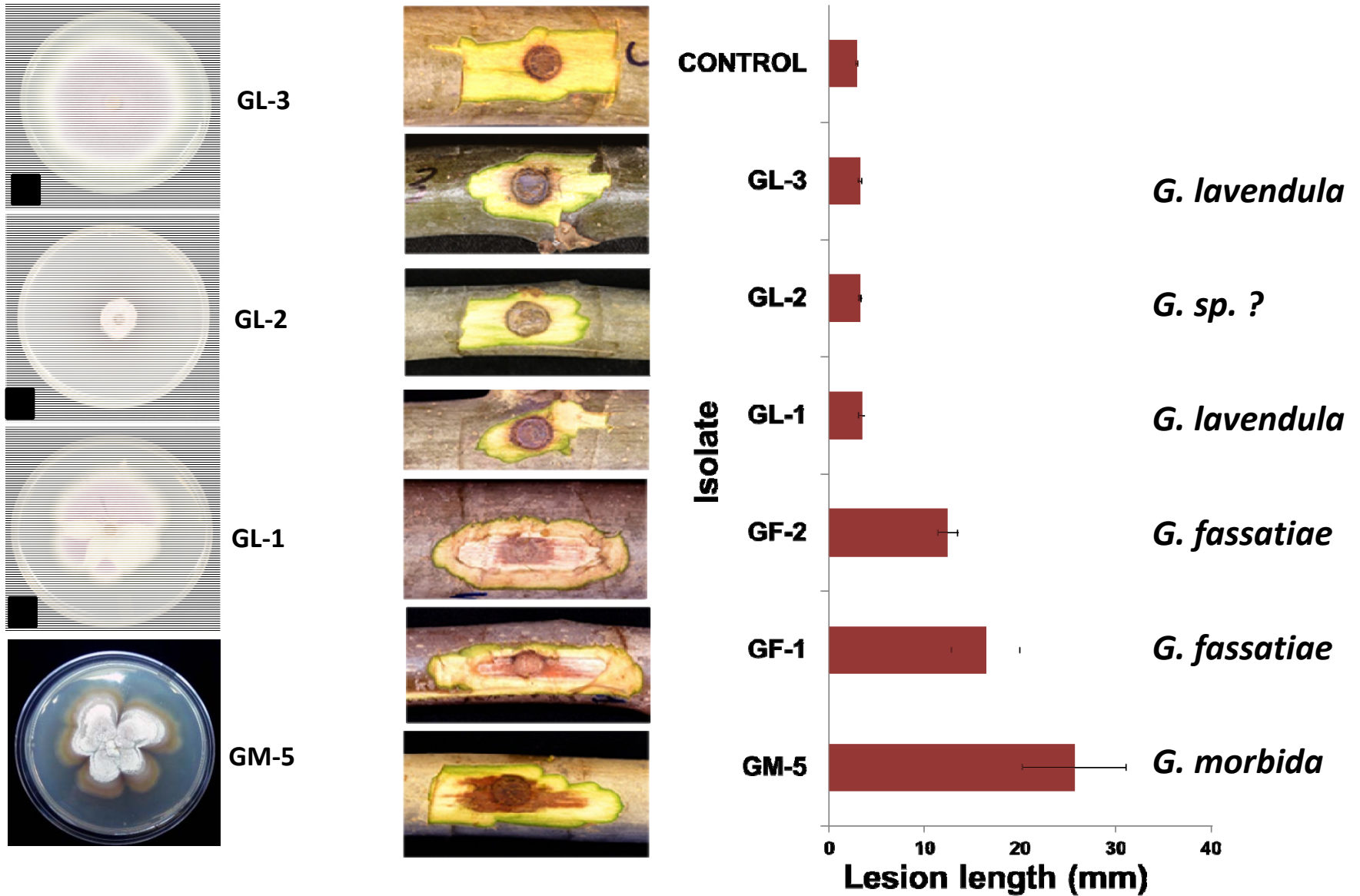


shallow bark canker
(Brennaria nigrifluens)

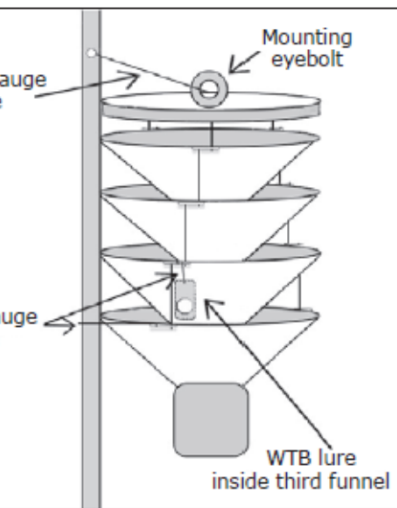


shallow bark canker and TCD
→ dying trees

Several *Geosmithia* species are associated with the WTB



Funnel Traps for Capturing Walnut Twig Beetle



J. A. King, UC Davis
 D. Schematic of a four-unit funnel trap showing the attachment between the trap and pole with heavy-gauge wire, the attachment and placement of the lure, and the attachment of the lowest funnel to the pole with light-gauge wire.

University of California
 Agriculture and Natural Resources
UC-IPM
 Research Integrated Pest Management Program

DETECTING AND IDENTIFYING WALNUT TWIG BEETLE: Monitoring Guidelines for the Invasive Vector of Thousand Cankers Disease of Walnut

STEVEN J. SEYBOLD, USDA Forest Service, Pacific Southwest Research Station, Davis, Calif.;
 PAUL L. DELGADO, Entomology, UC Davis; XINPING WU, HORTICULTURE, Entomology, UC Davis;
 MARY LOUISE FLINI, Entomology, UC Davis and UC Sustainable IPM Program

Walnut twig beetle (WTB), *Pityophthorus juglandis* (Oppler) is a small native phytophagous (plant-eating) insect recently associated with the fungus *Cronohium ashbyi* (Schubert et al. 2012). This fungus and WTB are the principal agents involved in Thousand Cankers Disease (TCD) (Schubert et al. 2012). WTB is the primary vector of *C. ashbyi* (Wu et al. 2012). This disease is fatal to walnut trees and is responsible for the gradual decline of numerous species of black walnuts in the western United States during the past decade (Kane et al. 2011; Flini et al. 2012; Tansil et al. 2012). The disease has spread widely to the western United States and has been detected in various states – including Tennessee (Kane 2012), Virginia (Kane 2012), and Pennsylvania (Kane 2012) – threatening the highly valuable native timber stands of eastern black walnuts, *Juglans nigra* (Millerton and Tansil 2013).

The beetle is now distributed discontinuously in the United States from eastern Oregon through to western Oregon and from southern Idaho to southern New Mexico to the West (Schubert et al. 2012). It was trapped in real time in Ohio in July 2012. Populations of WTB have been previously associated with the target tree type of stands of walnut that have found only where the beetle is present. Thus, capturing and identifying the key beetle is the key to early detection of the disease in new areas.

This publication provides detailed guidelines for using glass-mesh funnel traps to detect and monitor WTB. A trap user guide for field use and instruction sheets are also available at www.ipm.ucdavis.edu/PMG/Menu/ThousandCankers/Walnut/WalnutTWB.html. The purpose of this report is to define an accepted population of WTB or define a minimum population of WTB infestation has been recently discussed.

The trap and guidelines described here were developed in Southern California walnut orchard ecosystems with high population densities of WTB. Independently however, the trapping methodology has been field tested and demonstrated as a variety of native and introduced landscapes in California (Utah, Pennsylvania, Tennessee, Ohio, and Virginia) with low to intermediate population densities of WTB. The system uses a novel (and high-mass) trap design (Tansil) with the main purpose of aggregating pheromone (Schubert et al. 2012). The trap captures both sexes of the WTB while attracting few other insect species, including only low numbers of other native bark or cambium beetles (*Coleoptera: longhornidae* (Cleridae) 1 and 2, including wood-boring WTB larvae).

Figure 1. Adult male WTB lateral profile.

Figure 2. Four-unit funnel trap.



Walnut Twig Beetle

Pityophthorus juglandis

An ambrosia beetle,
Xyleborinus saxeseni

WTB

0.5 mm



♂



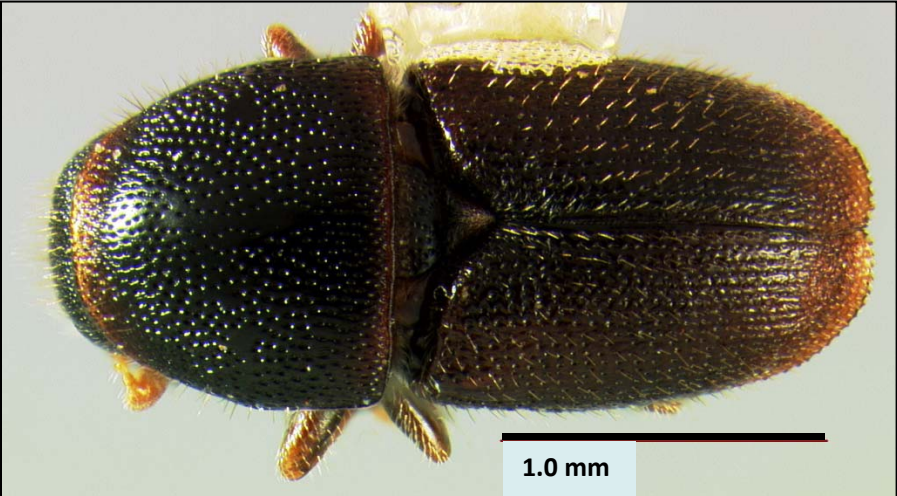
♀



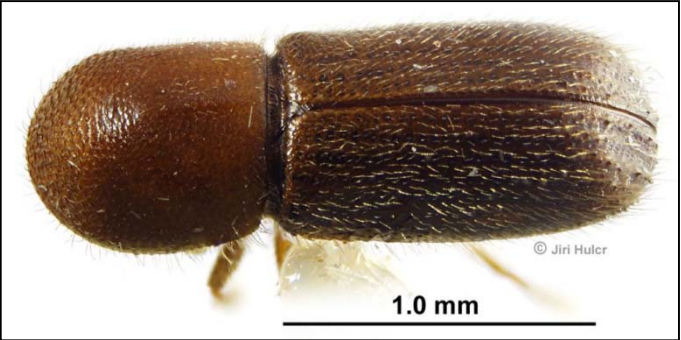
Other taxa in walnut orchards



Hypothenemus eruditus
bark beetle



Scolytus rugulosus
shot-hole borer

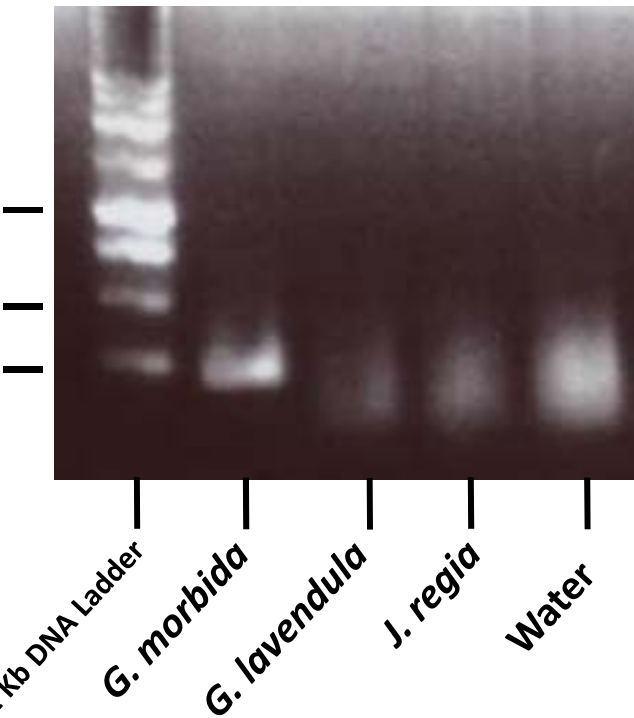


Cyclorhinidion hodoanum

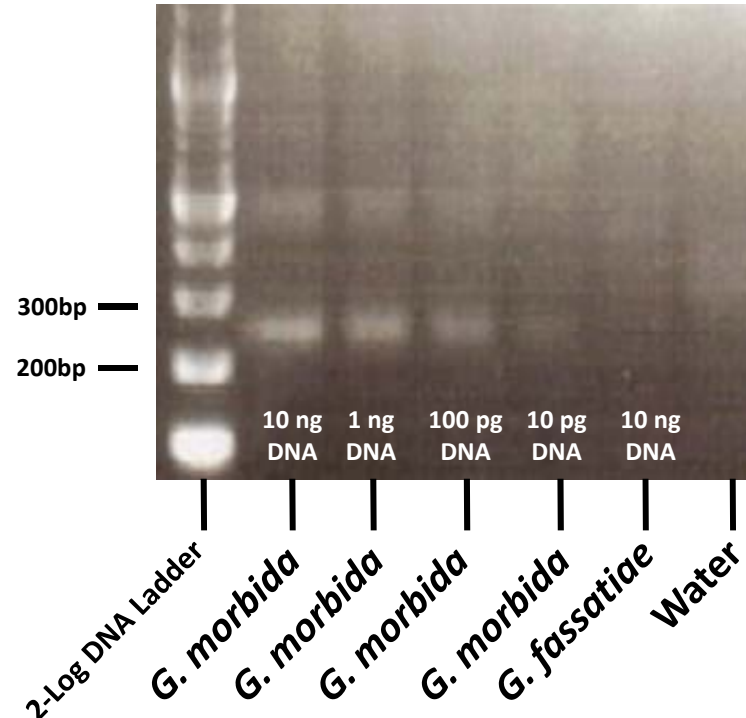
Recombinase Polymerase Amplification (RPA) Assay

Specific detection of *G. morbida* with 10 picogram DNA sensitivity!

Gel 1



Gel 2



Agarose gel electrophoresis of amplicon using *Geosmithia morbida*-specific *EF-1 α* primers

EF-1 α primer specificity for *Geosmithia morbida* over *G. lavendula*, *G. fassatiae*, and *J. regia*

Mil Ulceras Enfermedad de Nogales

Una Enfermedad nueva en California



Por Ricardo Hoenisch, Departamento de Fitopatología, UC Davis

Mil Ulceras de Nogales el género botánico *Juglans*



TCD management and future directions

Fungicides? Insecticides? Unlikely

Sanitation to mitigate spread – removal of dead and severely declining trees; grind or burn immediately.

<http://www.ipm.ucdavis.edu/EXOTIC/thousandcankers.html>

Do not transport/ship infested walnut material, especially where this is prohibited by state quarantines.

Need to keep beetles from landing on trees:

- ✓ host selection behavior and chemistry of attraction/repulsion
- ✓ host and pathogen VOC's as coattractants with pheromone

Improved diagnostic tools

Acknowledgements

Co-PI: Dr. Steven Seybold, USDA Forest Service and Dept. of Entomology and Nematology, UC Davis

UC Davis and UCCE

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Richard Hoenisch	}	Entomology
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Mary Lou Flint		
Yigen Chen	}	Plant Sciences
Paul Dallara		
Chuck Leslie	}	UCCE
Elizabeth Fichtner		
Marine Hasey		

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USDA-ARS

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