

# DEVELOPING SEMIOCHEMICALS FOR BARK BEETLES IN NEW SYSTEMS

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L.A. Mortenson, C.J. Fettig, D.F. Gomez,  
K. Roy, J.P. Audley, B.M. Bulaon,  
A. Mafra-Neto, A.S. Munson, & E.E. Davis



## Outline

1. *Bark beetle communication*
2. *Development of SPLAT® Verb*
3. *Recent applications of SPLAT® Verb into new systems including whitebark pine, limber pine, and rapid 'ōhi'a death (ROD)*
4. *Development of SPLAT® Verb+ and recent applications for southwestern pine beetle in West Texas*
5. *Q & A*



Top: Trees killed by mountain pine beetle.  
Bottom: Trees killed by rapid 'ōhi'a death (ROD).

## *Chemical ecology\* of the bark beetle-host system*



Western pine beetle galleries in the inner bark, California.

- *Pioneers use a combination of random landings, visual orientations, and **olfactory and gustatory cues**.*
- *Most tree-killing species have highly-evolved chemical communication systems. E.g., in western pine beetle, females release **exo-brevicomin**, which in combination with the host monoterpene **myrcene** is attractive to conspecifics. **Frontalin**, produced by males, enhances attraction. **Verbenone** is the primary anti-aggregation pheromone.*

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\*Chemical ecology seeks to understand the origin, function, and significance of natural chemicals that mediate interactions within, between, and among species.



***Semiochemical:*** a chemical emitted by one organism that affects the behavior of another organism, within or among species.

***Pheromone:*** a semiochemical that mediates intraspecific interactions.

***Anti-aggregation pheromones:*** pheromones released by insects to disperse their populations and reduce competition.

*Can we convince bark beetles that a suitable, susceptible host is not by masking its apparency?*

# RD&A sequence (years to decades)

## 1. Identify semiochemicals

- a) Relevant literature (cheap, efficient, limited)
- b) Sample the volatile profile of beetle extracts, frass, infested host materials, hosts, nonhosts, and the forest environment
- c) Gas chromatographic-electroantennographic detection analyses (GC-EAD, recordings of insect antennae)

## 2. Screen repellents – baited trap assays

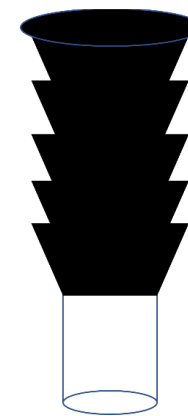
- a) Repeated many times (different locations, populations)
- b) >85% trap catch reduction
- c) Optimize doses, release rates, and combos

## 3. Efficacy of promising compound(s) at individual tree scale

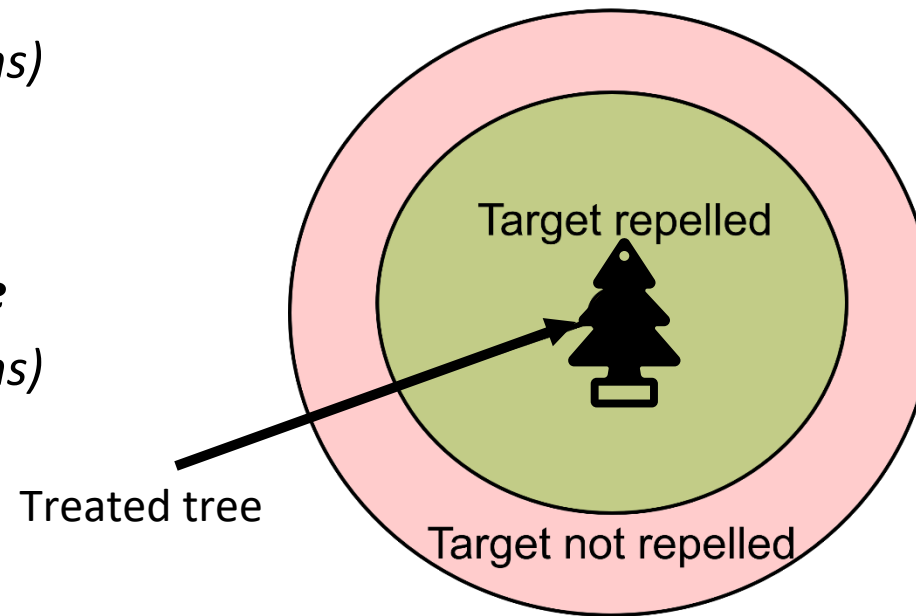
- a) Repeated many times (different locations, populations)
- b) Zone of inhibition

## 4. Efficacy of promising compound(s) at stand scale

- a) Optimize doses, release rates, and spacing



Zone of Inhibition



## Verbenone

- *Principle antiaggregant of several bark beetles including mountain and western pine beetles.*
- *Inhibits additional beetles from infesting the target tree, presumably limiting the number of beetles to a density that increases the likelihood of brood survival.*
- *Repels ambrosia beetles.*



A 5-g pouch was first registered by the U.S. Environmental Protection Agency (EPA) in 1999 for southern pine beetle.



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# SPLAT<sup>®</sup>

## Verb

Bark Beetle Repellent

*SPLAT is a matrix for the sustained passive release of insect pheromones. SPLAT Verb provides control of bark beetles by use of the anti-aggregation pheromone, repelling beetles from pine trees.*

 For Organic Production (optional marketing logo and claim)



**ACTIVE INGREDIENT:**

Bark Beetle Anti-Aggregation Pheromone  
Verbenone,  
4,6,6-Trimethylbicyclo(3.1.1) hept-3-en-2-one: 10.00%

**OTHER INGREDIENTS:**

90.00%

**TOTAL:**

100.00%

Net Contents: \_\_\_\_\_ Lbs \_\_\_\_\_ Kg

Batch Number: \_\_\_\_\_

EPA Reg. No. 80286-20  
EPA Est. No. 80286-CA-004

**KEEP OUT OF REACH OF CHILDREN  
CAUTION**

**FIRST AID STATEMENT**

**IF IN EYES:**

- Hold eye open and rinse slowly and gently with water for 15-20 minutes.
- Remove contact lenses, if present, after the first 5 minutes, then continue rinsing eye.
- Call poison control center or doctor for treatment advice

**IF ON SKIN OR CLOTHING:**

- Wash skin immediately with soap and water, then rinse with plenty of water for 15-20 minutes
- Take off contaminated clothing.

**IF SWALLOWED:**

- Call poison control center or doctor immediately for treatment advice.
- Do not induce vomiting unless told to do so by poison control center or doctor.
- Do not give anything by mouth to an unconscious person

**HOTLINE NUMBER**

- Have the product container or label with you when calling a poison control center or doctor or going for treatment
- For emergency information on (product, use, etc.), call the National Pesticide Information center at 1-800-858-7378, 6:30AM to 4:30PM Pacific Time (PT), seven days a week. During other time, call the poison control center 1-800-222-1222

General Classification: It is a violation of Federal Law to use this product in a manner inconsistent with its labeling. Do not apply this product in a way that will directly contact workers or other persons. Only protected handlers may be in the area during application. For any requirement specific to your State and Tribe, consult the State/Tribal agency responsible for pesticide regulation.



*Only verbenone product registered in CA  
is SPLAT<sup>®</sup> Verb, biodegrades in 1+ yr*



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# Mountain pine beetle (MPB) *(Dendroctonus ponderosae)*

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*SPLAT*<sup>®</sup> Verb – lodgepole pine, WY  
first publication = 2015

*SPLAT*<sup>®</sup> Verb – sugar pine, CA  
first publication = 2016







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*SPLAT<sup>®</sup> Verb – whitebark pine, CA, MT & OR  
first publication = 2021*

*Operationally used (Verb Pouch also, other  
states) in ski areas, some NFs & NPs; to  
protect blister rust resistant (+) trees*



Experimental applications of SPLAT<sup>®</sup> Verb to whitebark pine (**status = Threatened & Endangered, USFWS**), California.



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*SPLAT<sup>®</sup> Verb – mountain pine beetle  
and limber pine, Death Valley NP, CA*

***NPS, FHP, USFS PSWRS, RMRS***

*Limber pine adjacent and/or intermixed  
with **bristlecone pine**, concern over MPB  
colonizing limber pine being able to  
colonize (kill) nearby bristlecone pine*



*Thus far “SPLATed” limber  
pine have not experienced  
any observed mortality  
although MPB pressure  
(population) has  
seemingly crashed.  
Additional monitoring is  
needed.*



## Rapid 'Ōhi'a Death

**'Ōhi'a lehua** (*Metrosideros polymorpha*)  
Most ecologically and culturally important tree  
in Hawai'i

Caused by two species of ***Ceratocystis*** fungi  
and spread (wound required) in part via  
frass from four species of invasive  
*Xyleborini* ambrosia beetles





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## *SPLAT<sup>®</sup> Verb on healthy and ROD-affected 'Ōhi'a*

*No known pheromone communication in ambrosia beetles; use kairomones (interspecific semiochemical that benefits receiver, not emitter).*

*Tested low label rate (72 g) and high label rate (108 g) of SPLAT<sup>®</sup> Verb.*

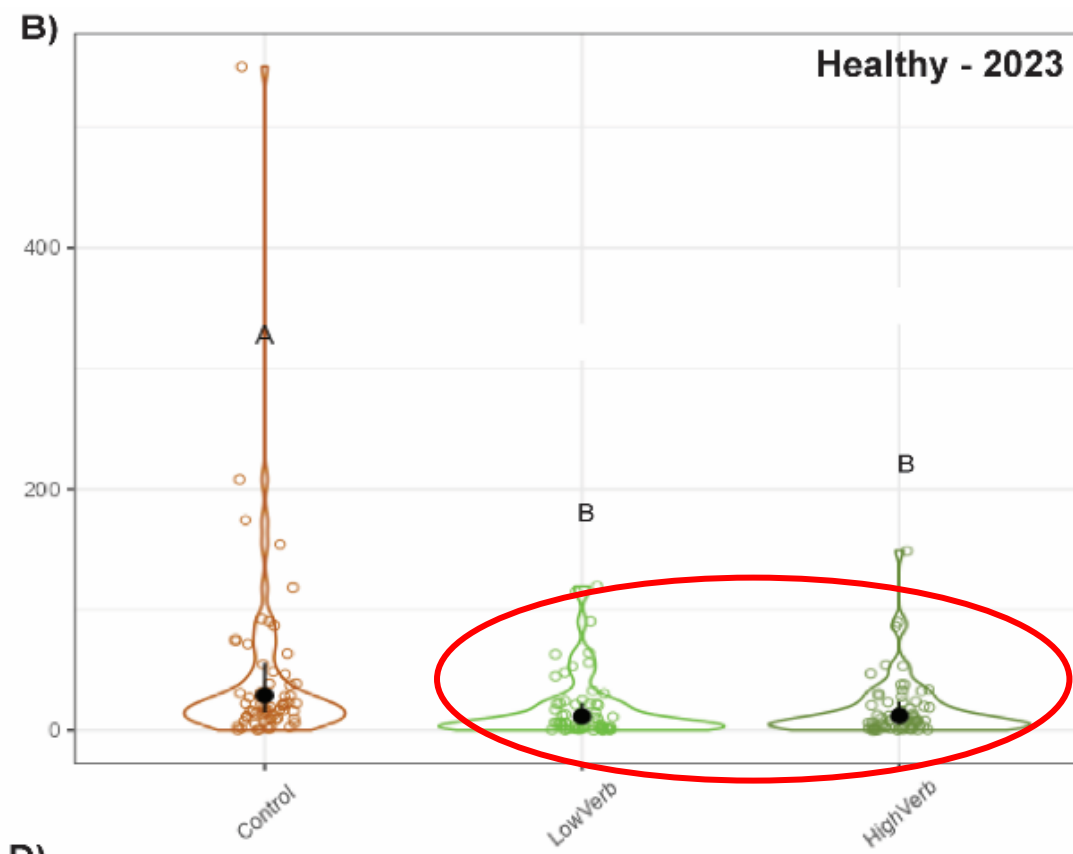
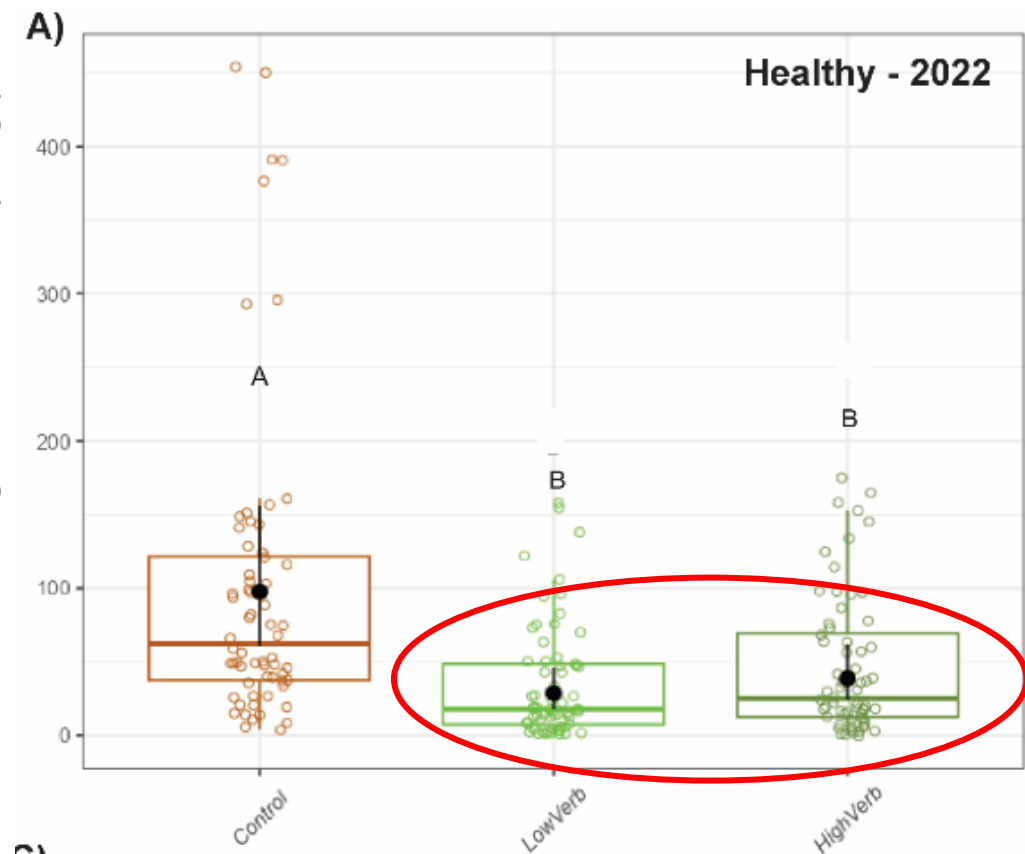
*Also tested SPLAT<sup>®</sup> Verb in conjunction with SPLAT<sup>®</sup> "Beetle Guard", verb + methyl salicylate (trapping assay); may signal that the tree is decomposing/dead/uninhabitable.*





# SPLAT<sup>®</sup> Verb on healthy 'Ōhi'a trees

No. of beetles landing on trees/sampling period

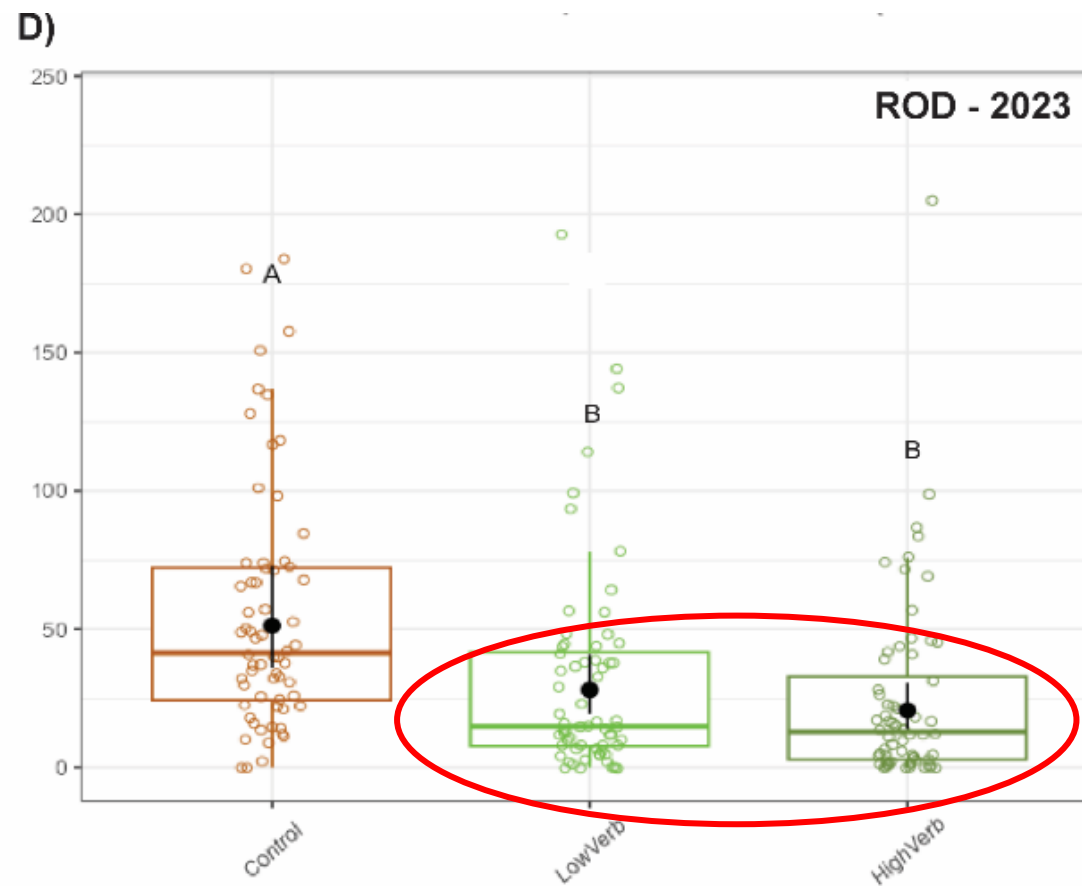
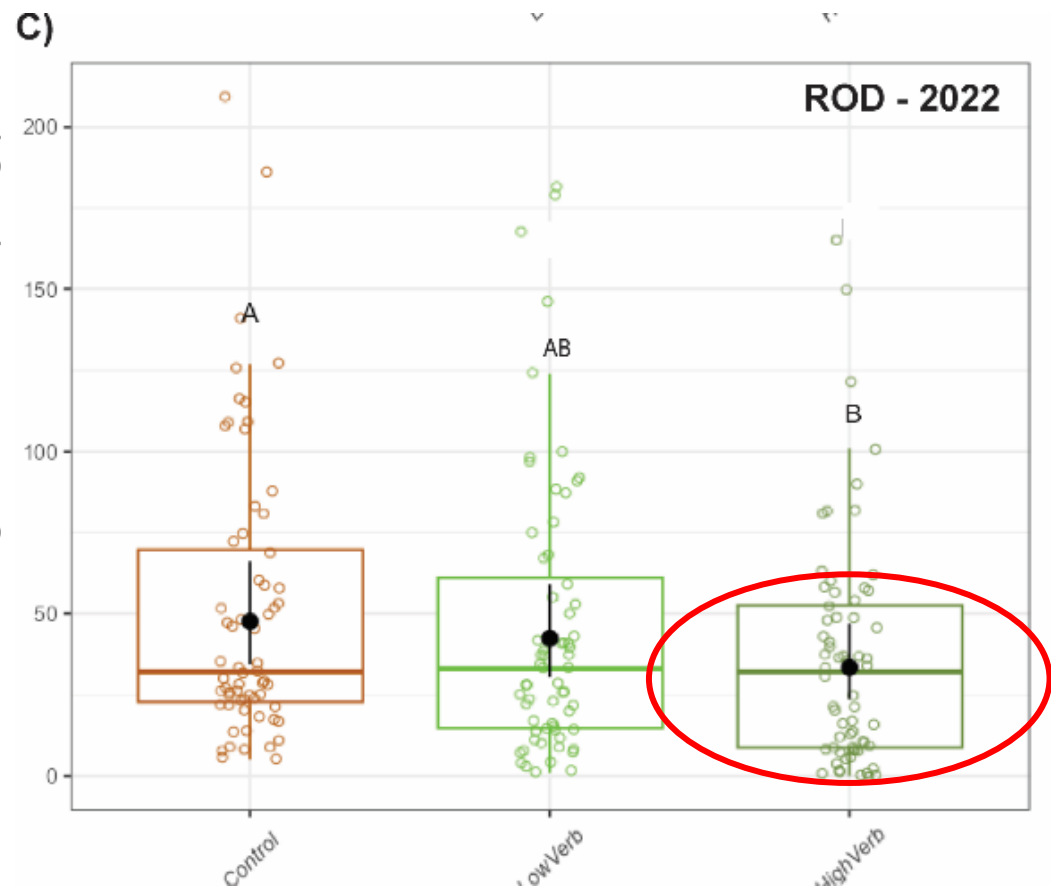


*Both dosages of SPLAT<sup>®</sup> Verb effectively reduced ROD-associated ambrosia beetle landings on **healthy trees**.*



# SPLAT<sup>®</sup> Verb ROD-affected 'Ōhi'a trees

No. of beetles landing on trees/sampling period



The high dosage was necessary to reduce ROD-associated beetle landings on **affected trees** in 2022.  
Both dosages reduced ROD-associated beetle landings on **affected trees** in 2023.



# Following years of work on western pine beetle “Verbenone Plus”

- Acetophenone
- (E)-2-hexen-1-ol + (Z)-2-hexen-1-ol
- (-)-verbenone

**Not yet commercially available, unfortunately!**

Also, still experimenting with potential additional additives (C. Homicz, PhD work)



THE DAVIS MOUNTAINS (MOSTLY A NATURE  
CONSERVANCY PRESERVE) HOLD THE FARTHEST SE  
POPULATION OF PONDEROSA PINE IN THE U.S. & THE  
LARGEST POPULATION IN TEXAS



HALF OF THE PONDEROSA PINE POPULATION LOST FROM  
WILDFIRES, BARK BEETLES, AND DROUGHT

# Southwestern pine beetle (*Dendroctonus barberi*)

Journal of Chemical Ecology (2021) 47:10–27  
<https://doi.org/10.1007/s10886-020-01233-y>



## Evidence for Semiochemical Divergence Between Sibling Bark Beetle Species: *Dendroctonus brevicomis* and *Dendroctonus barberi*

Brian T. Sullivan<sup>1</sup> · Amanda M. Grady<sup>2</sup> · Richard W. Hofstetter<sup>3</sup> · Deepa S. Pureswaran<sup>4</sup> · Cavell Brownie<sup>5</sup> · Daniel Cluck<sup>6</sup> · Tom W. Coleman<sup>7</sup> · Andrew Graves<sup>8</sup> · Elizabeth Willhite<sup>9</sup> · Lia Spiegel<sup>10</sup> · Dwight Scarbrough<sup>11</sup> · Andrew Orlemann<sup>12</sup> · Gerardo Zúñiga<sup>13</sup>

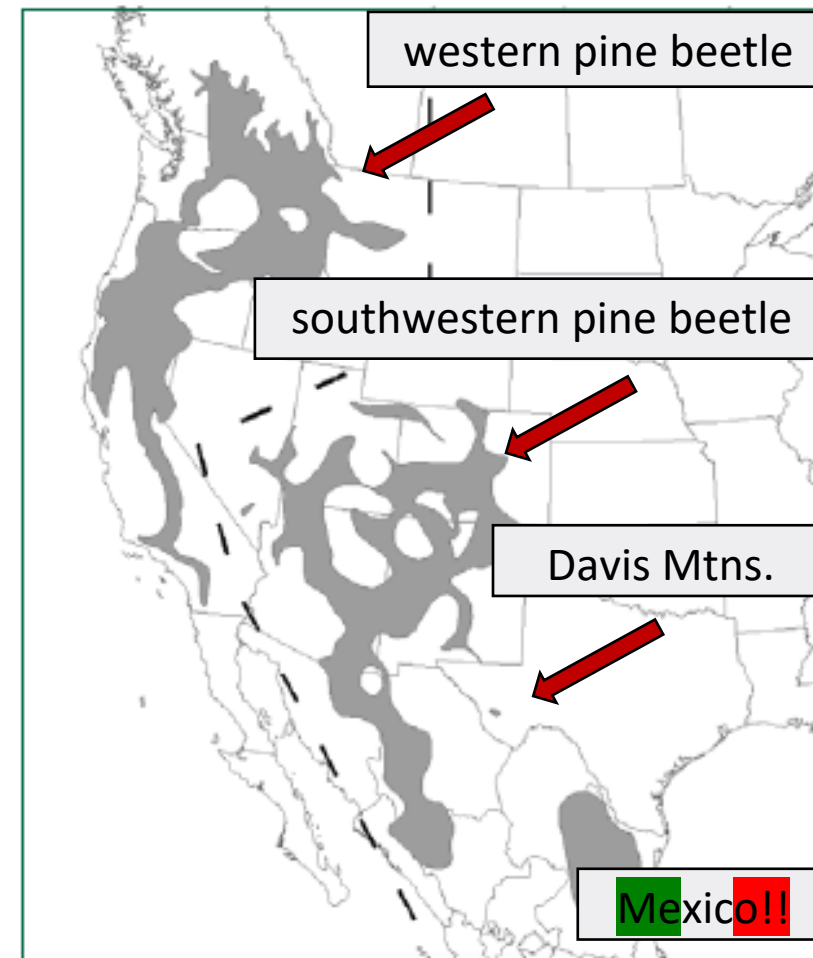
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Article

## Cryptic Species Discrimination in Western Pine Beetle, *Dendroctonus brevicomis* LeConte (Curculionidae: Scolytinae), Based on Morphological Characters and Geometric Morphometrics

Osiris Valerio-Mendoza<sup>1</sup>, Jazmín García-Román<sup>1</sup>, Moises Becerril<sup>1</sup>, Francisco Armendáriz-Toledano<sup>1,†</sup>, Gerardo Cuéllar-Rodríguez<sup>2</sup>, José F. Negrón<sup>3</sup>, Brian T. Sullivan<sup>4</sup> and Gerardo Zúñiga<sup>1,\*</sup>





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## *SPLAT<sup>®</sup> Verb+ & SWPB*

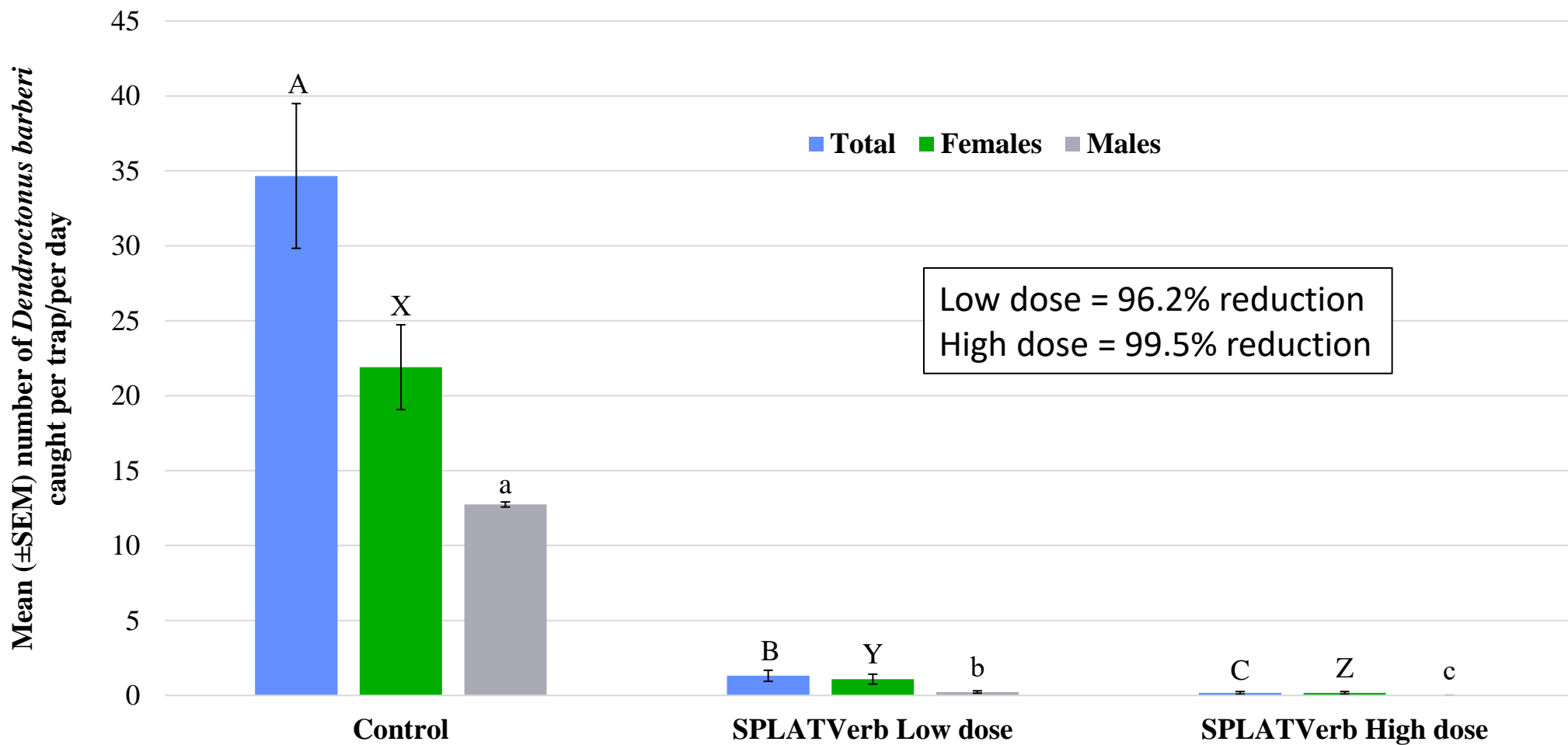
*Trap assays & tree protection  
(application)*

*17% mortality on untreated  
(control) plots. Unbaited design.*

*No ponderosa pine mortality  
on treated plots.*



**Fall assay** [spring assay = minimal pressure (mean for control = 2.3 beetles per day)]





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## ***Questions?***

*[christopher.fettig@usda.gov](mailto:christopher.fettig@usda.gov)*

*[leif.mortenson@usda.gov](mailto:leif.mortenson@usda.gov)*

*(Also: ongoing SPLAT<sup>®</sup> MCH research for spruce beetle management)*

