#### The History, Biology and Future of Biological Control

Dan Bean, Colorado Department of Agriculture, Palisade Insectary

Palisade, CO

Biocontrol in Action Workshop Phoenix, AZ April 30, 2025



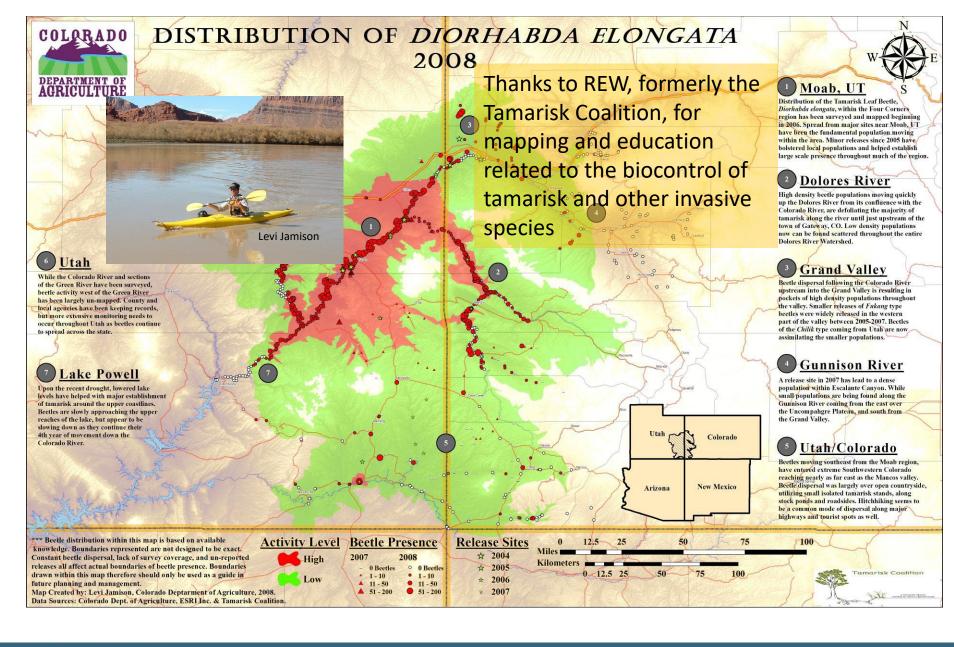
2007 (Nina Louden)

2010 (Sonya Daly)

Tamarisk Monitoring site on the Dolores River, CO



Diorhabda carinulata larvae



## What is biological control?

1. Biological control (biocontrol) is the use of natural enemies to control pests, including insect pests and noxious weeds.



Field bindweed stunted by the mite *Aceria malherbae* 



Macrocentrus ancylivorus stings host

## Types of biocontrol

- 1. Augmentative
- 2. Conservation
- 3. Classical

Establishing host specific natural enemies from the native range of the weed or insect pest into the introduced range.



- Tamarisk originated in Eurasia
- About 300 natural enemies were discovered
- Beetles in the genus *Diorhabda* were selected as biocontrol agents





## What is classical biological control?

- 1. Biological control (biocontrol) is the use of natural enemies, including insects, mites and pathogens, to control pests, including insect pests and noxious weeds.
- Classical biocontrol is a sustainable ecologically based pest control method. The goal is suppression of the weed or insect pest, not eradication. Often the desired results take years to achieve.



Field bindweed stunted by the mite Aceria malherbae

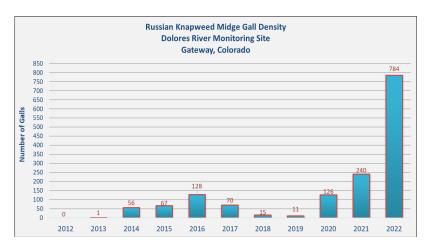




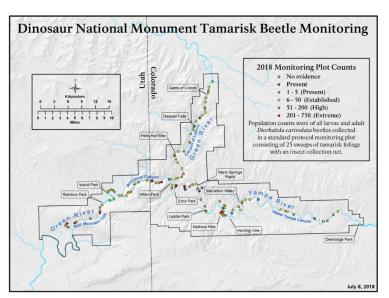
Leafy spurge herbivory by the red-headed borer, *Oberea*. Damage from feeding larvae (left) and feeding adult (right)

# Results of classical biocontrol require years to achieve

Invasive plant biocontrol occurs in <u>ecological time</u>, requiring years and sometimes decades to reach equilibrium.



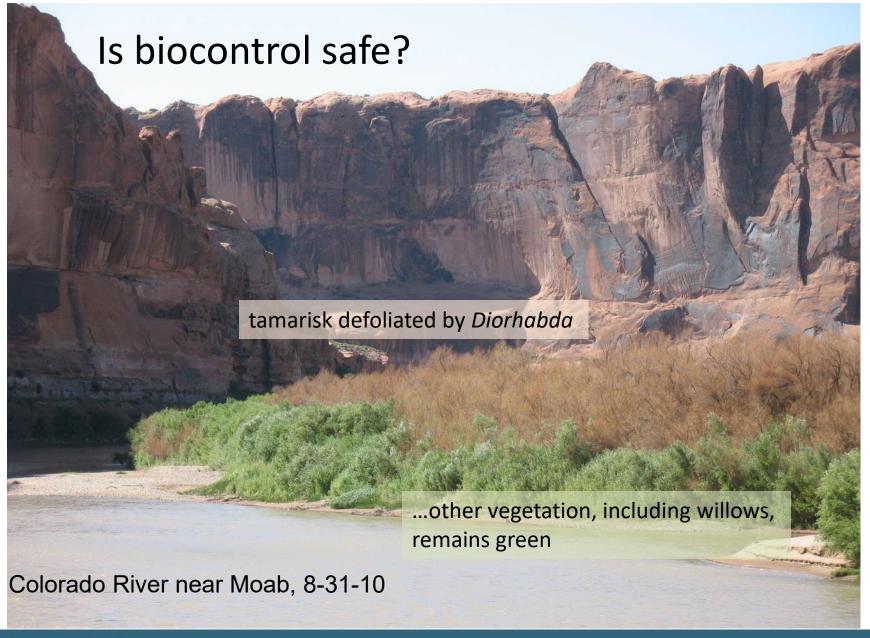
Russian knapweed, Dolores River, since 2012



Dinosaur Natl Monument invasive species project since 2006



Poudre Invasive Species Partnership, Larimer County, since 2013





# Safety considerations centered on host specificity

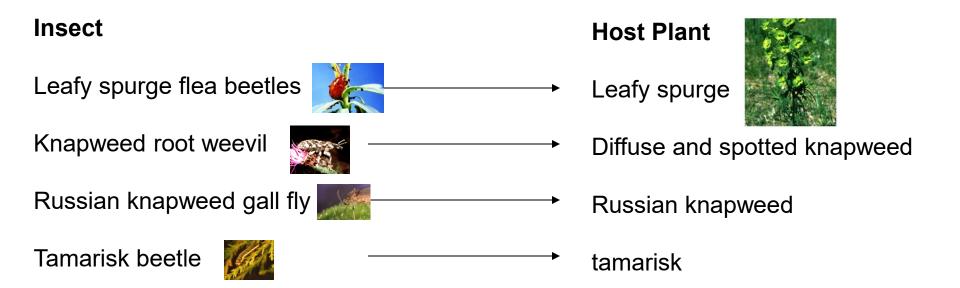
- 1. Do agents feed on agriculturally important plants?
- 2. Do agents feed on native plants? (after 1970)







# Generalists vs. Specialists: What makes them specialists?



- 1. Long range volatile attractants (smells)
- 2. Feeding stimulants
- 3. Ability to detoxify plant compounds

### Weed Biological Control is Safe

- Only specialists are used
- It takes at least ten years for an agent to be approved
- There has never been a case in modern weed biocontrol where a biocontrol agent switched host plants



**Host Range Testing** 

# Why do some non-native plants become invasive weeds?

- 1. Absence of natural enemies.
- 2. Unusual genetics, unique strains and hybrids
- 3. Better competitors in disturbed habitats
- 4. Some are drivers of ecosystem change



tamarisk
Tamarix ramosissima, chinensis, parviflora, etc



leafy spurge near Pine, CO Euphorbia esula

# Why do some non-native plants become invasive weeds?

#### **Enemy release hypothesis**

- 1. Absence of natural enemies.
- 2. Unusual genetics, unique strains and hybrids
- 3. Better competitors in disturbed habitats
- 4. Some are drivers of ecosystem change



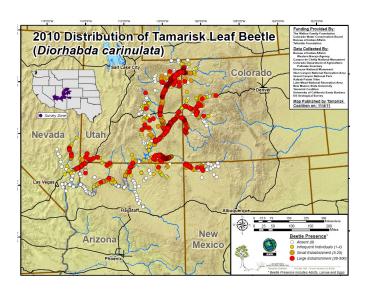
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leafy spurge near Pine, CO Euphorbia esula

Colorado's biocontrol program promotes integrated management in which biocontrol is part of larger plans to diminish impact of invasive species

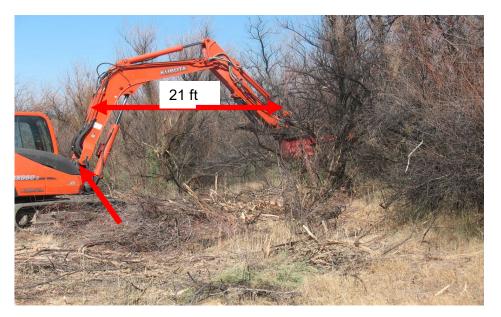
#### Monitoring





Colorado's biocontrol program promotes integrated management in which biocontrol is part of larger plans to diminish impact of invasive species

- Monitoring
- Mowing
- Grazing
- Herbicides



Physical removal of dead tamarisk

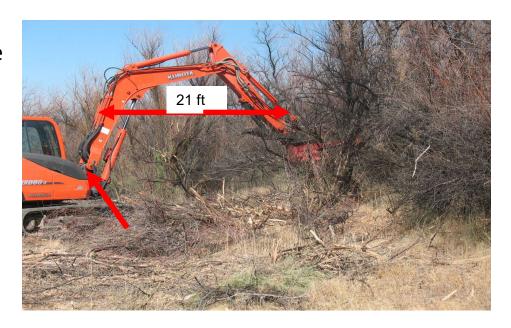
Colorado's biocontrol program promotes integrated management in which biocontrol is part of larger plans to diminish impact of invasive species

- Monitoring
- Mowing
- Grazing
- Herbicides
- Conservation
- Competition
- Restoration



Colorado's biocontrol program promotes integrated management in which biocontrol is part of larger plans to diminish impact of invasive species

- Sustainable
  - i. Self propagation
  - ii. Little or no resistance



#### Target choice for classical biocontrol

- A geographically widespread problem
- Difficult to control using conventional means
- Serious environmental and economic impacts
- Taxonomically distinct from native species

Tamarisk seen as a perfect target, **Saltcedar Consortium** formed for biocontrol development in the 1990s



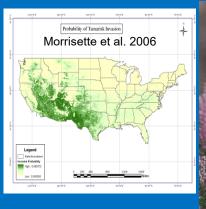
## Tamarisk is a dominant shrub in the west

- *Tamarix* spp. (aka tamarisk or saltcedar) occupy > 500,000 hectares in No. America
- *Tamarix* is the 3<sup>rd</sup> most common woody plant in Western riparian areas (Friedman et al. 2005)

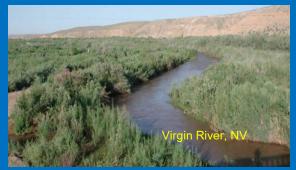




Colorado River









#### Tamarisk is a driver of ecosystem change

- Tamarisk in dense stands increases evapotranspiration (ET) and lowers water tables, which may help it to out compete native vegetation (Nagler et al 2014 Remote Sensing of Environment 140 206-219)
- 2. Tamarisk is fire adapted and with its fine structured leaves and branches **carries fire in riparian ecosystems** (Drus 2013 in "*Tamarix*: a case study of ecological change in the American West").
- 3. Tamarisk **alters soil chemistry** leading to unfavorable conditions for mycorrhizae associated with native vegetation, particularly cottonwoods (Meinhardt, KA and Gehring, CA 2012 Ecol App 22:532-49)



Cottonwood death following tamariskcarried fire, San Pedro River, AZ



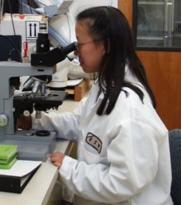
Diorhabda elongata was selected for use against tamarisk by the **Saltcedar** 

**Consortium**, a group of stakeholders and

biocontrol scientists









In the 1990s Jack DeLoach (right) headed up a program for tamarisk biocontrol development

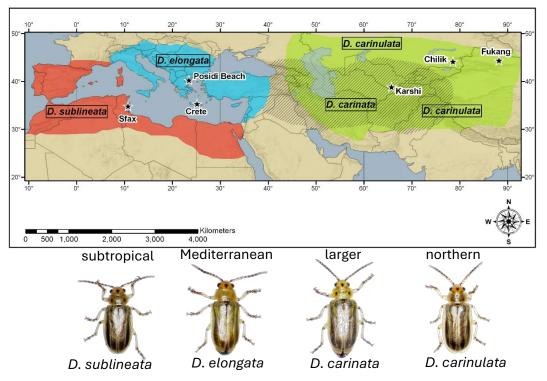
Host range testing as well as research on life cycle and behavior conducted at the USDA ARS facilities in Albany, CA (Ray Carruthers) and Temple, TX, (Jack DeLoach) show beetles in the genus *Diorhabda* to be promising as host specific control agents.



adult *Diorhabda elongata* 



feeding larvae

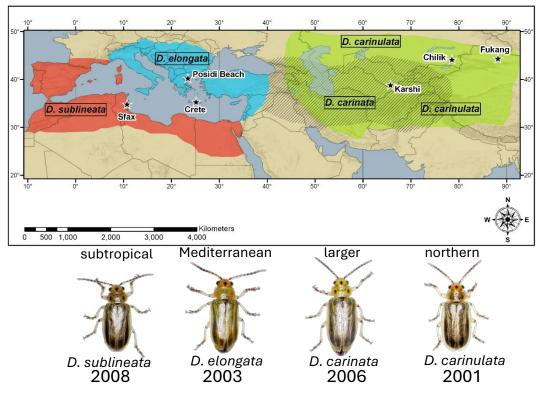


Tracy and Robbins 2009 Zootaxa 2101: 1-152

Widespread defoliation of *Tamarix* and range expansion of *Diorhabda* followed introduction

Four members of the *Diorhabda* species complex were introduced from Eurasia into North America for control of exotic *Tamarix*, beginning in 2001



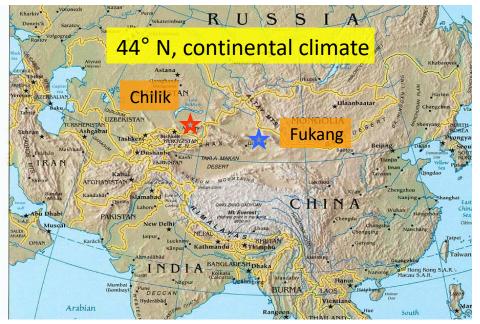


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The first biocontrol agent for tamarisk was Diorhabda carinulata, northern tamarisk beetle originated from two sites in the interior of central Asia.



#### **Northern Adapted Physiology and Phenology**

- Timing of entry and exit from diapause (dormancy)
- Diapause intensity
- Cold tolerance

**Well adapted** or **Maladapted** depended on where the beetle was released in the western US



# Beetles thrived at northern sites (Lovelock, Lovell, Delta)

- Cycles of defoliation and refoliation
- Decrease in flowering
- Die back of branches, decrease of plant biomass
- Mortality (about 30%, 70% at release site)









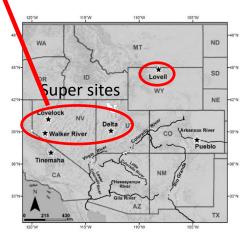
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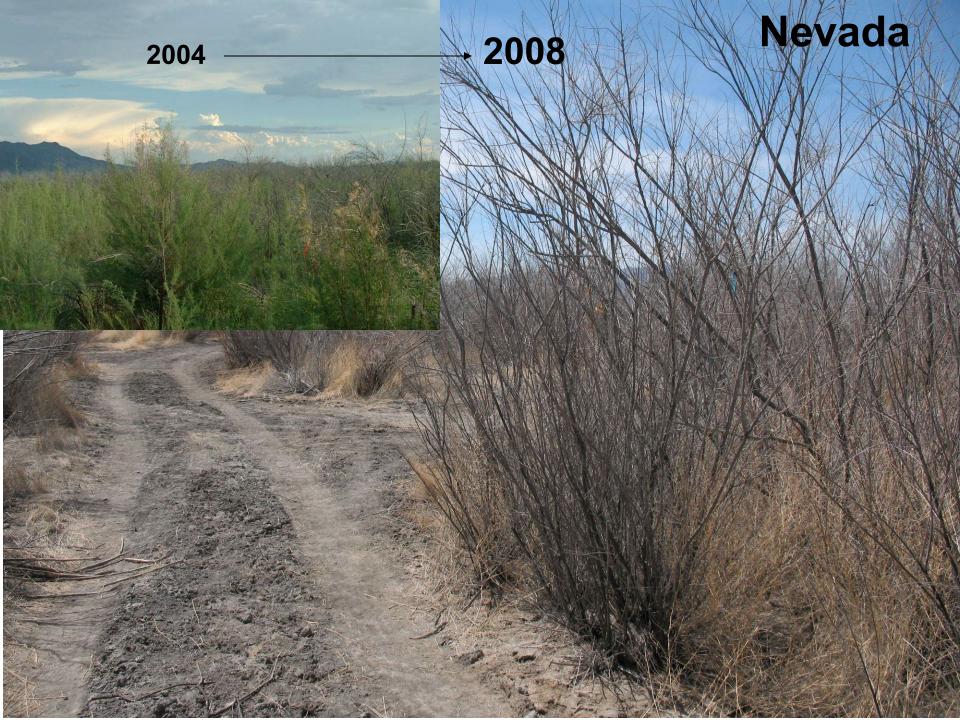




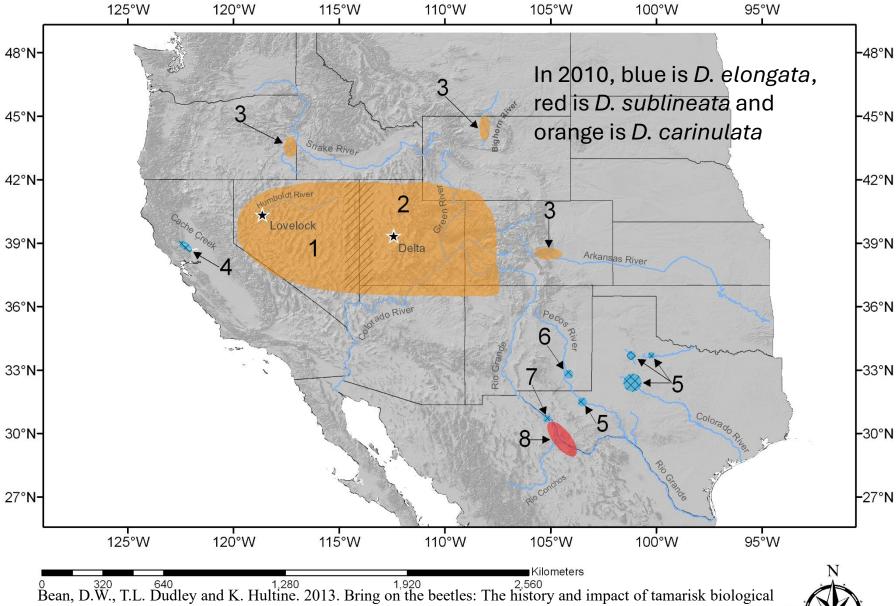




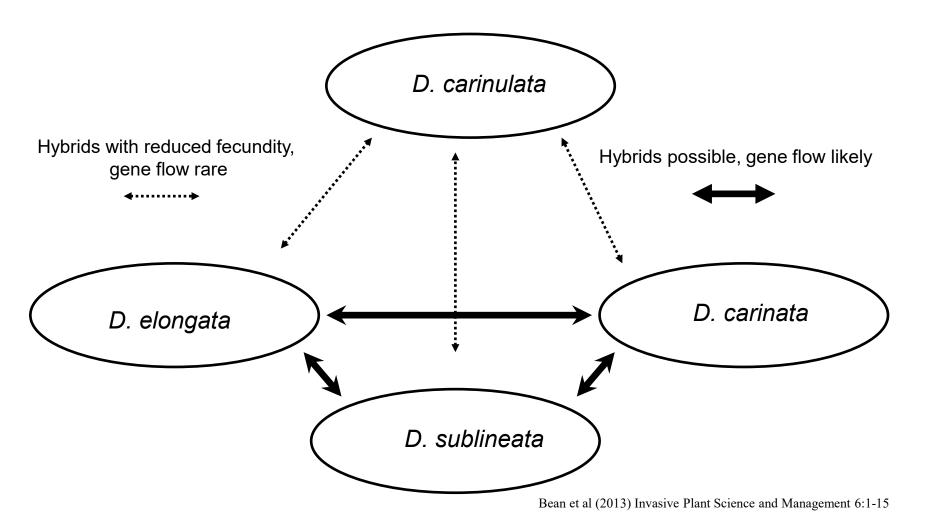
Saltcedar at Lovelock, NV, after two years of defoliation. Photos were taken in May, 2004, after foliage on uninfested plants had fully leafed out.



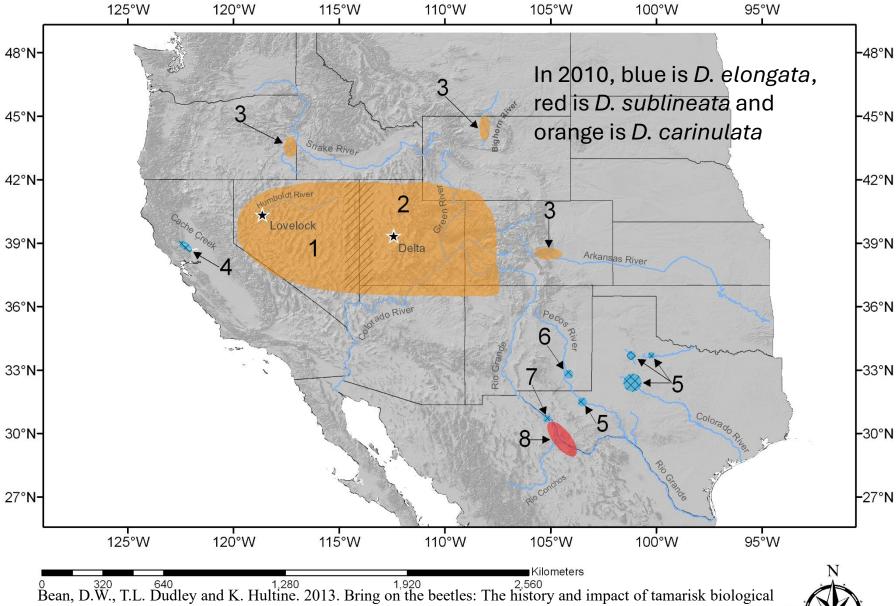




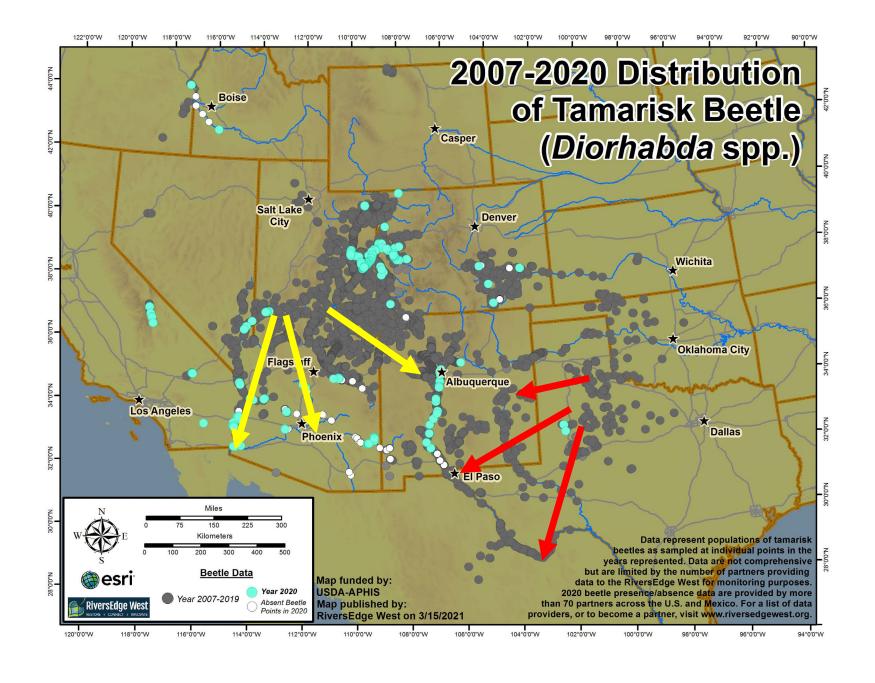
Bean, D.W., T.L. Dudley and K. Hultine. 2013. Bring on the beetles: The history and impact of tamarisk biological control. P. 377-403 In: Sher, A. and M. Quigley (eds). Tamarix: A case study of ecological change in the American West. Oxford Univ. Press.



- 1. D. carinata establishes at a site where a cage had washed away
- 2. Hybrids form between D. elongata, D. carinata and D. sublineata
- 3. D. carinulata adapts, through evolution, to southern conditions

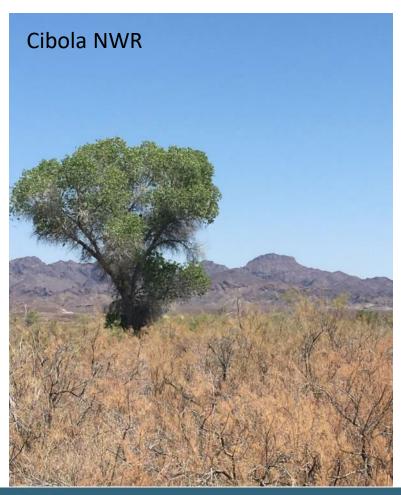


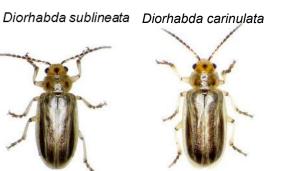
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Southward expanding *D. carinulata* undergo diapause cue evolution, while *D. sublineata*, -2020 Distribution carinata and elongata form a dynamic hybrid of Tamarisk Beetle swarm (Diorhabda spp.) In the western portion of the range D. carinulata\* move southward and In the southeastern portion evolve a shorter daylength of the range hybrid swarms cue for diapause form with D. carinata, D. sublineata and D. elongata Oklahoma City Flags Iff Albuquerque Los Angeles Dallas Phoenix K El Paso Miles Kilometers Data represent populations of tamarisk beetles as sampled at individual points in the years represented. Data are not comprehensive but are limited by the number of partners providing **Beetle Data** Map funded by: data to the RiversEdge West for monitoring purposes. **USDA-APHIS** Year 2020 2020 beetle presence/absence data are provided by more Absent Beetle Map published by: than 70 partners across the U.S. and Mexico. For a list of data providers, or to become a partner, visit www.riversedgewest.org. RiversEdge West on 3/15/2021

### The effective biocontrol of tamarisk

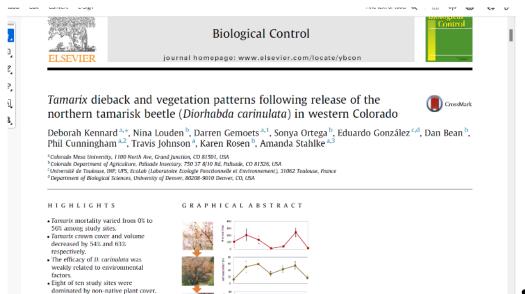


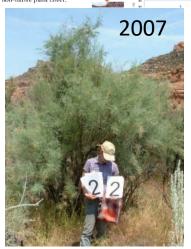




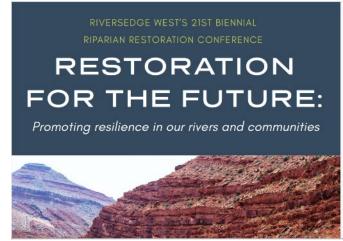
- Found in much of the arid west
- Saves 20,000+ acre-ft/yr in the upper CO basin
- Waves of defoliation/ refoliation

#### **Tamarisk Monitoring and Restoration following Biocontrol**









- Biomass reduction 60%
- Mortality- 30%
- Significant decrease in flowering

Work with Dr. Deb Kennard, Colorado Mesa University



Photo point showing dieback of tamarisk on the Green River, Dinosaur National Monument, seen in 2016 as gray dead branches. Photos were taken near Disaster Rapid, where John Wesley Powell lost one of his boats in 1869.

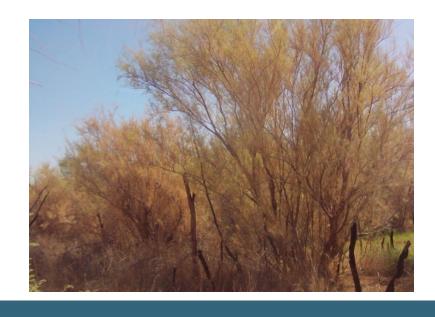
# The biology of *Diorhabda*

- Dispersal, aggregation, defoliation and the chemical cues that control them
- Seasonal timing of life cycle events and synchrony with host plant phenology
- Hybridization and genetics



June 8, 2019 Cibola NWR

June 18, 2019



## Swarming adults/ defoliating larvae



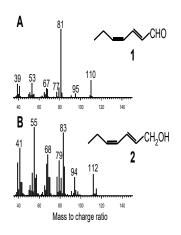






# Insect Attractants: practical uses in weed biological control

In collaboration with Bob Bartelt and Allard Cossé of USDA ARS NCAUR, Peoria



- 1. Male produced pheromone blend
- 2. Plant volatiles from *Tamarix* (mostly 'green leaf' volatiles)



Allard Cossé monitors trap baited with plant volatiles, spring 2004, Lovelock, NV



Pheromone blend attracts adults that are reproductively active

# The tamarisk beetle, *Diorhabda carinulata*, forms aggregations of reproductive males and females in response to a male-produced pheromone blend

Journal of Chemical Ecology, Vol. 31, No. 3, March 2005 (© 2005) DOI: 10.1007/s10886-005-2053-2

THE AGGREGATION PHEROMONE OF *Diorhabda elongata*,
A BIOLOGICAL CONTROL AGENT OF SALTCEDAR
(*Tamarix* SPP.): IDENTIFICATION OF TWO
BEHAVIORALLY ACTIVE COMPONENTS<sup>1</sup>

ALLARD A. COSSÉ,<sup>2,\*</sup> ROBERT J. BARTELT,<sup>2</sup> BRUCE W. ZILKOWSKI,<sup>2</sup> DANIEL W. BEAN,<sup>3</sup> and RICHARD J. PETROSKI<sup>2</sup>

- Discovered by Allard Cossé and Bob Bartelt of the USDA ARS
- Calls in swarms of reproductive beetles that can cause tamarisk defoliation

after aggregation they form mating pairs



Allard



Boh



Near Lovelock, NV, 2003

### First step: Collection of volatiles from feeding beetles





Collector tube with foliage and beetles

Beetles on foliage

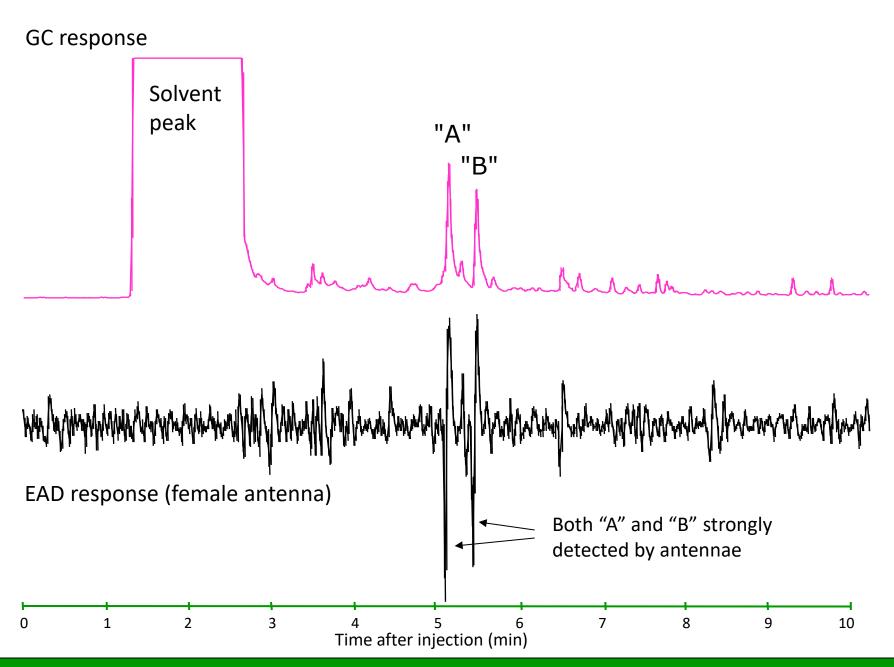


Close up of Super-Q filter

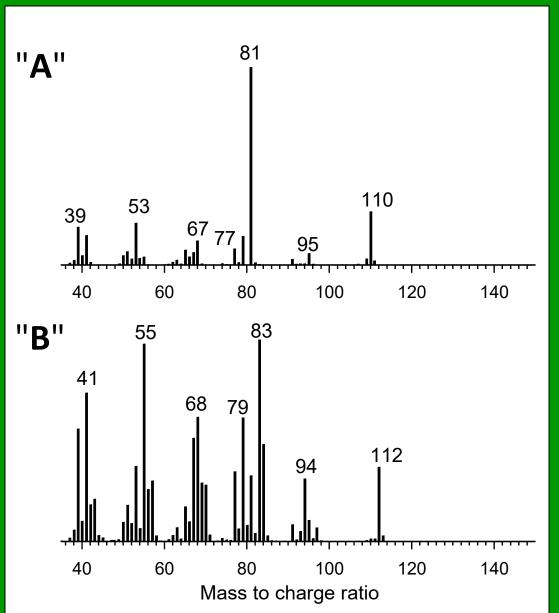
- Draw volatiles emitted from feeding beetles into filter of porous polymer ("Super-Q") with gentle vacuum; later on, rinse filter with solvent.
- On the plus side:

   Beetles + food is a
   "natural" situation; good chance of pheromone emission.
- On the minus side: plant compounds will also be collected.

### GC-EAD Response to Volatiles Collected from Feeding Male D. elongata



# Mass spectra of male-specific compounds and ID's, based on MS library and analytical comparison with standards





(2E,4Z)-2,4-heptadienal = "2E,4Z-7:Ald"



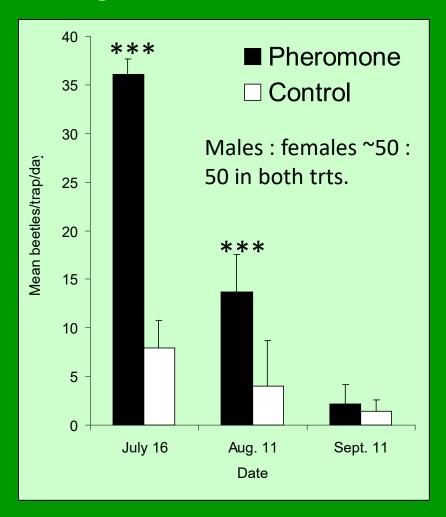
(2*E*,4*Z*)-2,4-heptadien-1-ol = "2*E*,4*Z*-7:OH"

Cossé et al., 2005, J. Chem. Ecol.

### Trapping results at Lovelock, Nevada, during 2003



Close up of sticky trap with pheromone dispensers in place.



Pheromone treatment significantly more attractive than control (P<0.001)

Reproductive tamarisk beetles were observed to avoid areas where larvae were abundant. Gaffke et al discovered a compound emitted by damaged foliage that is repellent to adult tamarisk beetles and could be the signal to indicate high densities of feeding larvae.

Environmental Entomology, XX(XX), 2020, 1–8 doi: 10.1093/ee/nvaa079

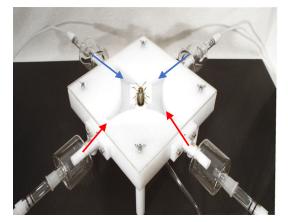


Chemical Ecology

An Herbivore-Induced Plant Volatile From Saltcedar (*Tamarix* spp.) Is Repellent to *Diorhabda carinulata* (Coleoptera: Chrysomelidae)

Alexander M. Gaffke, <sup>1,2,7</sup> Sharlene E. Sing, <sup>3</sup> Jocelyn G. Millar, <sup>4</sup>Tom L. Dudley, <sup>5</sup> Daniel W. Bean, <sup>6</sup> Robert K. D. Peterson, <sup>1</sup> and David K. Weaver<sup>1,6</sup>

<sup>1</sup>Department of Land Resources and Environmental Sciences, Montana State University, Bozeman, MT 59717, <sup>2</sup>Agricultural Research Service, Department of Agricultura, Center for Medical, Agricultural, and Veterinary Entomology, Gaineswille, Ft. 32608, <sup>3</sup>USDA Forest Service, Rocky Mountain Research Station, Bozeman, MT 59717, <sup>4</sup>Department of Entomology, University of California, Riverside, CA 92521, <sup>5</sup>Marine Science Institute, University of California, Santa Barbara, CA 93106, <sup>6</sup>Colorado Department of Agriculture, Palisade Insectary, Palisade, CO 81326, and <sup>6</sup>Corresponding author, e-mait: alexander.gaffke@usda.gov



 Adult reproductive beetles avoid tamarisk with a dense population of feeding larvae

 Foliage damaged by feeding tamarisk beetles emits a volatile compound repellent to tamarisk beetles

Feeding *Diorhabda* larvae

Behavioral assays show that a volatile compound (4-0xo-(E)-2-hexenal) is repellent to adult beetles. Beetle responses to air streams containing the compound (red arrows) are compared with responses to those without the compound (blue arrows).



Field trials conducted by Alex Gaffke, Sharlene Sing and David Weaver at Montana State University, show that semiochemicals can be used to manipulate of tamarisk beetle populations, resulting in defoliation of targeted plants.



Field demonstration of a semiochemical treatment that enhances *Diorhabda carinulata* biological control of *Tamarix* spp.

Accepted: 19 August 2019
Published online: 10 September 2019
Ale

Received: 2 July 2018

Alexander M. Gaffke<sup>1,2</sup>, Sharlene E. Sing<sup>3</sup>, Tom L. Dudley<sup>4</sup>, Daniel W. Bean<sup>5</sup>, Justin A. Russak<sup>5</sup>, Agenor Mafra-Neto<sup>0</sup>, Robert K. D. Peterson<sup>1</sup> & David K. Weaver<sup>0</sup>



Pheromone treated plants are defoliated



Alex Gaffke





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#### Using Chemical Ecology to Enhance Weed Biological Control

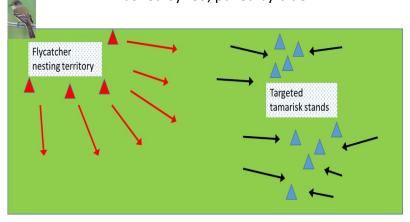
Alexander M. Gaffke 1,2, Hans T. Alborn 1, Tom L. Dudley 30 and Dan W. Bean 4,\*

- 1 Center for Medical, Agricultural, and Veterinary Entomology, Agricultural Research Service, United States Department of Agriculture, Gainesville, FL 32608, USA; agaff&e@agcenter.lsu.edu (A.M.G.); hans.albom@usda.eov (H.T.A.)
- Department of Entomology, Louisiana State University Agricultural Center, Baton Rouge, LA 70803, USA
- Marine Science Institute, University of California, Santa Barbara, CA 93106, USA; tdudley@msi.ucsb.edu
- Colorado Department of Agriculture, 750 37.8 Rd, Palisade, CO 81526, USA
- Correspondence: dan.bean@state.co.us; Tel.: +1-970-464-7916

Simple Summary: Signaling chemicals produced by one organism that bring about a behavioral response in a recipient organism are known as semiochemicals, with pheromones being a well-known example. Semiochemicals have been widely used to monitor and control insect pests in agricultural and forestry settings, but they have not been widely used in weed biological control. Here, we list

A Push/Pull strategy preserves tamarisk used for nesting

Pushed by red, pulled by blue



- As shown by Gaffke et al, beetles can be attracted to tamarisk stands and will defoliate them
- The southwestern willow flycatcher, a T&E species, sometimes nests in tamarisk so pushing beetles away from nesting territories may enhance ecosystem services
- Pulling beetles to targeted tamarisk with GLVs and pheromones, while
  pushing them away from possible nesting habitat with the repellent, is a
  strategy for selective suppression of tamarisk

# Tamarisk Control on Ute Mountain Ute Tribal Lands with Herbicides and Biological Control

Cynthia S. Brown, PhD¹, Hannah Ertl², Dan Bean, PhD³, Zeynep Özsoy, PhD⁴, Farley Ketchum Sr.⁵, and Emily Swartz<sup>6</sup>

(1) CSU Department of Agricultural Biology, Graduate Degree Program in Ecology; (2) Trees, Water & People Indigenous Lands Program; (3) Colorado Department of Agriculture Palisade Insectary; (4) Colorado Mesa University Department of Biological Sciences; (5) Ute Mountain Ute Tribe Environmental Programs Department; and (6) CSU Department of Forest and Rangeland Stewardship.

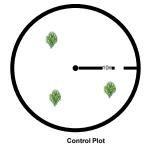
#### Introduction

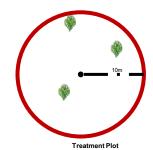
We tested the presence of tamarisk beetle (*Diorhabda carniculata*) GLV and pheromone lures in reducing re-sprout vigor of tamarisk following initial tamarisk treatment.

#### Methods

We used a paired plot design, where control and treatment plots had very similar characteristics. There were 4 plot pairs, or 8 plots total.

#### Paired Plot Design:





- We hung 10 Green Leaf Volatile (GLV) lures in treatment plots during spring leaf-out. GLV alert beetles that growing tamarisk is present.
- We hung 3 Pheromone (PHE) lures in treatment plots 3 times, every 3 weeks through the end of July. PHE are naturally produced by male beetles and stimulate beetle aggregation.

#### A PILOT STUDY



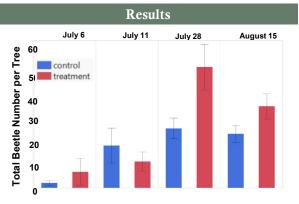


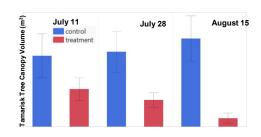
Mastication plot

Modified cut stump plot

We conducted sweep net surveys and monitored the response of tamarisk beetles in control and treatment plots.

We monitored defoliation and canopy volume in three target trees per plot.





- Trees with pheromone lures appeared to have more beetles later in the growing season than controls, but this effect was not statistically significant.
- Tamarisk canopies with pheromone lures appeared to be smaller than controls without lures and to decrease in size over time.

#### **Next Steps**

#### **Pheromone Treatment Scale-up**

- 20 acres of tamarisk treatment in summer/fall 2023
- SCALE UP Pheromone Study after herbicide treatment, beginning spring 2024.

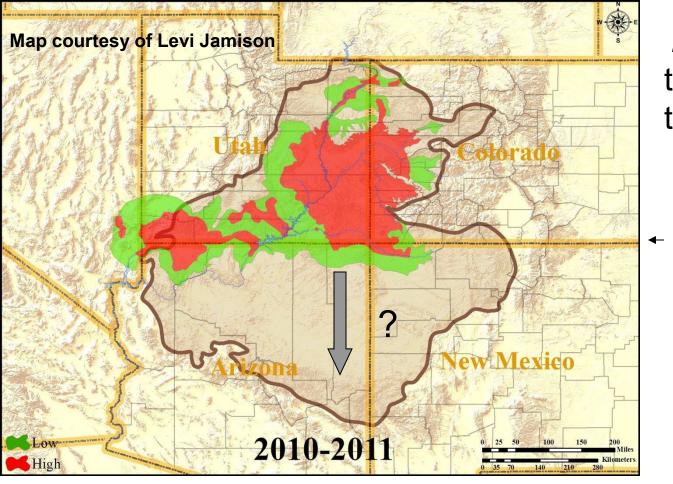












D. carinulata is the **northern** tamarisk beetle

← 37° N

How far and how fast populations of *D. carinulata* moved southward in the basin was a function of diapause timing

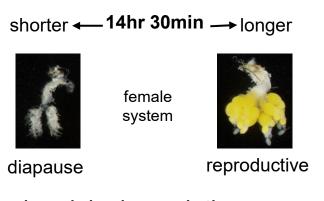
The environmental cues for diapause induction have evolved from being **northern adapted** to **southern adapted**.



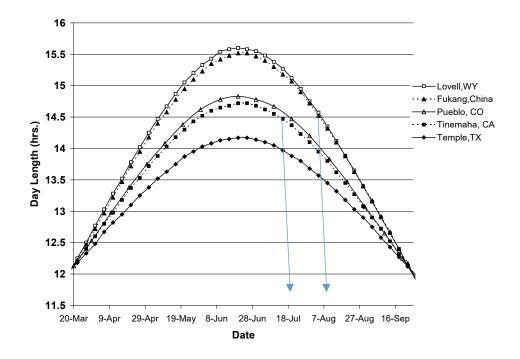
Diapausing beetles (photo: Nina Louden)



Daylength cues diapause or reproduction. 50% mix occurs at the critical daylength (CDL)



in original populations



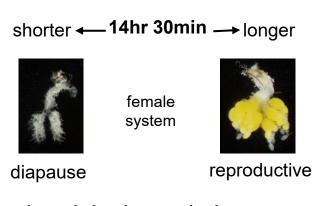
The environmental cues for diapause induction have evolved from being **northern adapted** to **southern adapted**.



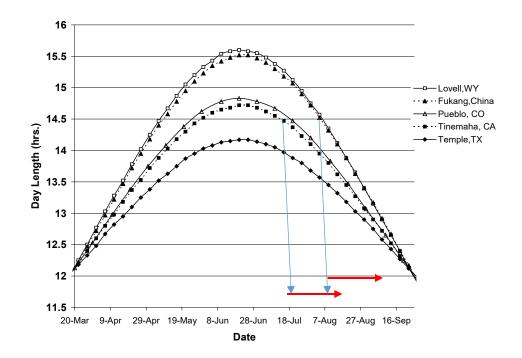
Diapausing beetles (photo: Nina Louden)



Daylength cues diapause or reproduction. 50% mix occurs at the critical daylength (CDL)



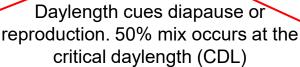
in original populations



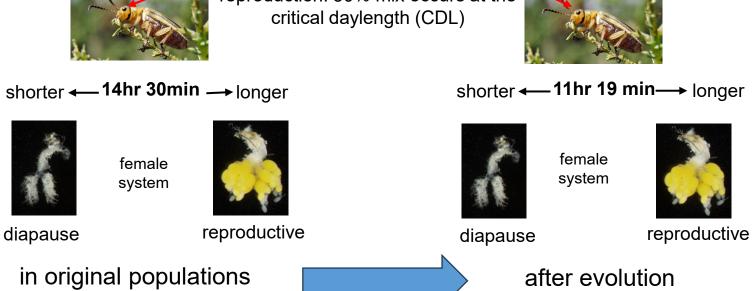
The environmental cues for diapause induction have evolved from being northern adapted to southern adapted.



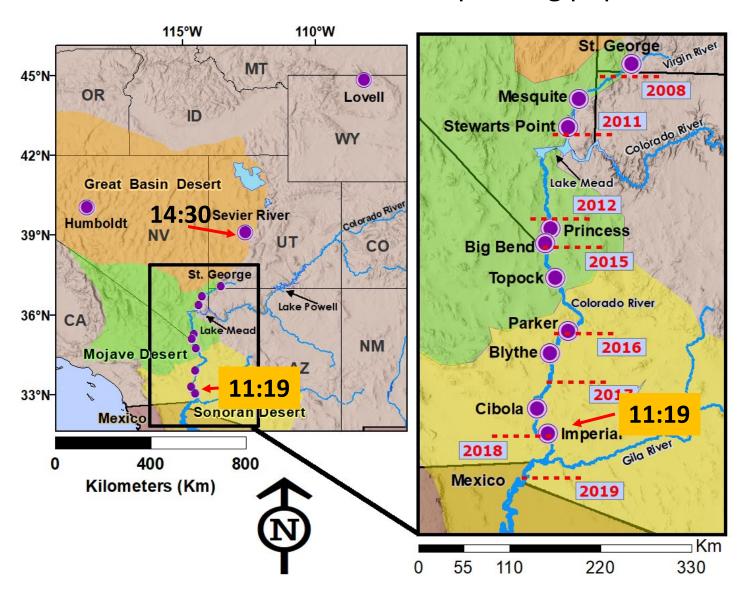
Diapausing beetles (photo: Nina Louden)





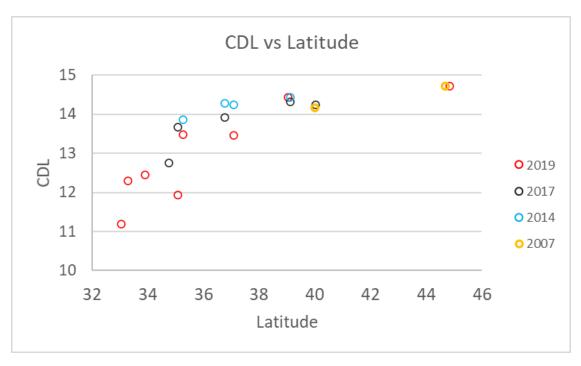


### CDL has evolved in southward expanding populations



Tom Dudley inspects a dead tamarisk shrub near Blythe, CA, 2019





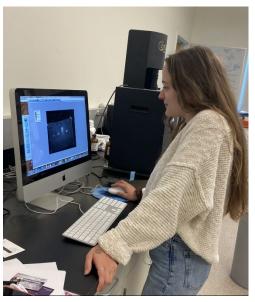
CDL measurements, over a 12-year period from 2007-2019, made under a thermoperiod of 35° day/ 15° night. Field collections were made at or near the leading front of range expansion in the intermountain west on each of the dates listed. The three sites north of the 38° N were original release sites (releases made in 2001).



Zeynep Özsoy mentored Amanda and Emma as undergrads and beyond



Amanda Stahlke brought bioinformatics to Biological Sciences at CMU



Emma Shelton and others are working to identify specific genes

The genome team, working with scientists from the USDA in Hawaii, sequenced the genomes of the four *Diorhabda* species used in biocontrol.



Goal: Sequence the top 100+ US agricultural arthropod pests and beneficial species

USDA-ARS's commitment and contribution to i5K and the Earth BioGenome Project







# Ag100Pest Initiative

We found that new sequencing technology makes assembly somewhat routine

Got a handful of biocontrol agents done too!

Diorhabda spp. and Aphthona nigriscutis







# Compare chromosome structure between the three species that readily hybridize and to *D. carinulata*:

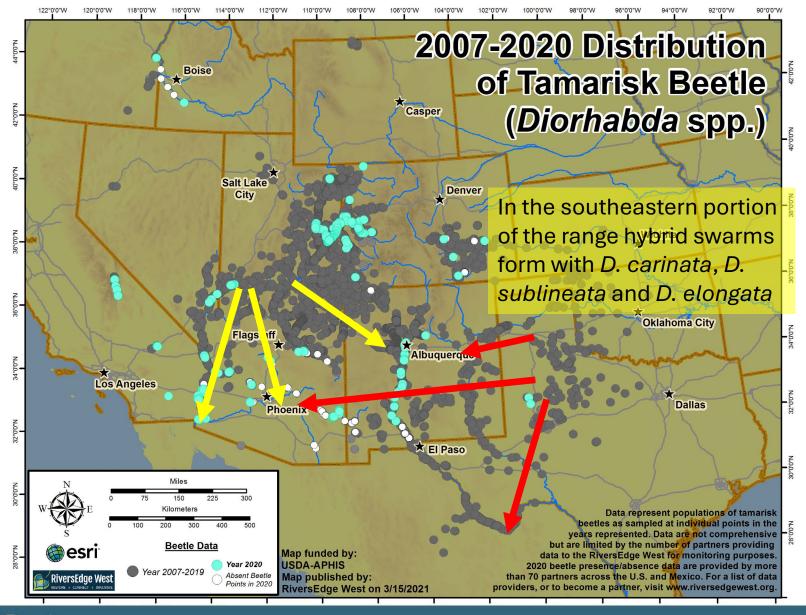
Maintenance of boundaries, explained by genome architecture

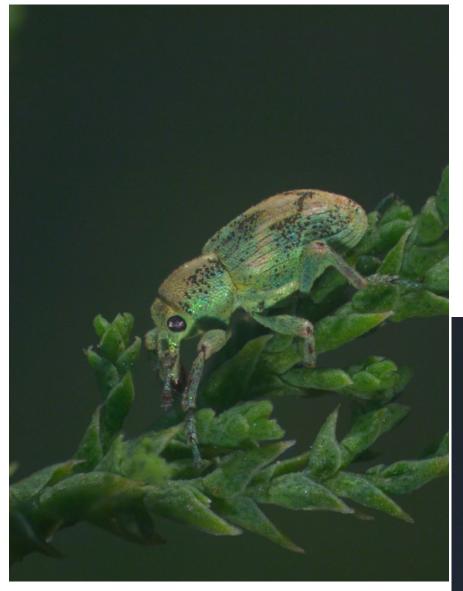
D. carinata

1 2 3 4 5 6 7 8 9 10 11 X Y

#### D. sublineata

*Diorhabda* sister species have 11 autosomes plus an XY. *D. sublineata* and *D. carinata* are similar at the chromosome level





# Coniatus splendidulus

A new tamarisk feeder enters the system 2007





# Coniatus damage Bill Williams River, AZ



Coniatus enter
Colorado in 2011
and are now
widespread



Coniatus larva on tamarisk, highly cryptic

Woven basket where *Coniatus* pupates
This offers protection from predators found in the leaf litter.



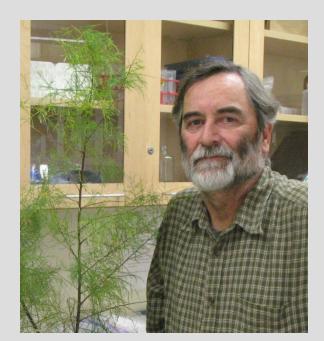
# Coniatus begin feeding earlier in the spring and remain active later in the summer/fall than Diorhabda



Tamarisk branch collected September 28, 2013. *Diorhabda* have been in diapause for about 30 days, *Coniatus* populations have exploded on the regrowth. Adults abundant, baskets abundant on branches with regrowth.

### Riparian restoration is the final step to biocontrol success

**Tom Dudley**, Marine Science Institute, University of California, Santa Barbara



**Bruce Orr**, Stillwater Sciences



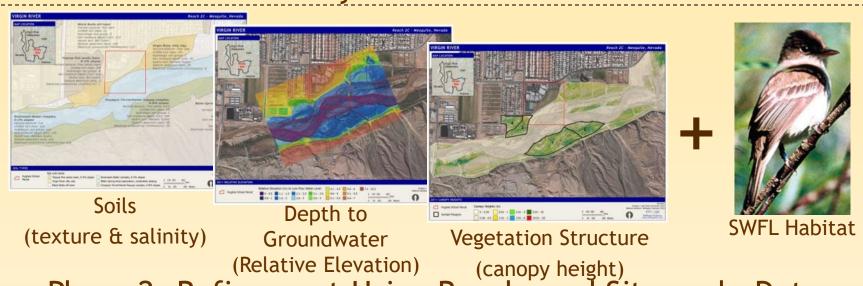


# **EcoHydrological Approach to Restoration**

Stillwater Sciences



### Phase 1: Identify Potential Restoration Areas



Phase 2: Refinement Using Reach- and Site-scale Data

### **Future Projects**

- Russian knapweed gall formers
- Hoary cress mites
- Russian olive mites
- Cheat grass agents (insects and mites)



