



SPECIES INVASIONS ACTIVITY 4990A

# Conservation Biocontrols

A summary of Moaz et al. (2011) and Greenstone et al. (2011)  
Presented by Amaris Bellord





# Supporting Knowledge




WHAT IS A  
CONSERVATION  
BIOCONTROL?



# Conservation Biocontrol

HOW CAN THEY HELP US MANAGE NATURAL LAND?

There are two kinds of conservation biocontrol – habitat or ecological conservation biocontrol and augmentative conservation biocontrols, both of which use native species to control a non-native or invasive species.





# Supporting Knowledge



WHAT IS INTEGRATED  
PEST MANAGEMENT?






# Integrated Pest Management

HOW CAN IT HELP US MANAGE NATURAL LAND?

Integrated pest management is an agricultural technique that focuses on strengthening agroecosystems with a variety of techniques, including biological control and habitat manipulation.





GREENSTONE ET AL. (2011)

Choosing natural enemies  
for conservation biological  
control: use of the prey  
detectability half-life to  
rank key predators of  
Colorado potato beetle

HOW DO YOU SELECT A  
GOOD BIOCONTROL AGENT?





# Greenstone et al. (2011)

## INTRODUCTION

- Predator feeding can be varied because they can be omnivorous and eat things like pollen or honeydew in addition to desired pests  
-> so how much are they actually eating?
- Predators aren't all equally effective at controlling pests.
- Even if remains are detected in a particular predator, it may not reflect direct predation by that predator – could have scavenged or eaten another predator.
- Even if an assay test is positive, representing predation, interpretation of assay data is not straightforward. What's the half life of decay? How long ago did the predator eat the prey? And importantly--How often do they eat it?



# Greenstone et al. (2011)





# Greenstone et al. (2011)

## INTRODUCTION

Goals:

1. Determine half-life for DNA detectability for *L. decemlineata*, the Colorado potato beetle (CPB)
2. Use half-lives to weight the incidence of prey in native predators.

Three generalists and one specialist\*:

- a. Spotted pink lady beetle – *Coleomegilla maculata*
  - b. Spined soldier bug – *Podisus maculiventris*
  - c. Ground beetle\* – *Lebia grandis*
  - d. Two-spotted stink bug – *Perillus bioculatus*
3. How does weighing half-lives affect the ranking of the effectiveness of these predators against traditional stomach content analysis?



# Greenstone et al. (2011)

## METHODS

### "Wild" Predator Study

- Bugs collected in agroecosystem potato field in Beltsville, Maryland at BARC (Beltsville Agricultural Research Center)
- Ten predator samples were taken between May 26 – July 25, 2006, and eight between June 4 – July 30, 2007, by hand-searching foliage, resulting in a total 351 predators collected.
- Stored in 75% EtOH at 4°C in individual 5-ml glass vials that were stored until DNA extraction



# Greenstone et al. (2011)

## METHODS

### Lab Feeding Study

- Leptinotarsa juncta (false potato beetle) was the control feed
- Use laboratory reared animals for all except L. grandis
  - L. grandis came from a field in August 2007
  - Were fed only L. juncta for 3 weeks before experiment
- Tested against 36 other species too!
- Used one egg of CPB 24-48 hours after oviposition as the experimental feed
- Starved predators for a period between 24 and 120 hours
- If refused to feed after 2 hours with CPB egg, excluded
- After feeding, put predators in individual petri dishes with water wicks and lights & heat to simulate being 15cm above ground in the BARC field because half-lives are temperature sensitive.

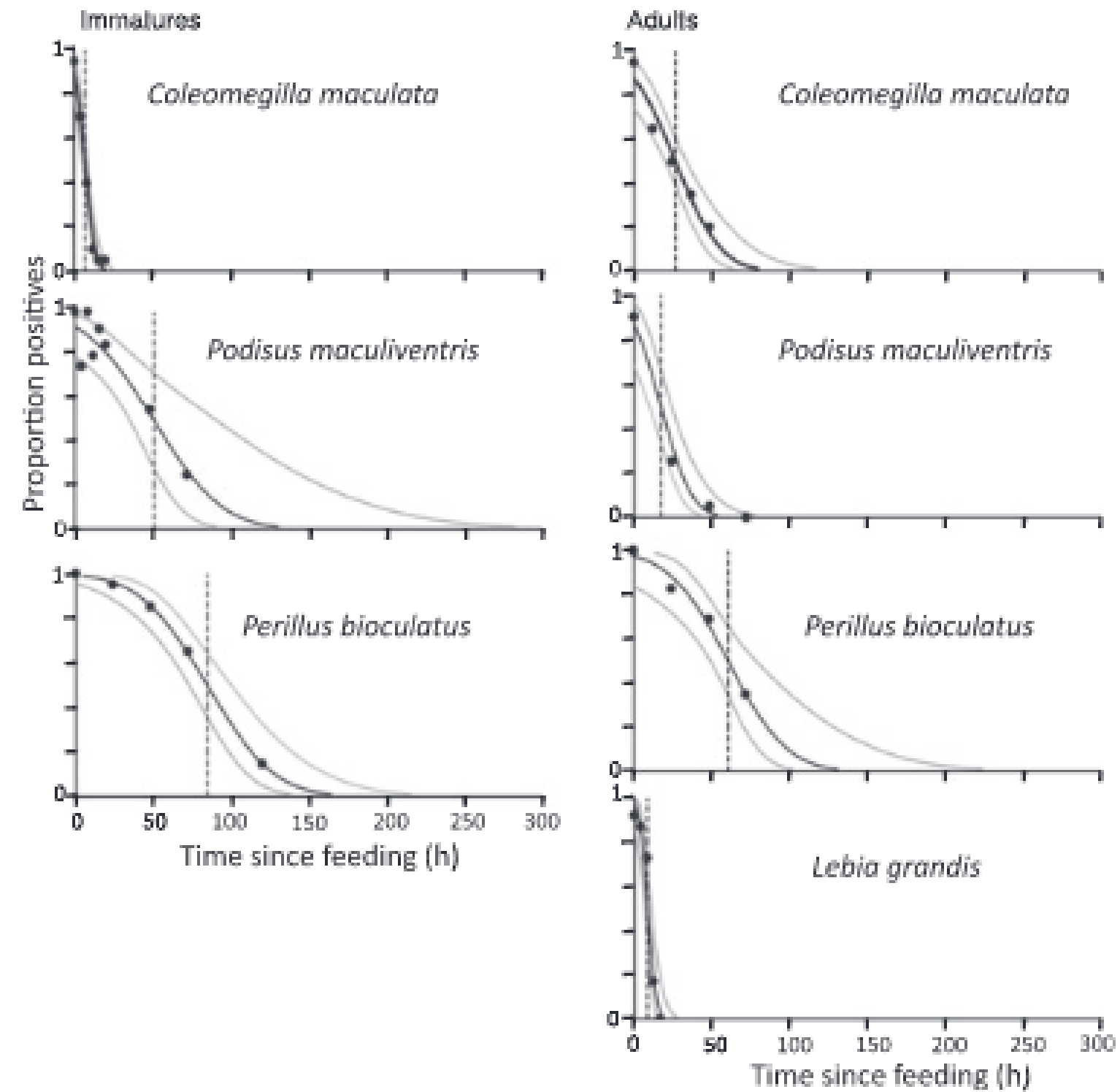


# Greenstone et al. (2011)

Species	Collecting locality (all in USA)		
ARANEAE			
Lycosidae			
<i>Pardosa milvina</i> (Hentz)	Beltsville, MD		
<i>Rabidosa rabida</i> (Walckenaer)	Beltsville, MD		
Theridiidae			
<i>Achaeearanea tepidarium</i> (CL Koch)	Ellicott City, MD		
INSECTA			
Anthocoridae			
<i>Orius insidiosus</i> (Say)	Charleston, SC		
Aphididae			
<i>Macrosiphum euphorbiae</i> (Thomas)	Beltsville, MD		
Cerambycidae			
<i>Tetraopes tetrophthalmus</i> (Förster)	Beltsville, MD		
Carabidae			
<i>Abacidus permundus</i> (Say)	Beltsville, MD		
<i>Agonum striatopunctatum</i> Dejean	Beltsville, MD		
<i>Agonum punctiforme</i> (Say)	Champaign, IL		
<i>Amara aenea</i> (DeGeer)	Beltsville, MD		
<i>Amara anthobia</i> A Villa & GB Villa	Beltsville, MD		
<i>Amara familiaris</i> (Duftschmid)	Beltsville, MD		
<i>Amara cupreolata</i> Putzeys	Beltsville, MD		
<i>Anisodactylus sanctaecrucis</i> (Fabricius)	Champaign, IL		
<i>Bembidion affine</i> Say	Beltsville, MD		
<i>Bembidion quadrimaculatum oppositum</i> Say	Beltsville, MD		
<i>Bradycellus insulsus</i> (Casey)	Beltsville, MD		
<i>Elaphropus anceps</i> (LeConte)	Beltsville, MD		
<i>Elaphropus xanthopus</i> (Dejean)	Beltsville, MD		
<i>Harpalus herbivagus</i> Say	Champaign, IL		
<i>Harpalus fulgens</i> Csiki	Beltsville, MD		
<i>Harpalus indigenus</i> Casey	Champaign, IL		
<i>Stenolophus dissimilis</i> Dejean	Beltsville, MD		
<i>Stenolophus conjunctus</i> (Say)	Beltsville, MD		
Coccinellidae			
<i>Coccinella septempunctata</i> L.	Beltsville, MD		
<i>Epilachna varivestis</i> Mulsant	Beltsville, MD		
<i>Harmonia axyridis</i> (Pallas)	Beltsville, MD		
Lygaeidae			
<i>Geocorus punctipes</i> (Say)	Beltsville, MD		
Pentatomidae			
<i>Euschistus servus</i> (Vollenhoven)	Beltsville, MD		
<i>Oebalus pugnax</i> (Fabricius)	Beltsville, MD		
Thripidae			
<i>Franklinella occidentalis</i> (Pergande)	Charleston, SC		



# Greenstone et al. (2011)



**Figure 1** Results of *Leptinotarsa decemlineata* DNA half-life studies for immature and adult predators. Regressions and 95% fiducial limits were fitted by a two-parameter probit model (Proc PROBIT; SAS Institute, 1999). The dotted vertical line indicates the half-life.



# Greenstone et al. (2011)

**Table 6** Incidence (%) of cytochrome oxidase I DNA detected by polymerase chain reaction assay in predators collected from conventionally tilled potato plots at the Beltsville Agricultural Research Center in 2006 and 2007 (combined). For each species, the raw incidence was used to calculate the corresponding time since feeding from its probit model, which was then entered into the probit model for *Podisus maculiventris* nymphs to calculate the adjusted incidence

Predator	Incidence (%)		
	Life stage	Raw	Adjusted
<i>Coleomegilla maculata</i>	Adult	10.6	39.4
	Larva	0	0
<i>Lebia grandis</i>	Adult	32.1	88.0
<i>Perillus bioculatus</i>	Adult	84.5	73.0
	Nymph	95.1	75.2
<i>Podisus maculiventris</i>	Adults	75.0	90.4
	Nymph	77.8	77.8

adults < *P. bioculatus* adults < *P. bioculatus* nymphs < *P. maculiventris* nymphs < *L. grandis* adults < *P. maculiventris* adults. The adjusted rankings place adults of the polyphagous pentatomid *P. maculiventris* first, ahead of the stenophagous carabid *L. grandis*. This is a dramatic change from the original rankings based on raw incidence (Table 4), which placed all stages of the two pentatomids first, with the stenophagous *P. bioculatus* before the polyphagous *P. maculiventris*.



# Greenstone et al. (2011)

## RESULTS

- *L. decemlineata* DNA in the guts of 351 field-collected predators ranged from 11 to 95%
- Before adjustment for half-life: *C. maculata* adults < *L. grandis* adults < *P. maculiventris* adults < *P. maculiventris* nymphs < *P. bioculatus* adults < *P. bioculatus* nymphs
- After adjustment for half-life: *C. maculata* adults < *P. Bioculatus* adults < *P. Bioculatus* nymphs < *P. Maculiventris* nymphs < *L. grandis* adults < *P. Maculiventris* adults
- This revealed *P. maculiventris* and *L. grandis* to be more effective biological control agents of *L. decemlineata* than the other species considered, suggesting that these would be good targets for conservation management.

# Greenstone et al. (2011)

## DISCUSSION

- First study to measure prey detectability half-lives for arthropod predators of an insect pest, and to use them to evaluate and rank the impacts of adults and juvenile predator species.
- Showed the effectiveness of a new method to detect the value of different predators.
- Method also supports low habitat disruption to determine possible biological control agents. Can collect sample predators for lab analysis instead of repeated disturbance or pest releases.



**MAOZ ET AL. (2011)**

Biocontrol of perseae  
mite, *Oligonychys*  
*perseae*, with an exotic  
spider mite predator and  
an indigenous pollen  
feeder

MULTIPART EXPERIMENT ON  
TWO BIOCONTROL METHODS



## INTRODUCTION

- Persea mite, which causes necrotic spotting on avocado trees, first noted in Israel in 2001.
- Israel has a history as a pesticide-free farm system, so they're very interested in continuing this tradition.
- Phytoseiids can feed on spider mites but their webs protect them.
- Previous studies have shown that generalist predators of spider mites can be effective if provided with additional food sources (i.e. pollen).
- In California, *Euseius hibisci* was identified as a successful predator of persea mites, and Israel has a native *Euseius*.
- Wanted to compare *Neoseiulus californicus* (non-native) and *Euseius scutalis* (native) as biocontrols



## **METHODS**

### *N. californicus* Experiment

- Bought 2000 *N. californicus*, reared 100,000 for use in experiment
- Conducted on Haas avocado trees in five orchards:
  - Kibbutz Hanita in Western Galilee
  - Kibbutz Yodfat in Lower Galilee
  - Kibbutz Dalia in Menashe Heights
  - Kibbutz Ein Hahoreh in Hefer Valley
  - Kibbutz Negba in Northern Negev
- Started in the spring/summer once persimmon mites were spotted
- Each orchard had 5 treatment trees and 5 control trees
- 2,000 mites per tree were released twice, 14 days apart
- Used rapid counting: Every two weeks they counted motile stages on the leaves by counting distal tips as described in Machlitt (1998) to get mite density per leaf

# **Maoz, et al. (2011)**





## **METHODS**

### Biocontrol Survey

- Conducted a two-year survey from 2002–2003 on local phytoseiid species
  - Looked at predatory mites from five orchards every 14 days April to November (when perseas mites are most active) and once per month December to March
  - Five trees sampled per orchard, ten leaves taken per tree for a total count of all predators.
- Found that *E. scutalis* was the most prominent locally, so moved forward with further experiments

## **METHODS**

Four types of *E. scutalis* experiments: leaf discs, seedlings, corn pollen & trees, and Rhodes grass.

### Leaf discs

- Six perseas mites (*O. perseae*) placed individually on six leaf circles floating in water
- 3 of 6 discs got an *E. scutalis*
- Repeated fifteen times for a total of 45 treatment & 45 control discs to compare surviving perseas mites



## **METHODS**

### Seedlings

- Pests and mites collected from untreated orchard trees
- 23 replications set up as three blocks over time
  - n1 = 5
  - n2 = 8
  - n3 = 10
- Seedlings were placed in a controlled walk-in chamber
- 10 *E. scutalis* were placed on each seedling and given 0.05 g corn pollen on an upper leaf 2x a week
- Once population had established, pollen was brushed away
- Seedlings then infested with 30 perseas mites
- For 6 weeks, added pollen twice a week to different leaves for treatment. No additional pollen for control.
- At end, counted all mites and predators found on plants.

## **METHODS**

### Seedlings subexperiment

- 3rd block of seedlings (n=10) was monitored differently
  - sampled one leaf from each of the ten trees every fourteen days
  - counted all mites and predators on leaf and then again at end to model population dynamics over time



## **METHODS**

### Corn Pollen & Trees

- Conducted July to October 2007
- Sprayed trees with water and applied corn pollen to half
- 5 control trees & 5 treatment trees in each orchard
- Rapid counting as in Machlitt, et al. (1998) every 14 days
- Also selected six limbs to beat 3 times and catch fallen mites
- Repeated measures: one leaf per plant every fourteen days

## **METHODS**

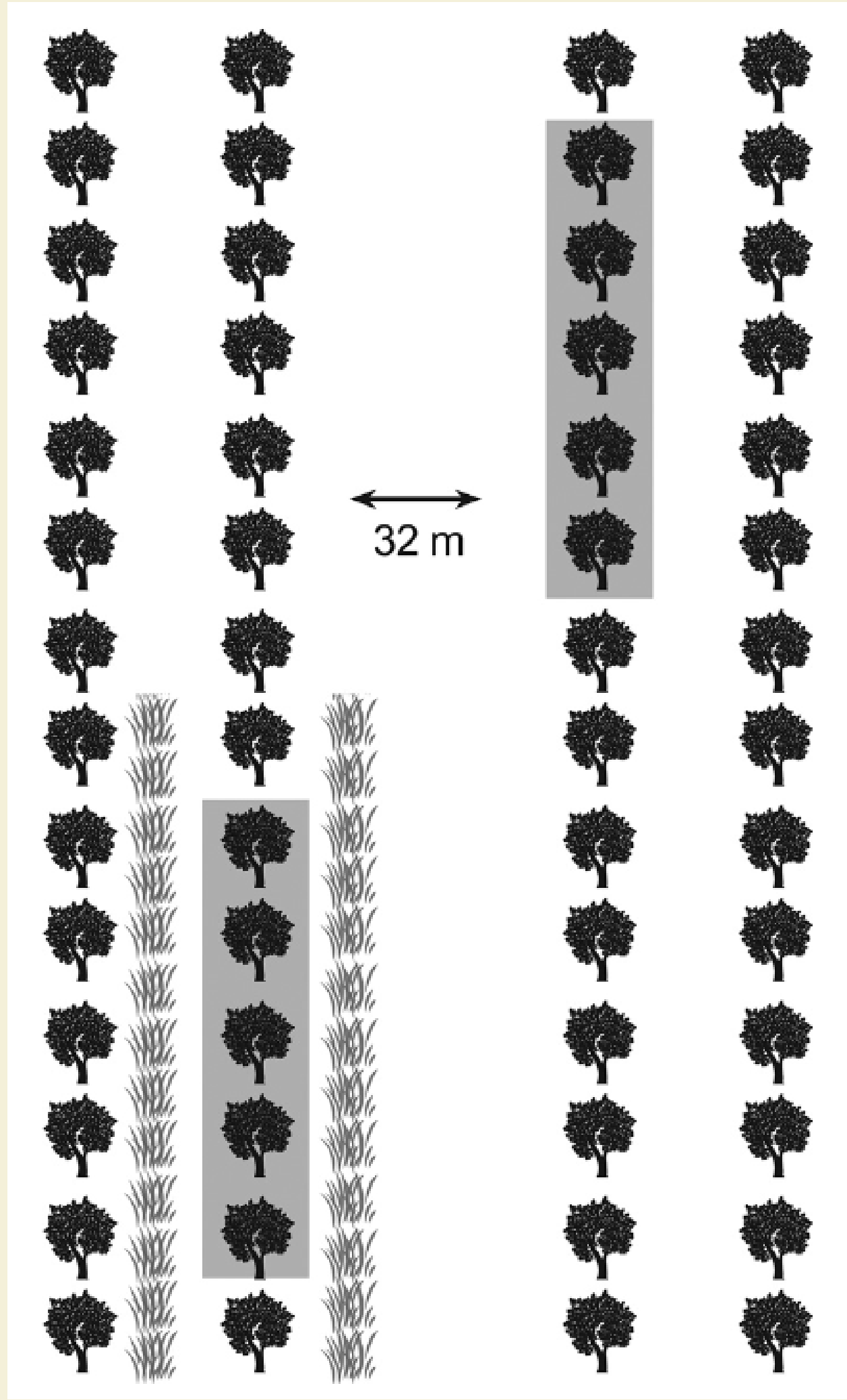
### Rhodes grass

- Conducted August–November 2008
- Patches of Rhodes grass were established between the rows of two orchards (A & B) in a randomized blocks design, replicated four times per orchard for a total of eight replications
- 5 treatment and 5 control per replication (80 total trees)
- Pollen traps used to quantify amount of pollen exposure
- Repeated measures: One leaf per plant every fourteen days

### Rhodes grass subexperiment

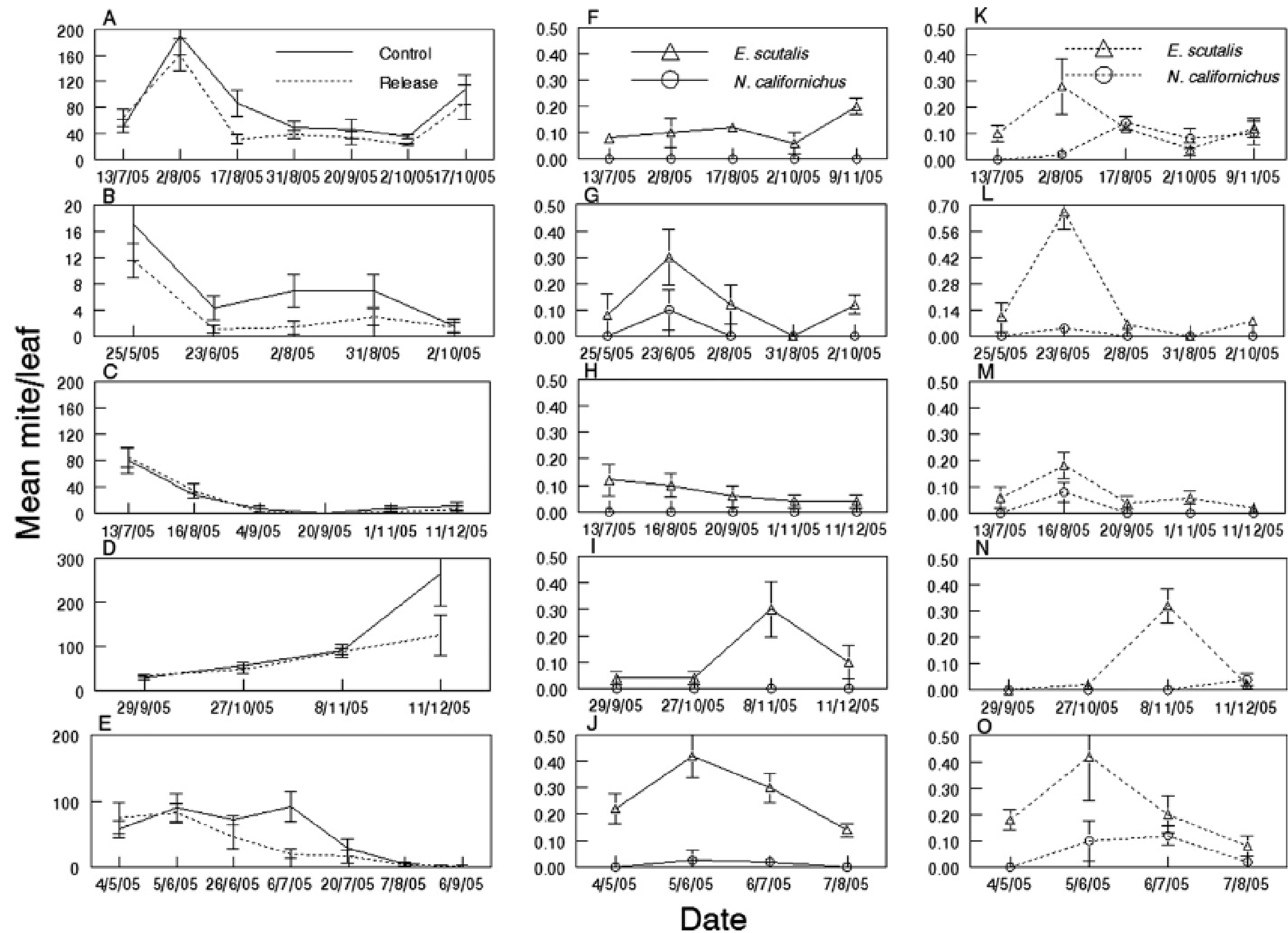
- Measured number and total weight of fruit from Rhodes grass and control tree for comparison of yield

# Maoz et al. (2011)



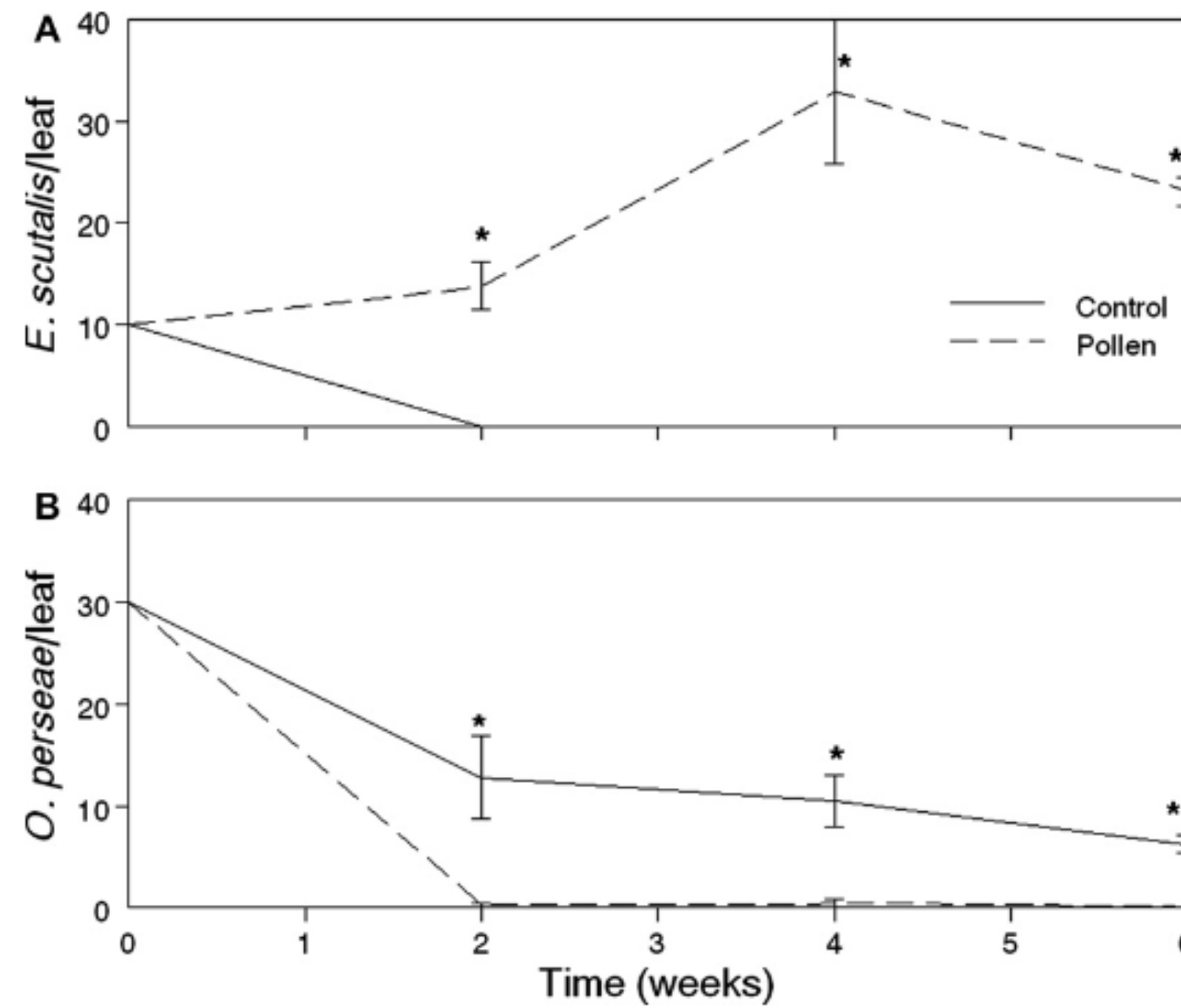


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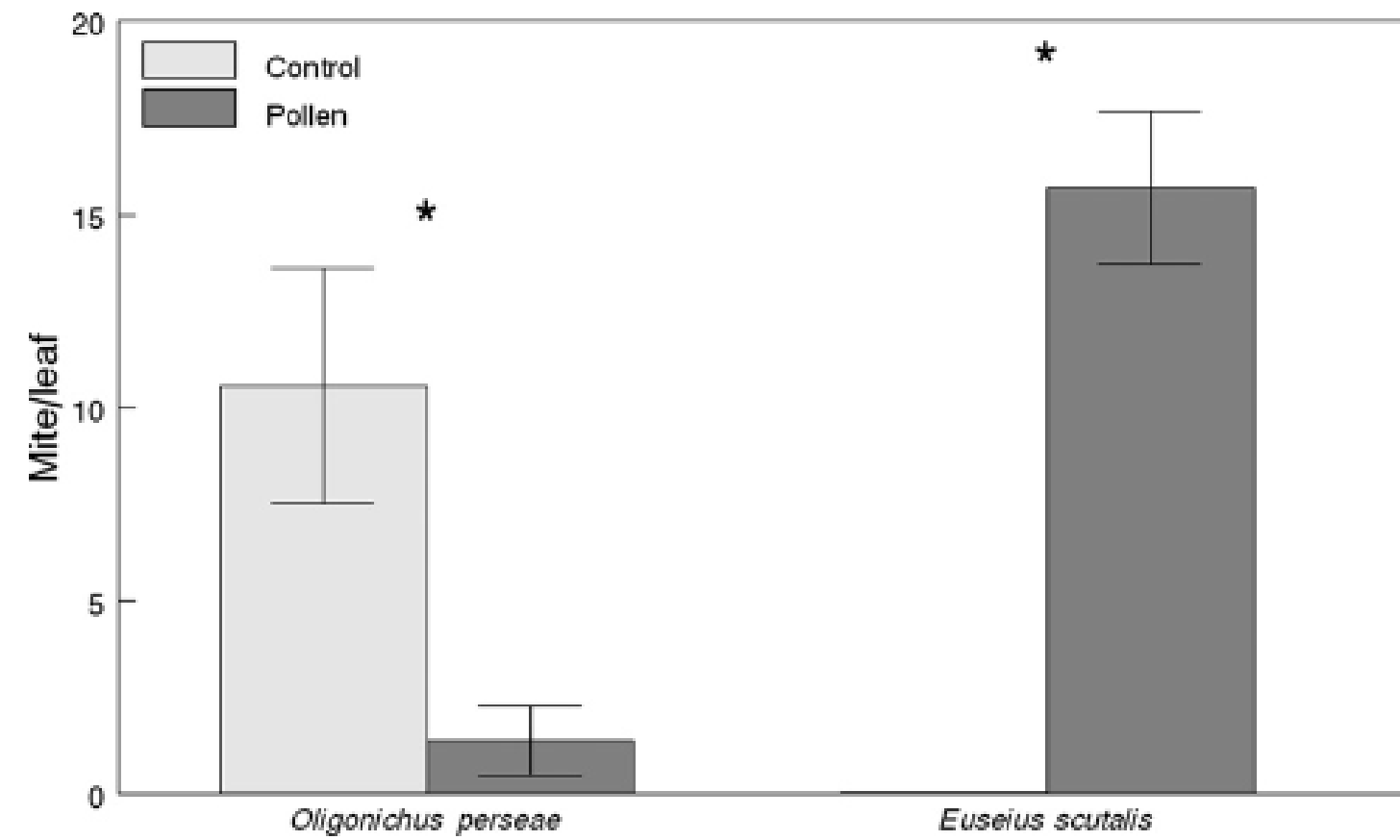
**Fig. 2.** Mean mites/leaf  $\pm$  SE in avocado orchards in five growing regions in Israel, 2005. (A–E) *Oligonychus perseae*; (F–J) *Neoseiulus californicus* and *Euseius scutalis* on control trees; (K–O) *Neoseiulus californicus* and *Euseius scutalis* on release trees. From north to south: (A,F,K) Western Galille, Kibbutz Hanita; (B,G,L) Lower Galilee, Kibbutz Yodfat; (C,H,M) Menashe Heights, Kibbutz Dalia; (D,I,N) Hefer Valley, Kibbutz Ein Hahoreh; (E,J,O) Northern Negev, Kibbutz Negba.

# Maoz et al. (2011)



**Fig. 3.** Motile *Euseius scutalis*  $\pm$  SE (A) and all life stages of *Oligonychus perseae*  $\pm$  SE (B) on control and corn-pollen provisioned avocado seedlings. \* indicates significant differences ( $p < 0.05$ ) between treatments within mite species and sampling time.

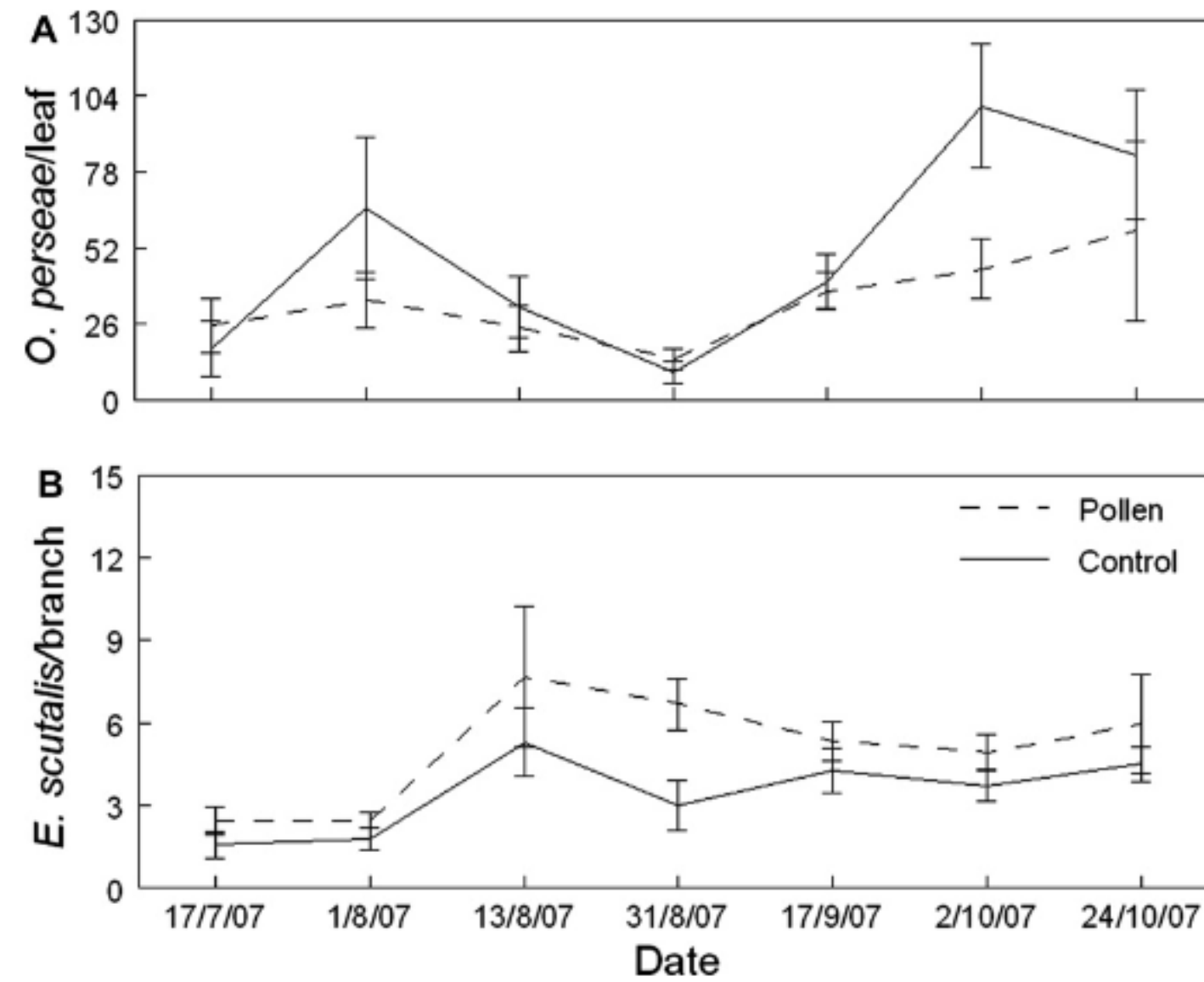
# Maoz et al. (2011)



**Fig. 4.** Mean mites/leaf  $\pm$  SE of *Oligonychus perseae* (eggs and motile stages) and *Euseius scutalis* (motile stages) on avocado seedlings, following six weeks of corn pollen provisioning (twice a week) compared to un-treated control seedlings. \*Significant differences between treatments within mite species (Mann-Whitney U test; *E. scutalis*:  $Z = -5.96$ ,  $p < 0.001$ ; *O. perseae*:  $Z = -4.52$ ,  $p < 0.001$ ).

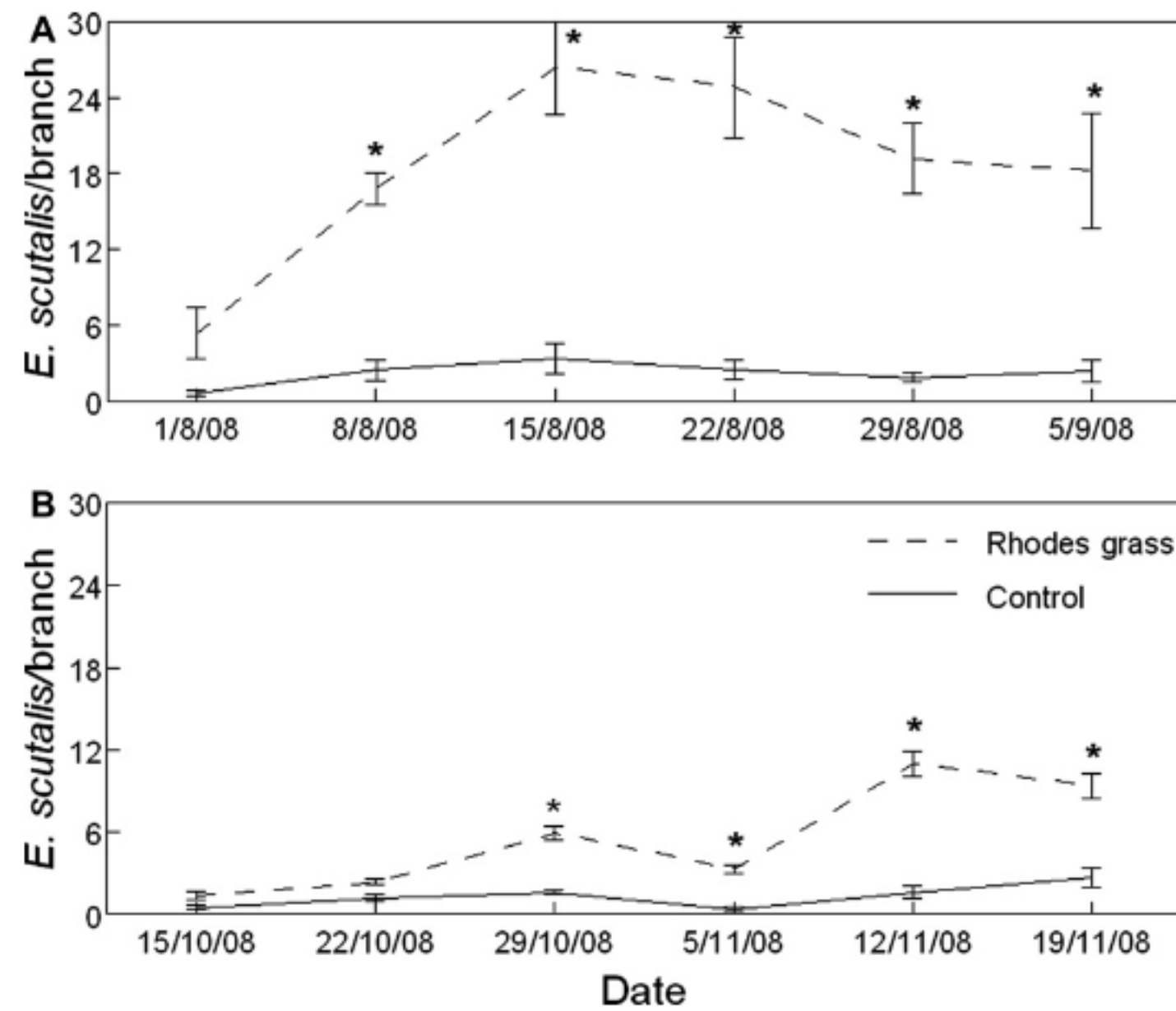


# Maoz et al. (2011)



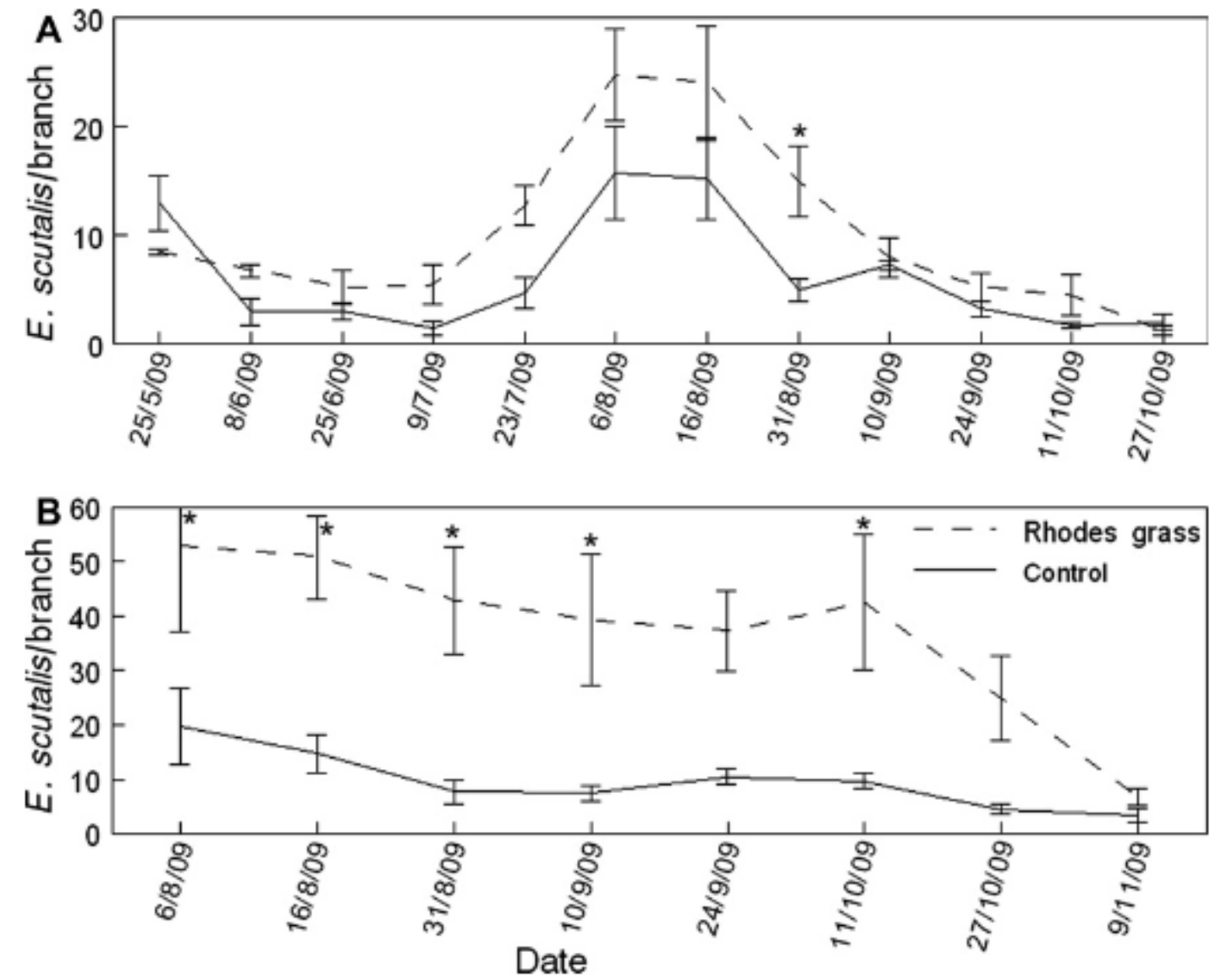
**Fig. 5.** Mean number of *Oligonychus perseae*/leaf  $\pm$  SE (A) and *Euseius scutalis*/branch  $\pm$  SE (B) on trees treated with pollen every fourteen days compared to non-treated control trees, Western Galilee, Israel, 2008.

# Maoz et al. (2011)



**Fig. 6.** Mean number of *Euseius scutalis*/Branch  $\pm$  SE in two orchards (A,B) on avocado trees adjacent to Rhodes grass patches compared to control trees five rows away, in Orchards A and B in Kibbutz Beit Haemeq, Western Galilee, Israel, 2008. \*Significant differences ( $p < 0.05$ ) between treatments within orchards and sampling dates.

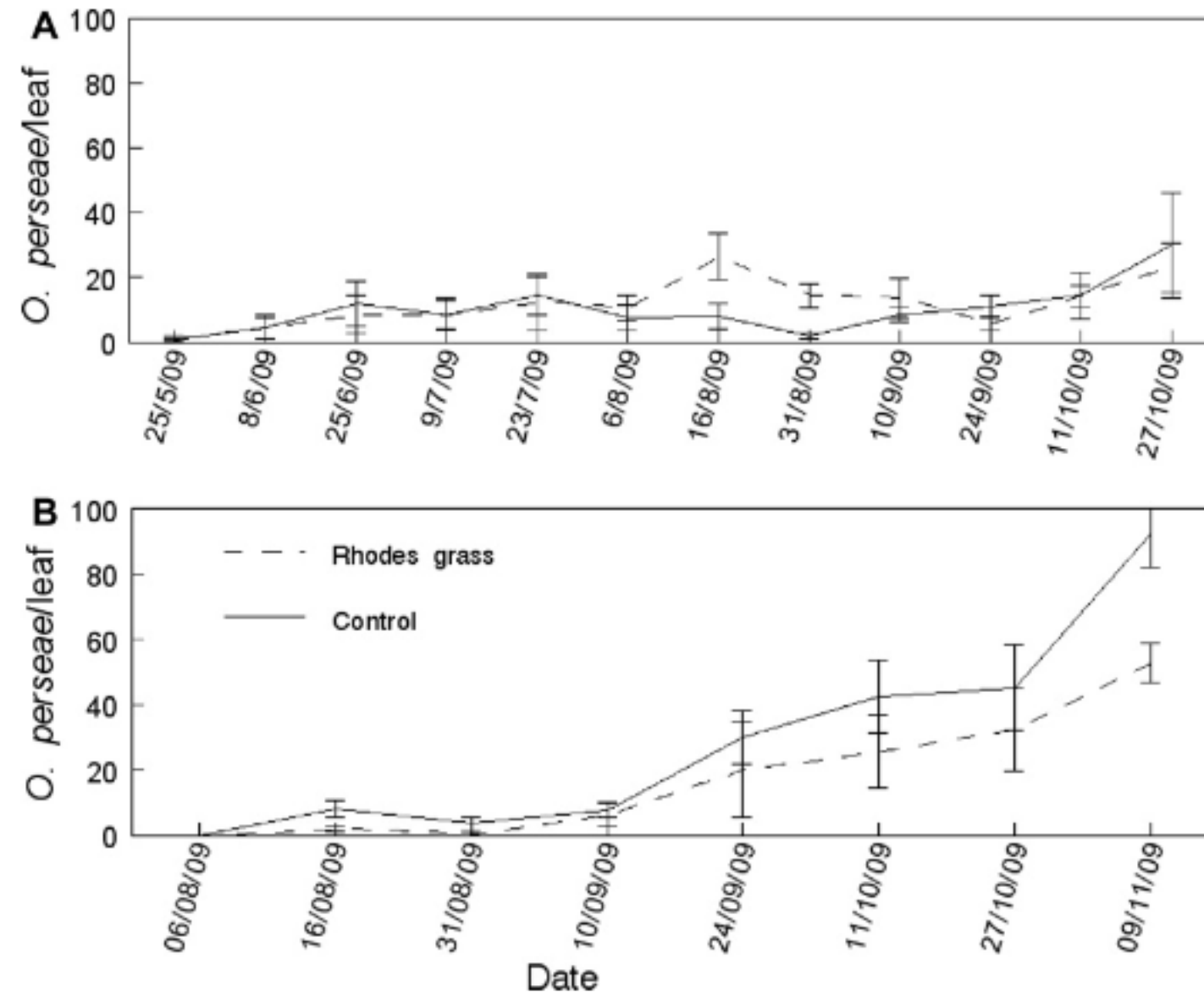
# Maoz et al. (2011)



**Fig. 7.** Mean number of *Euseius scutalis*/Branch  $\pm$  SE in two orchards (A,B) on avocado trees adjacent to Rhodes grass patches compared to control trees five rows away, in Orchards A and B in Kibbutz Beit Haemeq, Western Galilee, Israel, 2009. \*Significant differences ( $p < 0.05$ ) between treatments within orchards and sampling dates.



# Maoz et al. (2011)



**Fig. 8.** Mean number of *Oligonychus perseae*/leaf  $\pm$  SE in two orchards (A,B) on avocado trees adjacent to Rhodes grass patches compared to control trees five rows away, in Orchards A and B in Kibbutz Beit Haemeq, Western Galilee, Israel, 2009.

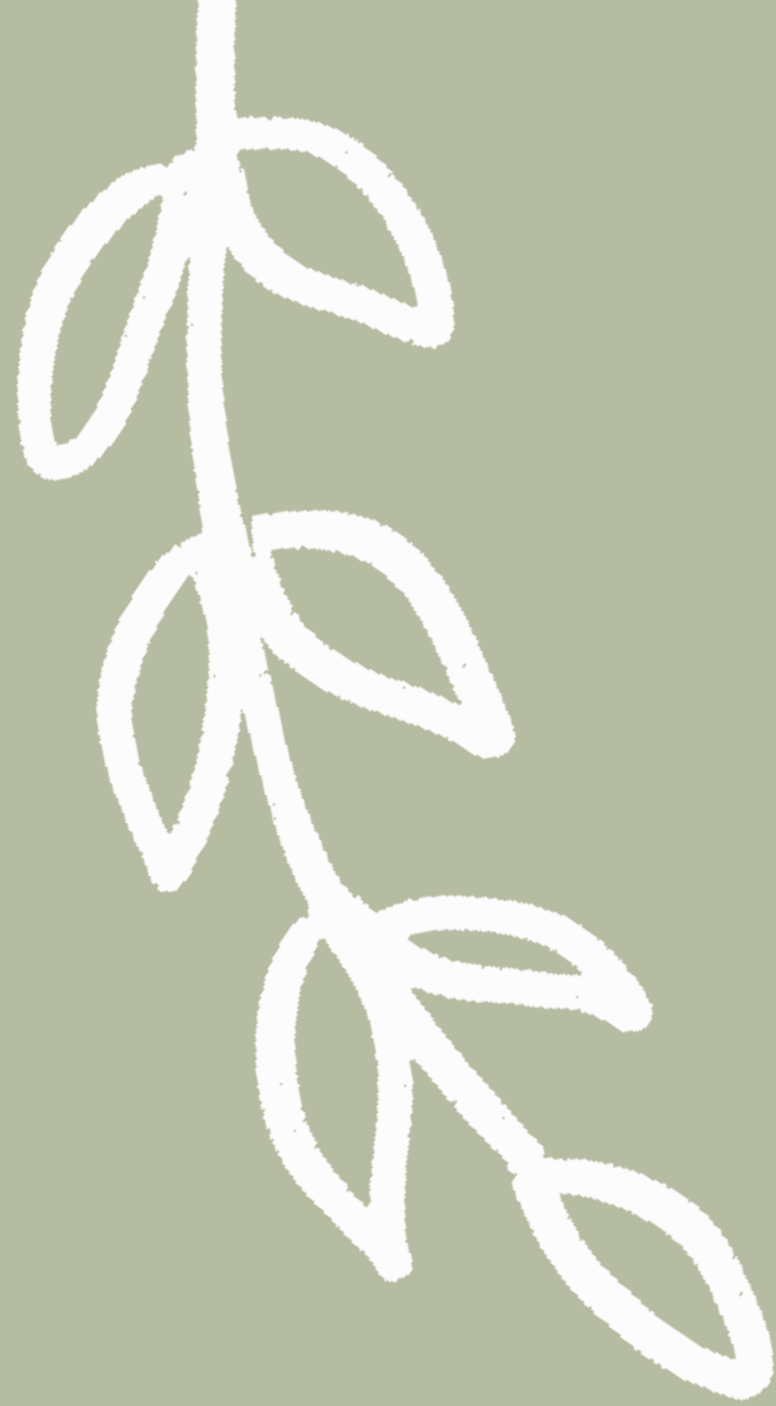
## RESULTS

- *N. californicus* releases on perseas mite populations were significant in three of five orchards
- **But** *N. californicus* populations remained low at 1/3– 1/8 of orchard *E. scutalis* populations
- *E. scutalis* was 96% of native predators, so an easy choice
- 31% and 81% survival on discs with and without *E. scutalis*
- Seedlings without corn had reduced *E. scutalis* populations.
- Seedlings with corn pollen had reduced perseas mites.
- Trees with corn pollen had lower perseas mite populations.
- Rhodes grass significantly increased *E. scutalis* populations and did not significantly decrease perseas mite populations.
- Rhodes grass experiment had no significant yield differences.

## **DISCUSSION**

- Most recovered predators in the survey were Euseius, probably because they're native/better adapted to the local environment.
- The spider mites just couldn't establish, like *D. aligarhensis* here in California.
- Rhodes grass increased *E. scutalis* and *E. scutalis* decreased perseas mites, but Rhodes grass did not decrease perseas mites.
- Still may be valuable to plant Rhodes grass in avocado orchards, as there is evidence for *E. scutalis* as a good conservation biological control.

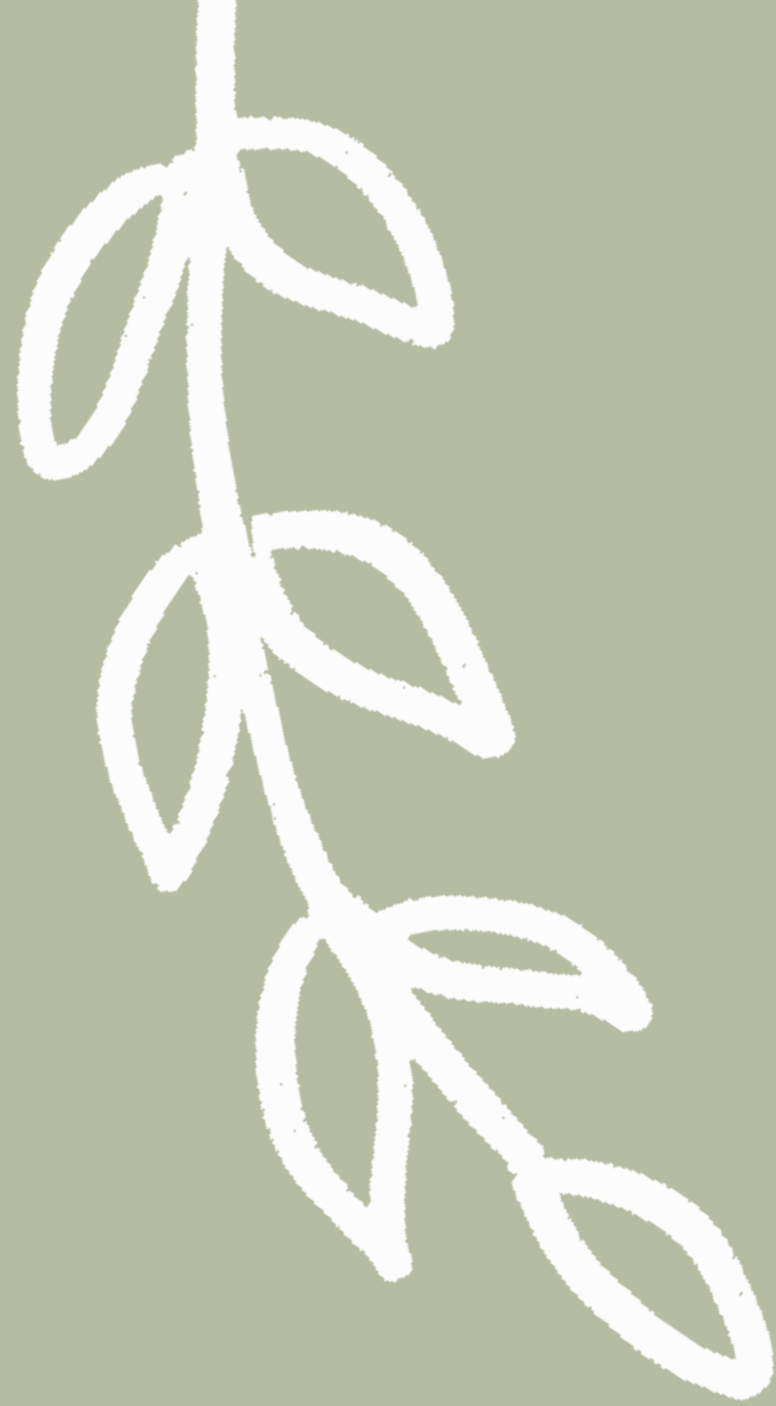




## BIOCONTROL SELECTION

We learned about two different biocontrol selection techniques: surveying local areas, which is perhaps intuitive, and measuring prey selection by predators with DNA half-life analysis – maybe not so intuitive.

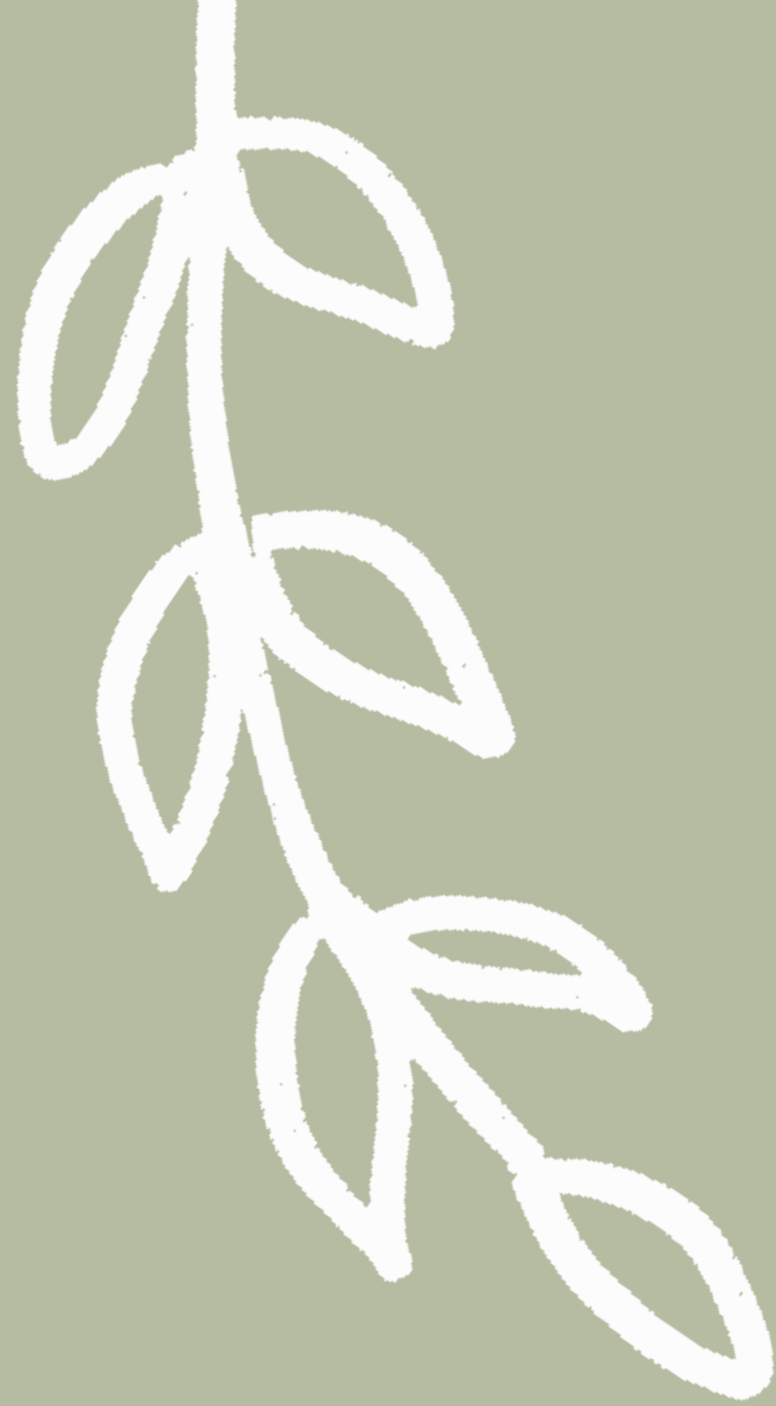
# Takeaways



# Takeaways

## IPM & AGRICULTURAL PRACTICE

Conservation ecologists have to look to agricultural practices for updated land management techniques, especially Integrated Pest Management (IPM).

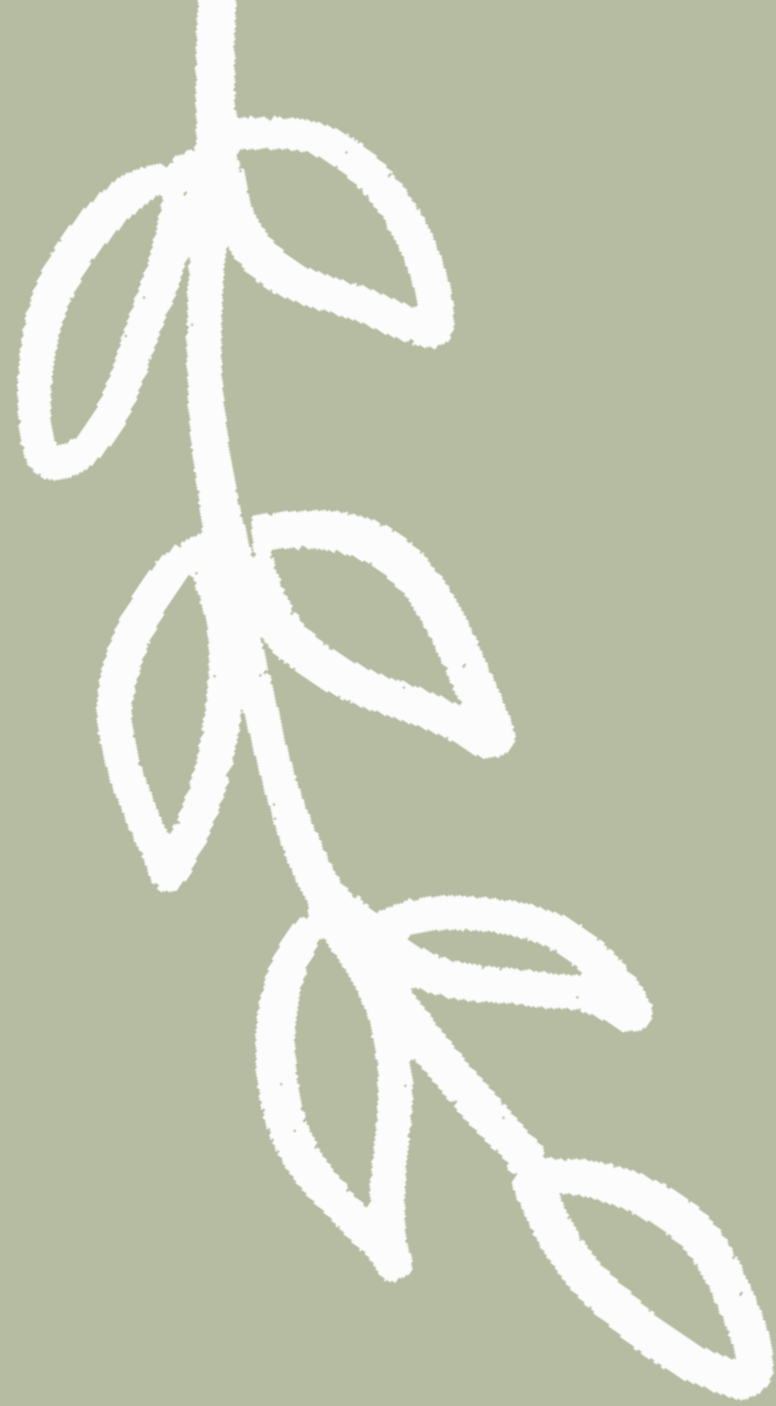


## SISTER CLIMATES

It's important to consider parallel natural land management techniques that are being used by sister countries in areas with similar climates, especially when those countries have a vested agricultural interest in the management of a species.

# Takeaways





# Takeaways

## ECOLOGY

Effective biocontrols may require support species or prefer a variety of pest life stages. A solid understanding of life history, feeding preferences, and local ecological factors that can affect the control species is necessary to not only select a high-quality biocontrol but also to maintain it.

# Citations

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