

Three-cornered Alfalfa Hopper and Other Potential Vectors of Red Blotch



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Grapevine Red Blotch Virus (GRBV)

Background and Timeline



Grapevine Red Blotch Virus (GRBV)

- **Geminiviridae, characterized in 2012**
- **(Krenz et al. 2012, Al Rwahnih et al. 2013, Sudarshana et al. 2015)**

Symptoms

- **Reddening of leaf veins, blotchy red areas on leaf**

Impacts

- **Reduced crop vigor, yield and quality, delayed maturation of fruit**

Geography

- **First reported 2008 @ Oakville Station (Calvi 2011)**
- **Widespread in North America (Krenz et al. 2014)**
- **Archival samples contain virus (Al Rwahnih et al. 2015)**

Spread

- **Humans? Nursery stock? Insects? (Poojari et al. 2013)**

Grapevine Red Blotch Virus (GRBV)

Background and Timeline

Daane / Wilson Research Group

1. Survey insects and plants for virus (2015-2017)
2. Transmission experiments (2015-2018)
3. Threecornered Alfalfa Hopper (TCAH) studies (2017-2020)



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Grapevine Red Blotch Virus (GRBV)

Plant and Insect Surveys

- Can we find the virus outside of vineyards?
- What insects carry the virus?
- Association between insects and positive plants?



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Plant Hosts of Red Blotch Virus

Family	Genus/Species	n	% Positive
Adoxaceae	<i>Sambucus spp.</i>	0/4	0
Apiaceae	<i>Conium maculatum</i>	0/2	0
	<i>Foeniculum vulgare</i>	0/1	0
Apocynaceae	<i>Vinca major</i>	0/17	0
Araliaceae	<i>Hedera helix</i>	0/2	0
Asteraceae	<i>Artemisia spp.</i>	0/3	0
	<i>Baccharis pilularis</i>	0/11	0
	<i>Calendula sp.</i>	0/1	0
	<i>Taraxacum officinale</i>	0/2	0
Betulaceae	<i>Alnus spp.</i>	0/14	0
Brassicaceae	<i>Brassica spp.</i>	0/4	0
	<i>Raphanus raphanistrum</i>	0/1	0
Convulvulaceae	<i>Convolvulus arvensis</i>	0/1	0
Cucurbitaceae	<i>Marah sp.</i>	0/4	0
Ericaceae	<i>Arbutus menziesii</i>	0/9	0
	<i>Arctostaphylos spp.</i>	0/11	0
Fabaceae	<i>Vicia faba</i>	0/6	0
	<i>Vigna sp.</i>	0/4	0

Plant Hosts of Red Blotch Virus

Family	Genus/Species	n	% Positive
Fagaceae	<i>Quercus agrifolia</i>	0/35	0
	<i>Quercus douglassi</i>	0/2	0
	<i>Quercus lobata</i>	0/2	0
Juglandaceae	<i>Juglans sp.</i>	0/8	0
Lauraceae	<i>Umbellularia californica</i>	0/14	0
Malvaceae	<i>Malva parviflora</i>	0/3	0
Oleaceae	<i>Fraxinus latifolia</i>	0/11	0
	<i>Olea europaea</i>	0/11	0
Poaceae	<i>Avena sativa</i>	0/8	0
Rosaceae	<i>Heteromeles arbutifolia</i>	0/40	0
	<i>Prunus spp.</i>	0/10	0
	<i>Rosa spp.</i>	0/5	0
	<i>Rubus spp.</i>	0/28	0
Salicaceae	<i>Salix spp.</i>	0/23	0
Sapindaceae	<i>Acer sp.</i>	0/2	0
	<i>Aesculus californica</i>	0/10	0
Vitaceae	<i>Vitis californica x vinifera</i>	18/68	26
	<i>Vitis vinifera</i>	19/20	95

Plant Hosts of Red Blotch Virus

Additional ground covers from Cieniowicz et al. 2019

Family	Genus/Species	Common names
Fabaceae	<i>Vicia faba</i>	Fava bean
	<i>Vicia Americana</i>	Purple vetch
	<i>Trifolium</i> spp.	Red/white clover
	<i>Pisium sativa</i>	Field peas
Poaceae	Various species	Mixed grasses

Grapevine Red Blotch Virus (GRBV)

Plant and Insect Surveys

- Can we find the virus outside of vineyards?
- What insects carry the virus?
- Association between insects and positive plants?



Insects Positive for Red Blotch Virus

Family	Genus/Species	Mean \pm SEM	n	% Positive
Aphididae		9.8 \pm 1.2	0/122	0
Berytidae		2.7 \pm 1.1	0/7	0
Cicadellidae	<i>Acanalonia</i> sp.	1.0 \pm 0.0	0/3	0
	<i>Aceratagallia</i> spp.	4.0 \pm 1.1	0/55	0
	<i>Acinopterus angulatus</i>	1.3 \pm 0.1	2/26	8
	<i>Alconeura</i> sp.	2.0 \pm 0.0	0/2	0
	<i>Colladonus coquilletti</i>	1.3 \pm 0.2	4/6	67
	<i>Colladonus montanus reductus</i>	1.0 \pm 0.0	1/2	50
	<i>Colladonus</i> sp.	1.0 \pm 0.0	1/2	50
	<i>Deltocephalus fuscinervosus</i>	2.5 \pm 0.4	0/66	0
	<i>Dikraneura rufula</i>	1.0 \pm 0.0	0/3	0
	<i>Dikrella californica</i>	1.0 \pm 0.0	0/2	0

Insects Positive for Red Blotch Virus

Family	Genus/Species	Mean \pm SEM	n	% Positive
Cicadellidae (cont'd)	<i>Empoasca</i> spp.	3.7 \pm 0.8	0/41	0
	<i>Erythroneura elegantula</i>	5.3 \pm 0.6	0/156	0
	<i>Erythroneura variabilis</i>	3.7 \pm 0.5	0/66	0
	<i>Euscelidius schenkii</i>	1.4 \pm 0.3	0/12	0
	<i>Graphocephala atropunctata</i>	1.0 \pm 0.0	0/2	0
	<i>Macrosteles quadrilineatus</i>	1.0 \pm 0.0	0/4	0
	<i>Osbornellus</i> sp.	2.0 \pm 0.0	0/2	0
	<i>Scaphytopius</i> spp.	1.1 \pm 0.1	9/19	47
	<i>Thamnotettix zelleri</i>	2.0 \pm 0.4	0/8	0
Cixiidae	<i>Melaniolarus</i> sp.	2.2 \pm 0.4	0/10	0
Delphacidae		1.2 \pm 0.1	1/21	5
Lygaeidae	<i>Nysius raphanus</i>	5.2 \pm 3.1	0/24	0
Membracidae	<i>Spissistilus festinus</i>	1.7 \pm 0.2	6/38	16
Miridae	<i>Lygus</i> spp.	3.8 \pm 0.5	1/99	1
Pentatomidae		1.5 \pm 0.5	0/2	0
Psyllidae		1.6 \pm 0.3	0/18	0
Rhopalidae	<i>Boisea rubrolineata</i>	4.0 \pm 0.0	0/2	0
Tingidae	<i>Corythuca</i> sp.	7.2 \pm 2.7	0/14	0

Insects Positive for Red Blotch Virus

Additional positives from Cieniowicz et al. 2019

Family	Genus/Species
Aphididae	
Cicadellidae	<i>Osbornellus borealis</i>
	<i>Empoasca</i> sp.
	<i>Graphocephala atropunctata</i>
Cixiidae	<i>Melaniolarus</i> sp.
Phylloxeridae	

Grapevine Red Blotch Virus (GRBV)

Plant and Insect Surveys

- Can we find the virus outside of vineyards?
- What insects carry the virus?
- Association between insects and positive plants?

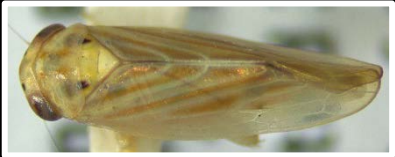




Lygus spp.



Acinopterus angulatus



Colladonus coquilletti



Colladonus montanus



Spissistilus festinus



Scaphytopius spp.

Key Questions

- How high are populations?
- How frequently do they test positive?
- Are they ever found on *Vitis*?

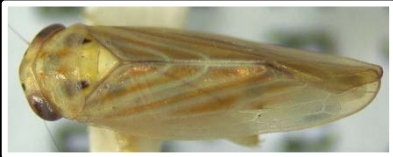


Lygus spp.

- High abundance
- 1% positive
- Not on grape vines



Acinopterus angulatus



Colladonus coquilletti



Colladonus montanus



Spissistilus festinus



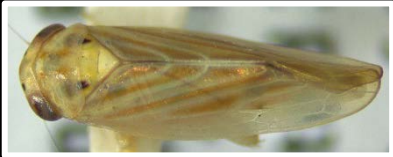
Scaphytopius spp.



Lygus spp.



Acinopterus angulatus



Colladonus coquilletti



Colladonus montanus



Spissistilus festinus



Scaphytopius spp.

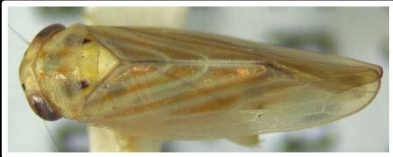
- Low abundance
- 8% positive
- Not on grape vines



Lygus spp.



Acinopterus angulatus



Colladonus coquilletti



Colladonus montanus

- Low abundance
- 50-67% positive
- Not on grape vines



Spissistilus festinus



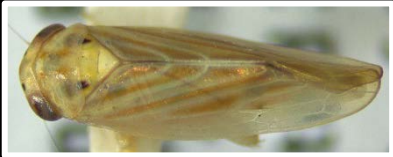
Scaphytopius spp.



Lygus spp.



Acinopterus angulatus



Colladonus coquilletti



Colladonus montanus



Spissistilus festinus



Scaphytopius spp.

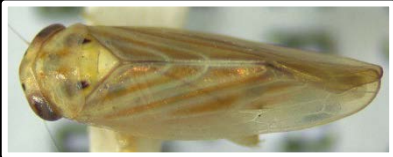
- Low abundance
- 16% positive
- Sometimes on grape vines



Lygus spp.



Acinopterus angulatus



Colladonus coquilletti



Colladonus montanus



Spissistilus festinus



Scaphytopius spp.

- High abundance
- 47% positive
- Frequently on grape vines

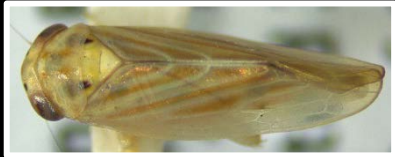
Candidate Rank



Lygus spp.



Acinopterus angulatus



Colladonus coquilletti



Colladonus montanus



Spissistilus festinus



Scaphytopius spp.

Very Low

Low

Medium

High

Grapevine Red Blotch Virus (GRBV)

Plant and Insect Surveys

Results

Key Points:

- Multiple species tested positive
 - This DOES NOT mean that they are a vector
- Differences in...
 - Abundance
 - Frequency of positive GRBV
 - Activity in vine canopy
- Lab vs. Field Transmission
 - Transmission *efficiency* is key

Grapevine Red Blotch Virus (GRBV)

Background and Timeline

Daane / Wilson Research Group

1. Survey insects and plants for virus (2015-2017)
2. Transmission experiments (2015-2018)
3. Threecornered Alfalfa Hopper (TCAH) studies (2017-2019)



A Leafhopper-Transmissible DNA Virus with Novel Evolutionary Lineage in the Family *Geminiviridae* Implicated in Grapevine Redleaf Disease by Next-Generation Sequencing

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1 Department of Plant Pathology, Washington State University, Inigated Agriculture Research and Extension Center, Prosser, Washington, United States of America, **2** Eureka Genomics, Sugar Land, Texas, United States of America

Abstract

A graft-transmissible disease displaying red veins, red blotches and total reddening of leaves in red-berried wine grape (*Vitis vinifera* L.) cultivars was observed in commercial vineyards. Next-generation sequencing technology was used to identify etiological agent(s) associated with this emerging disease, designated as grapevine redleaf disease (GRD). High quality RNA extracted from leaves of grape cultivars Merlot and Cabernet Franc with and without GRD symptoms was used to prepare cDNA libraries. Assembly of highly informative sequence reads generated from Illumina sequencing of cDNA libraries, followed by bioinformatic analyses of sequence contigs resulted in specific identification of taxonomically disparate viruses and viroids in samples with and without GRD symptoms. A single-stranded DNA virus, tentatively named Grapevine redleaf-associated virus (GRLAV), and *Grapevine fanleaf virus* were detected only in grapevines showing GRD symptoms. In contrast, *Grapevine rupestris stem pitting-associated virus*, *Hop stunt viroid*, *Grapevine yellow speckle viroid 1*, *Citrus exocortis viroid* and *Citrus exocortis Yucatan viroid* were present in both symptomatic and non-symptomatic grapevines. GRLAV was transmitted by the Virginia creeper leafhopper (*Erythroneura ziczac* Walsh) from grapevine-to-grapevine under greenhouse conditions. Molecular and phylogenetic analyses indicated that GRLAV, almost identical to recently reported Grapevine Cabernet-associated virus from New York and Grapevine red blotch-associated virus from California, represents an evolutionarily distinct lineage in the family *Geminiviridae* with genome characteristics distinct from other leafhopper-transmitted geminiviruses. GRD significantly reduced fruit yield and affected berry quality parameters demonstrating negative impacts of the disease. Higher quantities of carbohydrates were present in symptomatic leaves suggesting their possible role in the expression of redleaf symptoms.

Citation: Poojari S, Alabi OJ, Fofanov VY, Naidu RA (2013) A Leafhopper-Transmissible DNA Virus with Novel Evolutionary Lineage in the Family *Geminiviridae* Implicated in Grapevine Redleaf Disease by Next-Generation Sequencing. PLOS ONE 8(6): e64194. doi:10.1371/journal.pone.0064194

Editor: Darren P. Martin, Institute of Infectious Disease and Molecular Medicine, South Africa

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Competing Interests: The authors have the following interests to report: Viacheslav Y. Fofanov is employed by Eureka Genomics. There are no patents, products in development or marketed products to declare. This does not alter the authors' adherence to all the PLOS ONE policies on sharing data and materials, as detailed online in the guide for authors.

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Introduction

Nearly seventy viruses and other infectious sub-cellular obligate parasites, collectively referred to as graft-transmissible agents (GTAs), have been documented in grapevines (*Vitis* spp.) [1], [2]. Among all diseases caused either directly or indirectly by these GTAs, grapevine leafroll disease is considered as the most economically important disease affecting plant vigor and longevity and causing significant losses in fruit yield and impacting berry quality attributes [3], [4], [5]. Other virus diseases, such as rugose wood complex, fanleaf infectious degeneration and fleck complex, represent a group of disorders distributed widely in several grape-growing countries around the world [1], [2]. Besides these 'traditional' virus diseases, which can cause significant problems to grape production, other diseases due to GTAs have limited

geographic distribution causing relatively less economic damage to grape production.

In addition to viruses, several viroids belonging to the family *Positiviridae* are ubiquitous in cultivated grapevines [6], [7], [8], [9]. They are *Hop stunt viroid* (HPSVd, genus *Hopstovoid*), *Grapevine yellow speckle viroid 1* (GYSVd-1, genus *Apscavirid*) and 2 (GYSVd-2, genus *Apscavirid*), *Citrus exocortis viroid* (CEVd, genus *Positivirid*) and *Australian grapevine viroid* (AGVd, genus *Apscavirid*). Although these viroids are found in symptomless grapevines, GYSVd-1 has been implicated in vein-banding and yellow speckle symptoms, likely due to a synergistic interaction between GYSVd-1 and *Grapevine fanleaf virus* (GFLV, genus *Nepovirus*, family *Comoviridae*) [10], [11].

Besides their negative impacts on yield and quality of grapes, the introduction and subsequent spread of viruses and other GTAs to healthy vineyards is of great concern for sanitation and grapevine



Grapevine Red Blotch Virus (GRBV)

Transmission Experiments – WSU

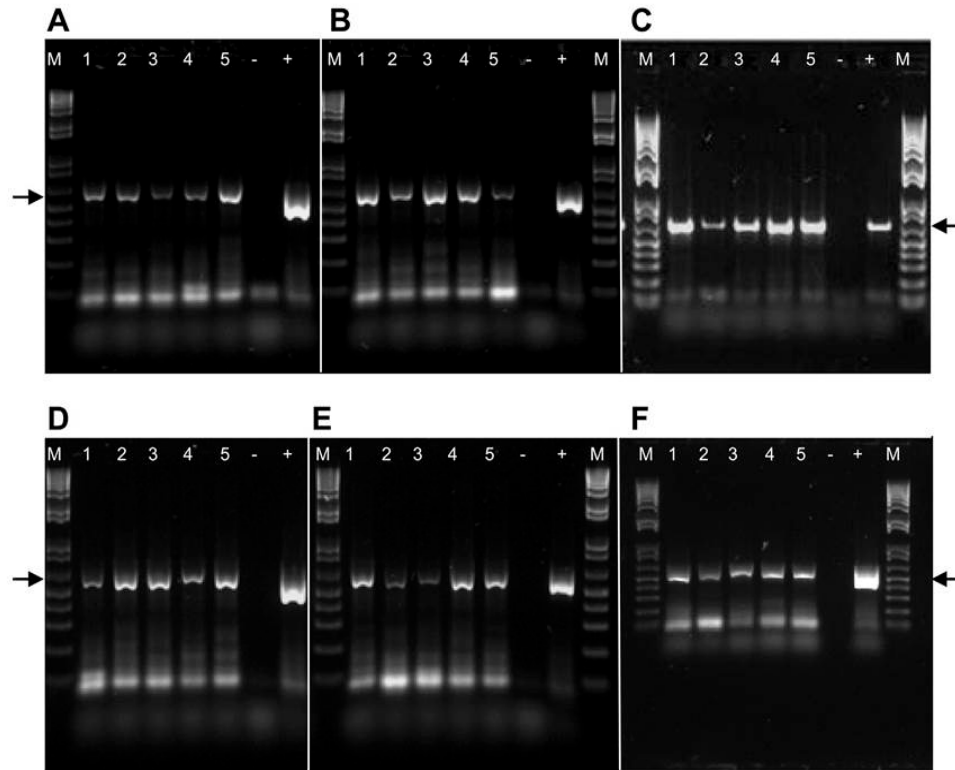


Figure 5. Detection of Grapevine redleaf-associated virus (GRLaV) in grapevines inoculated with viruliferous Virginia creeper leafhoppers. An agarose gel (0.8%) showing amplification of partial IR and CP regions of GRLaV from (A) Cabernet Franc, (B) Chardonnay, (C) Merlot, (D) Pinot Noir, (E) Pixie and (F) Sangiovese. Lanes 1 to 5 represent individual grapevines, '+' represents positive control for GRLaV, '-' represents healthy control negative for GRLaV, and 'M' represents 1 kb plus DNA ladder (Invitrogen). Arrows represent ~550 bp DNA band specific to GRLaV. doi:10.1371/journal.pone.0064194.g005



Methods

- 50 insects / plant
- 6 cultivars (A-F)
- 48 hr Acquisition AP
- 48 hr Inoculation AP
- PCR tests in 4 wks

Our initial goal was to repeat this work and then test similar insects (e.g., WGLH), and common vineyard pests

Grapevine Red Blotch Virus (GRBV)

Transmission Experiments

Focus on Common/Abundant Candidates

Common Name	Species
Western grape leafhopper	<i>Erythroneura elegantula</i>
Variegated leafhopper	<i>Erythroneura variabilis</i>
Virginia creeper leafhopper	<i>Erythroneura ziczac</i>
Grape whitefly	<i>Trialeurodes vittatas</i>
Grape mealybug	<i>Pseudococcus maritimus</i>
Vine mealybug	<i>Planococcus ficus</i>
Blue-green sharpshooter	<i>Graphocephala atropunctata</i>
Foliar phylloxera	<i>Daktulosphaira vitifoliae</i>
Threecornered alfalfa hopper	<i>Spissistilus festinus</i>

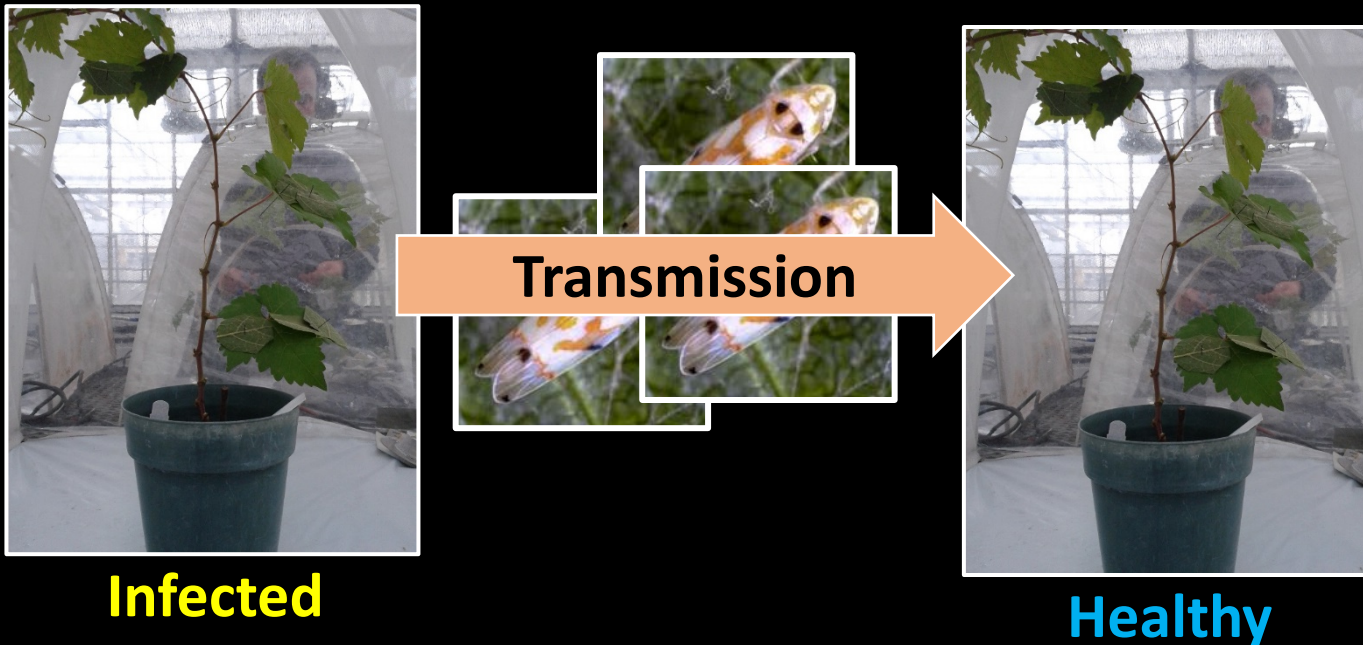
Grapevine Red Blotch Virus (GRBV)

Transmission Experiments

Focus on Common/Abundant Candidates

Experimental Setup

- Potted vines caged in greenhouse
- Cohorts of 10-100 individuals/species
- 3-10 replicates/trial



Grapevine Red Blotch Virus (GRBV)

Transmission Experiments

Focus on Common/Abundant Candidates

Experimental Setup

- 48-72 hr Acquisition (AAP)
- 48-72 hr Inoculation (IAP)



Grapevine Red Blotch Virus (GRBV)

Transmission Experiments

Focus on Common/Abundant Candidates

Experimental Setup

- Vines held for 2 years to observe symptoms and test with PCR



Grapevine Red Blotch Virus (GRBV)

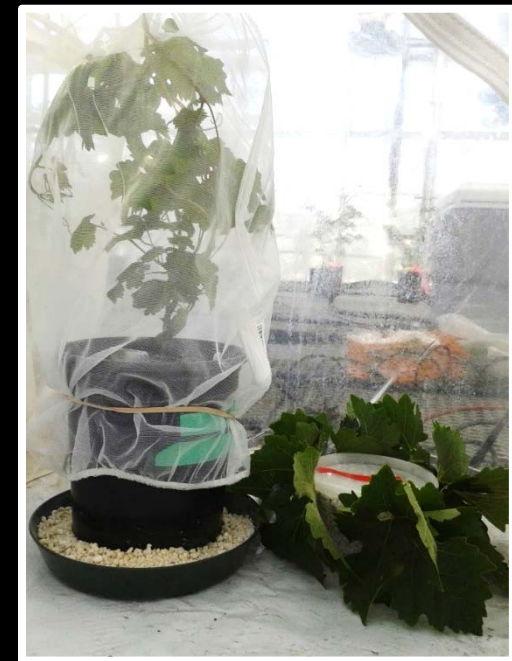
Transmission Experiments

Focus on Common/Abundant Candidates

Experimental Setup

Some variations and repeat experiments

- “Bouquets” of mature leaves from field vines
- Longer AAP/IAP (5 days)
- Paired infected/healthy vines



Grapevine Red Blotch Virus (GRBV)

Transmission Experiments – Results

Family	Species Tested	Acquires	Transmits
Cicadellidae	<i>Erythroneura elegantula</i>	No*	No
	<i>Erythroneura variabilis</i>	No	No
	<i>Erythroneura ziczac</i>	Yes	No
	<i>Graphocephala atropunctata</i>	No	No
Aleyrodidae	<i>Trialeurodes vittatus</i>	No	No
Pseudococcidae	<i>Pseudococcus maritimus</i>	No	No
	<i>Planococcus ficus</i>	Yes	No
Phylloxeridae	<i>Daktulosphaira vitifoliae</i>	Yes	No
Membracidae	<i>Spissistilus festinus</i>	Yes	No

* Yes in field collections

Grapevine Red Blotch Virus (GRBV)

2018-2020 – Field Transmission Exp. – UCB



- Worries over virus titer led to field and lab inoculations
- Focus on TCAH and *Scaphytopius*

Methods

- 48hrs AAP in the field
- Insects brought to lab + placed on clean plants for 48 hrs IAP
- Check vines regularly for symptoms and PCR evaluations for 2 years

Grapevine Red Blotch Virus (GRBV)

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Grapevine Red Blotch Virus (GRBV)

TCAH as Vector – Addressing Grower Concerns

Phylogeny of Geminivirus Coat Protein Sequences and Digital PCR Aid in Identifying *Spissistilus festinus* as a Vector of Grapevine red blotch-associated virus

Brian W. Bahder, Frank G. Zalom, Maya Jayanth, and Mysore R. Sudarshana

First, third, and fourth authors: U.S. Department of Agriculture-Agriculture Research Services, Department of Plant Pathology, University of California, One Shields Ave., Davis 95616; and first and second authors: Department of Entomology and Nematology, University of California, One Shields Ave., Davis 95616.
Accepted for publication 18 April 2016.

ABSTRACT

Bahder, B. W., Zalom, F. G., Jayanth, M., and Sudarshana, M. R. 2016. Phylogeny of geminivirus coat protein sequences and digital PCR aid in identifying *Spissistilus festinus* as a vector of Grapevine red blotch-associated virus. *Phytopathology* 106:1223-1230.

Grapevine red blotch-associated virus (GRBaV) is a single-stranded DNA virus, proposed to be a member of the family *Geminiviridae*, and is associated with grapevine decline and blotch symptoms in North America.

Spissistilus festinus, a geminivirus known to be transmitted by a family that is closely related to leafhoppers, was tested as a vector of GRBaV, hemipteran species within an area where virus spread was suspected were collected. Among the species tested, *Spissistilus festinus* (Hemiptera: Membracidae) was identified as a vector from a grapevine infected with GRBaV in the laboratory.

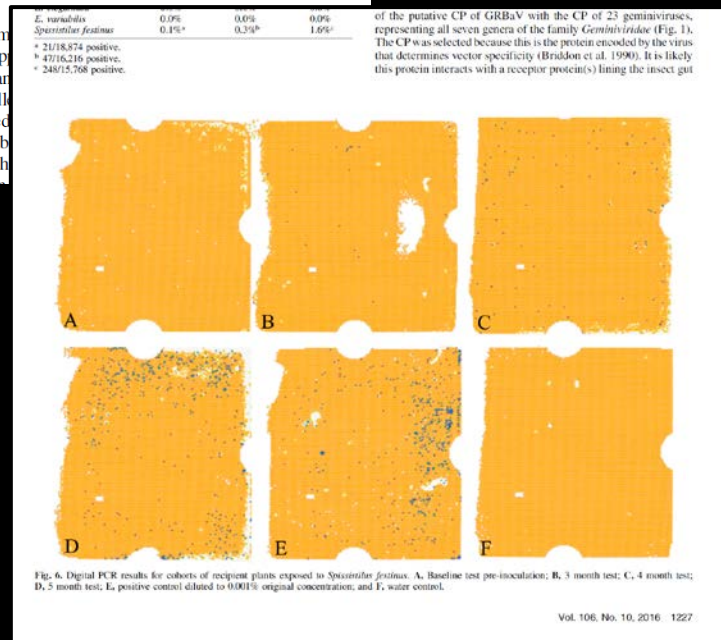


Fig. 3. *Spissistilus festinus* adult A, male and B, female feeding on alfalfa plants.

Grapevine Red Blotch Virus (GRBV)

TCAH Ecology

Distribution

- Widely distributed in US
- Especially southern regions
- Present in North Coast – but not a pest of grapes

Pest Status?

- Legumes – soybean, peanut, alfalfa

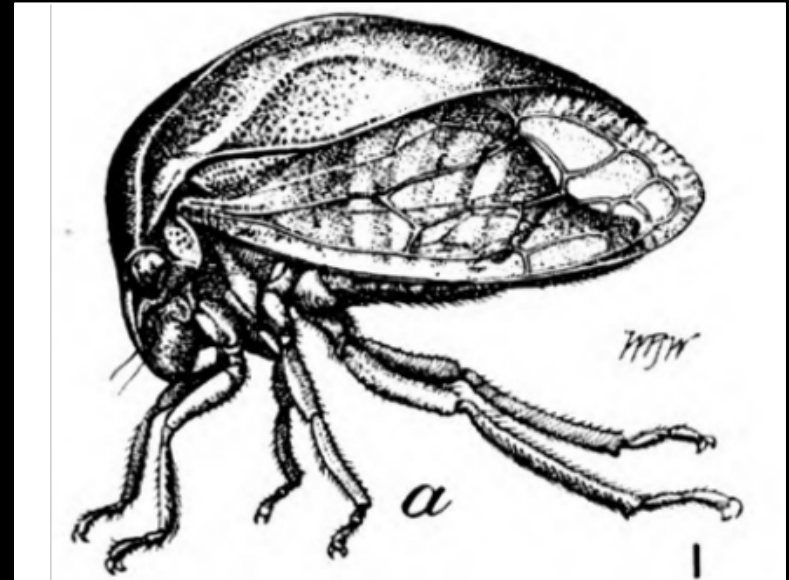
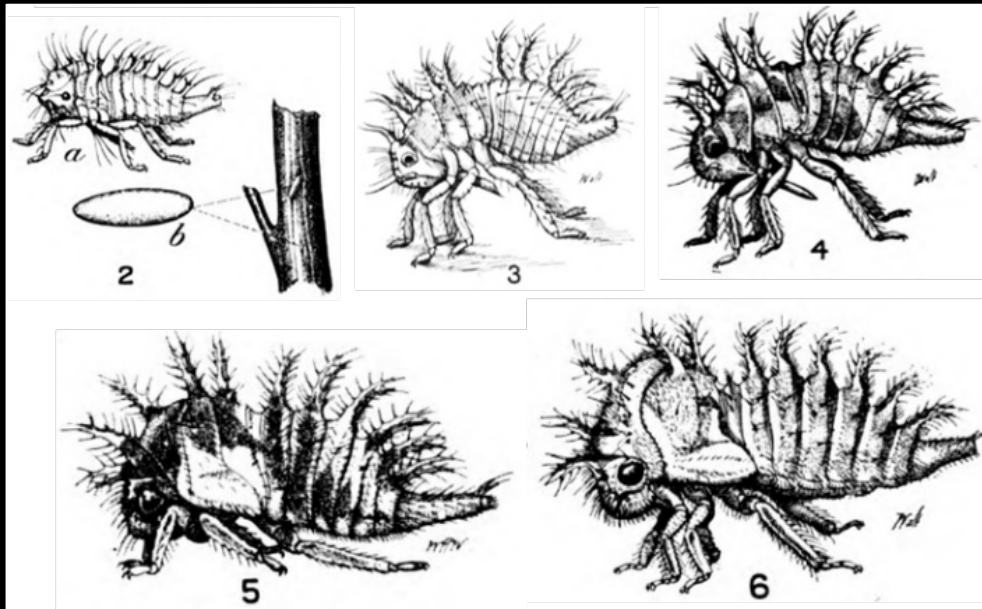


Grapevine Red Blotch Virus (GRBV)

TCAH Ecology

Life Cycle – General

- Adults → Eggs → 5 nymphal instars → Adults
- 2-4 generations/year
- Preference for legumes – but can develop on other species
- Overwinter in adult stage in sheltered areas

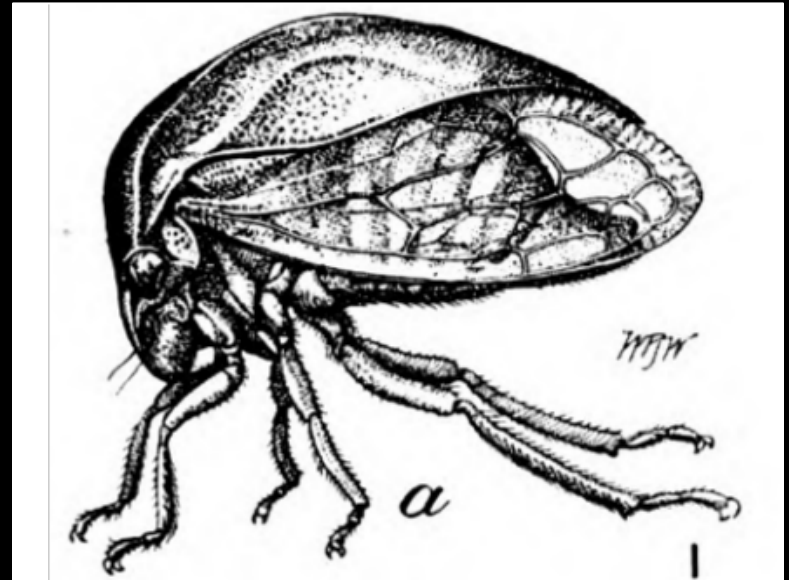
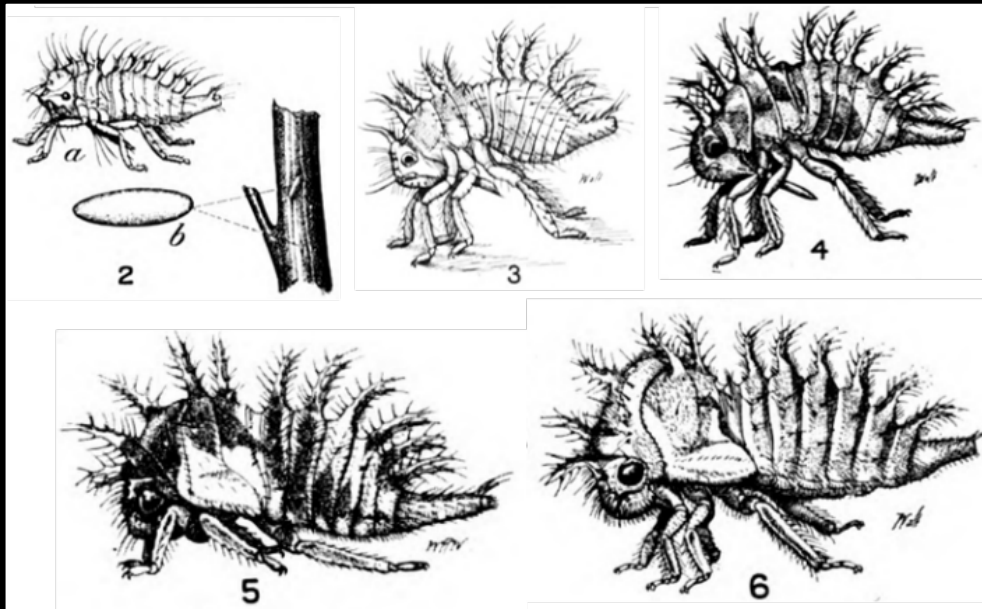


Grapevine Red Blotch Virus (GRBV)

TCAH Ecology

Life Cycle – Vineyards

- Adults appear on ground covers in the spring
- Feed/reproduce on ground covers (likely legumes)
- Petiole girdling later in season
- 2 generations/year

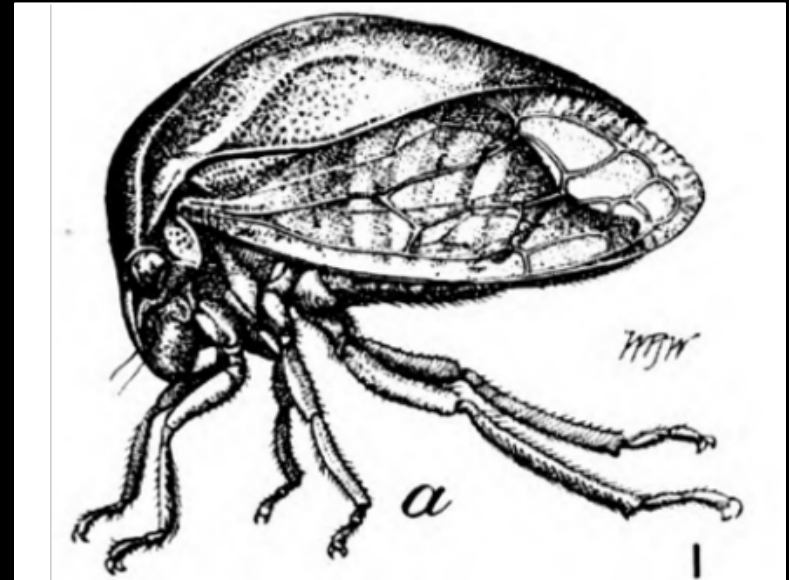
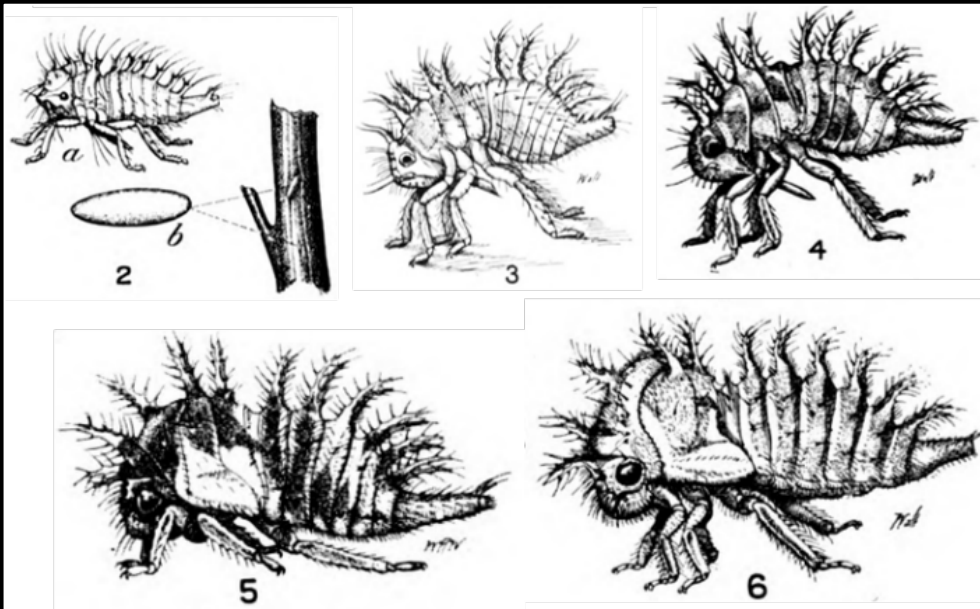


Grapevine Red Blotch Virus (GRBV)

TCAH Ecology

Key Questions

- When and how much activity in vine canopy?
- Overwinter in natural habitats? Vineyard edge activity?



Grapevine Red Blotch Virus (GRBV)

TCAH Ecology

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- When and how much activity in vine canopy?
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Fig. 1. Map illustrating two red blotch disease epidemics in a 1.5-ha Cabernet Sauvignon and a 2.0-ha Cabernet franc vineyard within 10 years post-planting in California, and the proximity of these vineyards to the Napa River and riparian habitat. In the Cabernet Sauvignon vineyard, red overlay indicates diseased vines in 2017 at the onset of the study and orange indicates new GRBV infections in 2018. In the Cabernet franc vineyard, red overlay indicates initial disease incidence recorded previously for 2014 to 2016 (Cieniewicz et al. 2017a). Purple and orange indicate new infections in 2017 and 2018, respectively. White grids indicate areas where insect survey was conducted in 2015 and 2016 in the Cabernet franc (Cieniewicz et al. 2018b) and in 2017 and 2018 in the Cabernet Sauvignon (this study).

Grapevine Red Blotch Virus (GRBV)

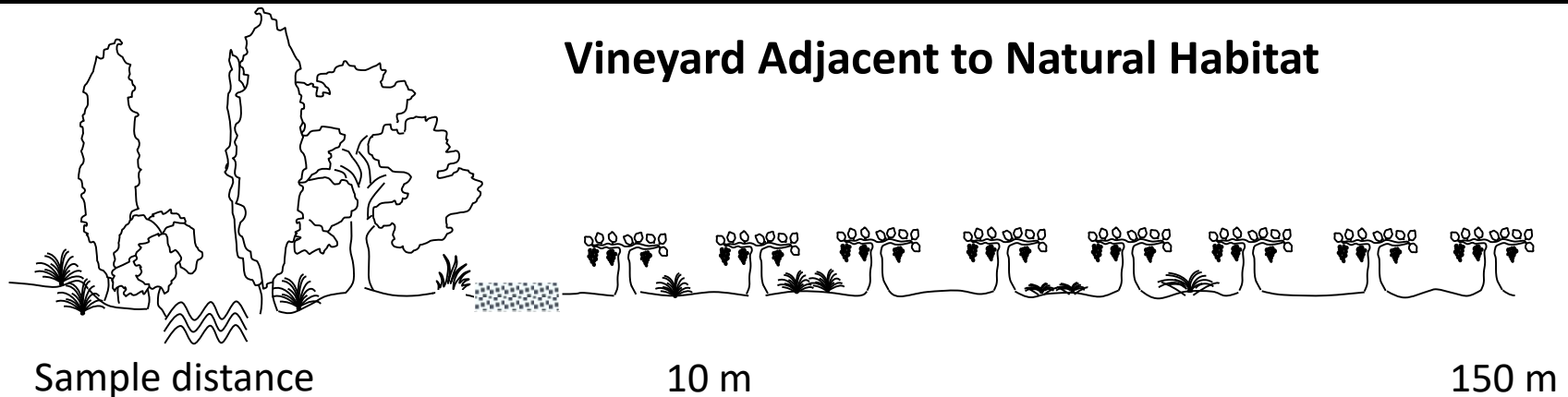
TCAH Ecology

Key Questions

- When and how much activity in vine canopy?
- Overwinter in natural habitats? Vineyard edge activity?

Transect Study (2017-2018)

- 5 vineyard sites
- Sampled 2x/month March – November (2017-2018)



Grapevine Red Blotch Virus (GRBV)

TCAH Transect Study

Sampling Methods

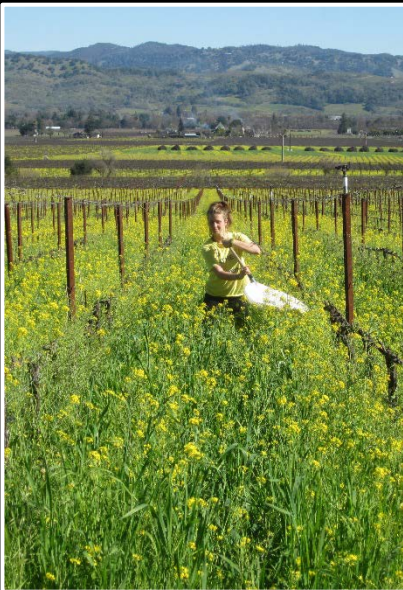


Grapevine Red Blotch Virus (GRBV)

TCAH Transect Study Sampling Methods

Samples at Vineyard Edge and Interior

- Ground covers – Yellow sticky-traps (Mar-Nov)
- Vine Canopy – Yellow sticky-traps (Mar-Nov)
- Petiole girdling – 1-2x/month (June-Oct)



Grapevine Red Blotch Virus (GRBV)

TCAH Transect Study

Focal Organisms



Spissistilus festinus

Three-cornered Alfalfa Hopper

“TCAH”



Scaphytopius spp.

Sharp-nosed Leafhopper

“Scaphy”



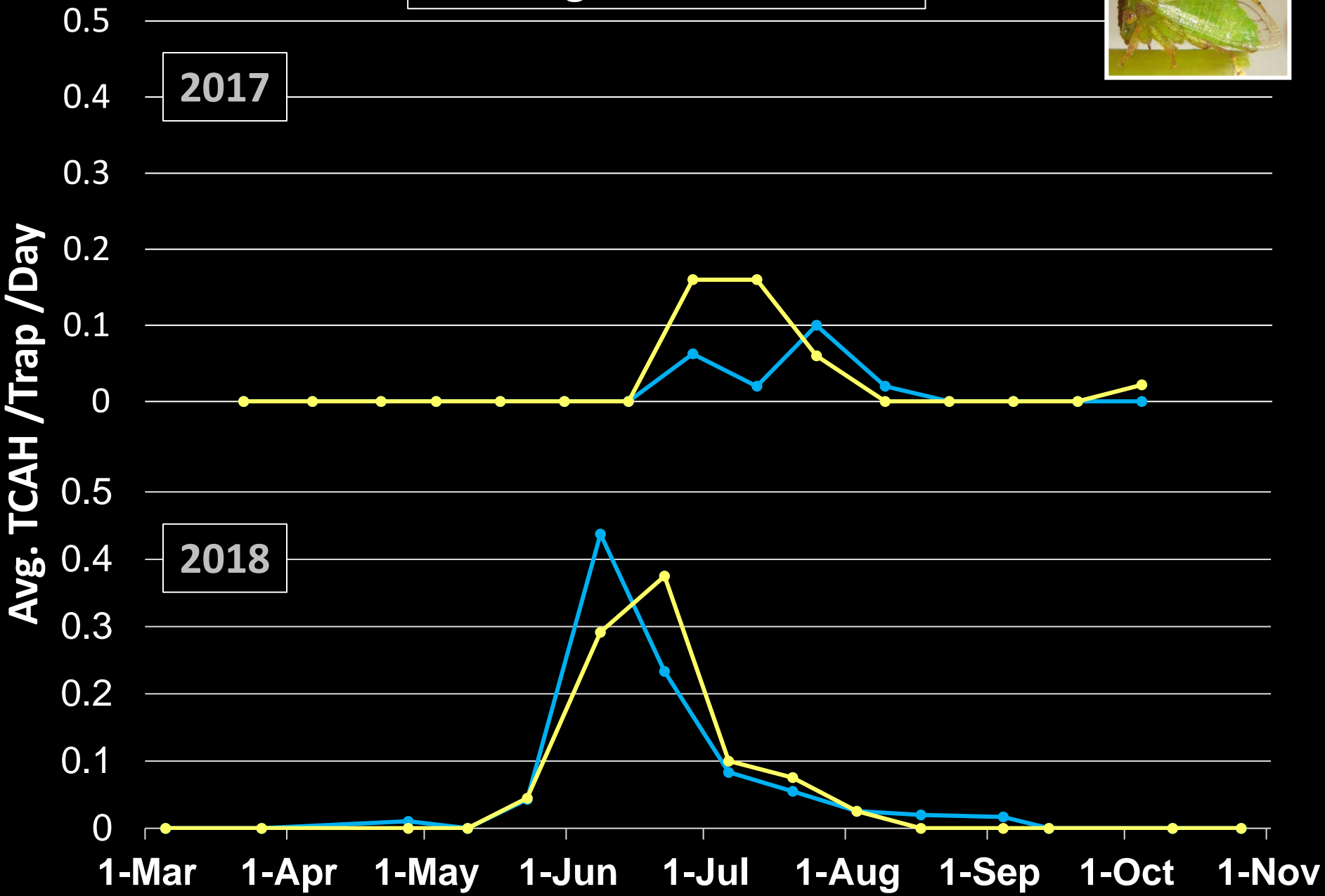
Spissistilus festinus

Three-cornered Alfalfa Hopper

“TCAH”

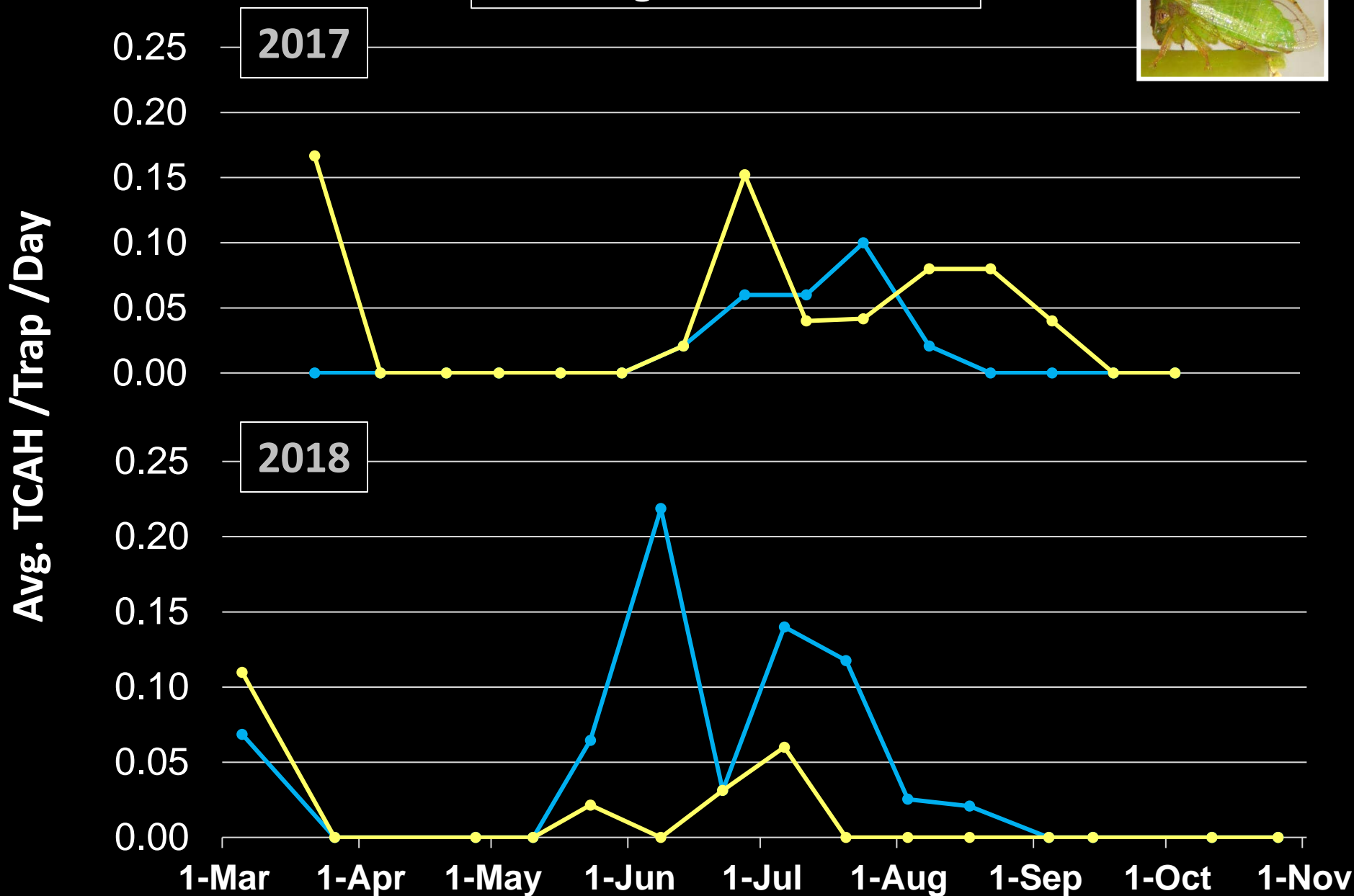
Vine Canopy – No differences between edge/interior

Edge Interior

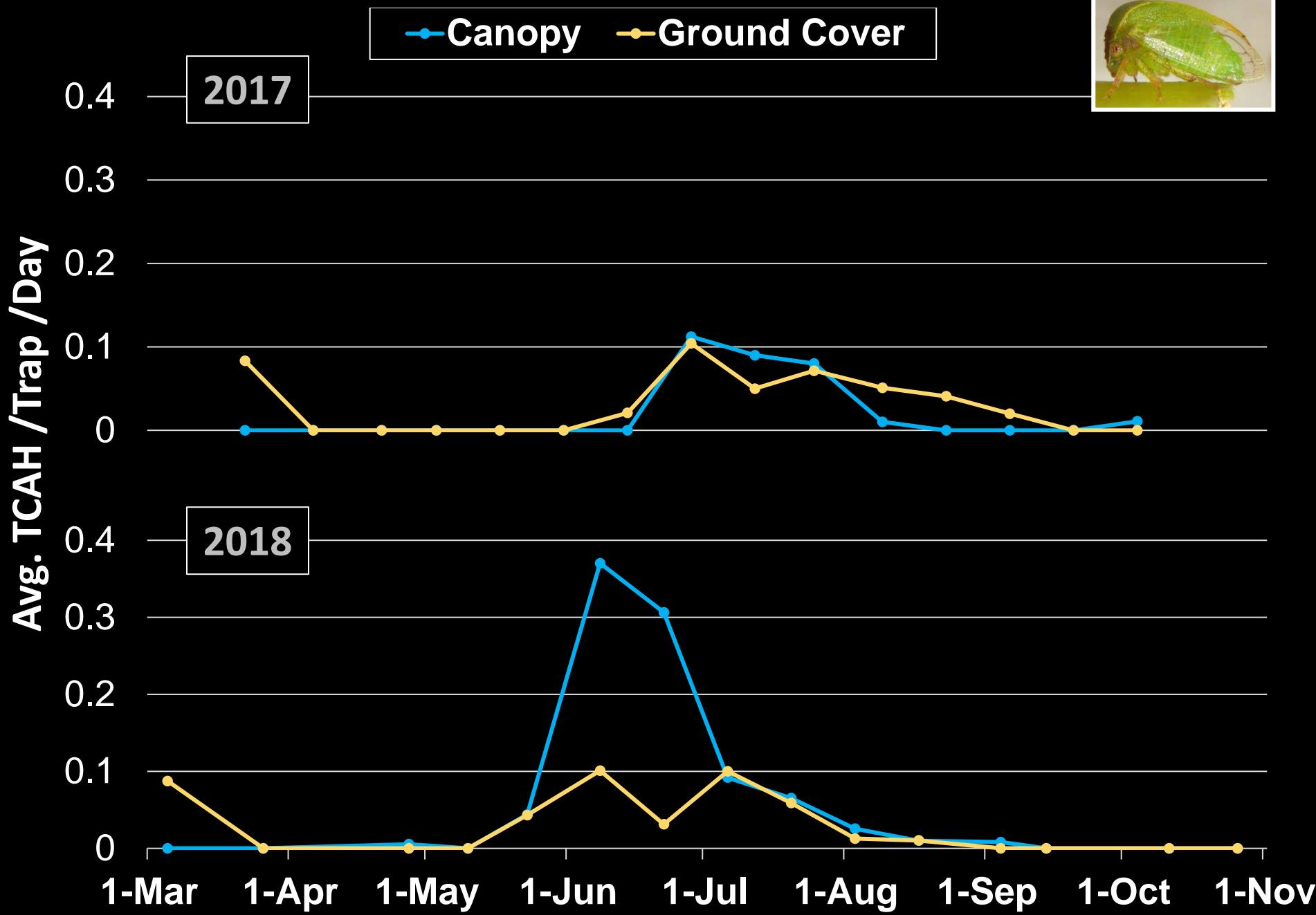


Ground Covers – No differences between edge/interior

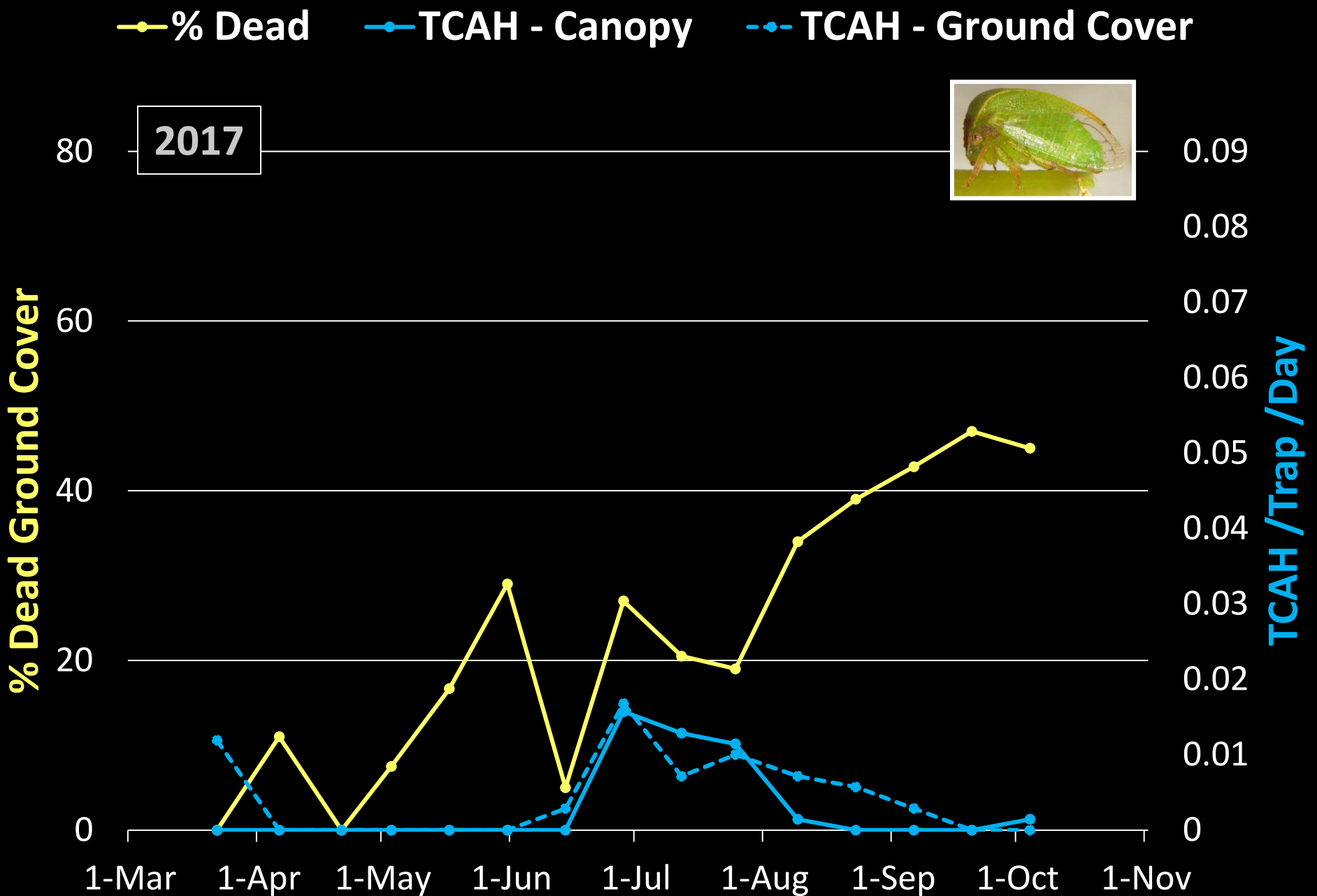
Edge Interior



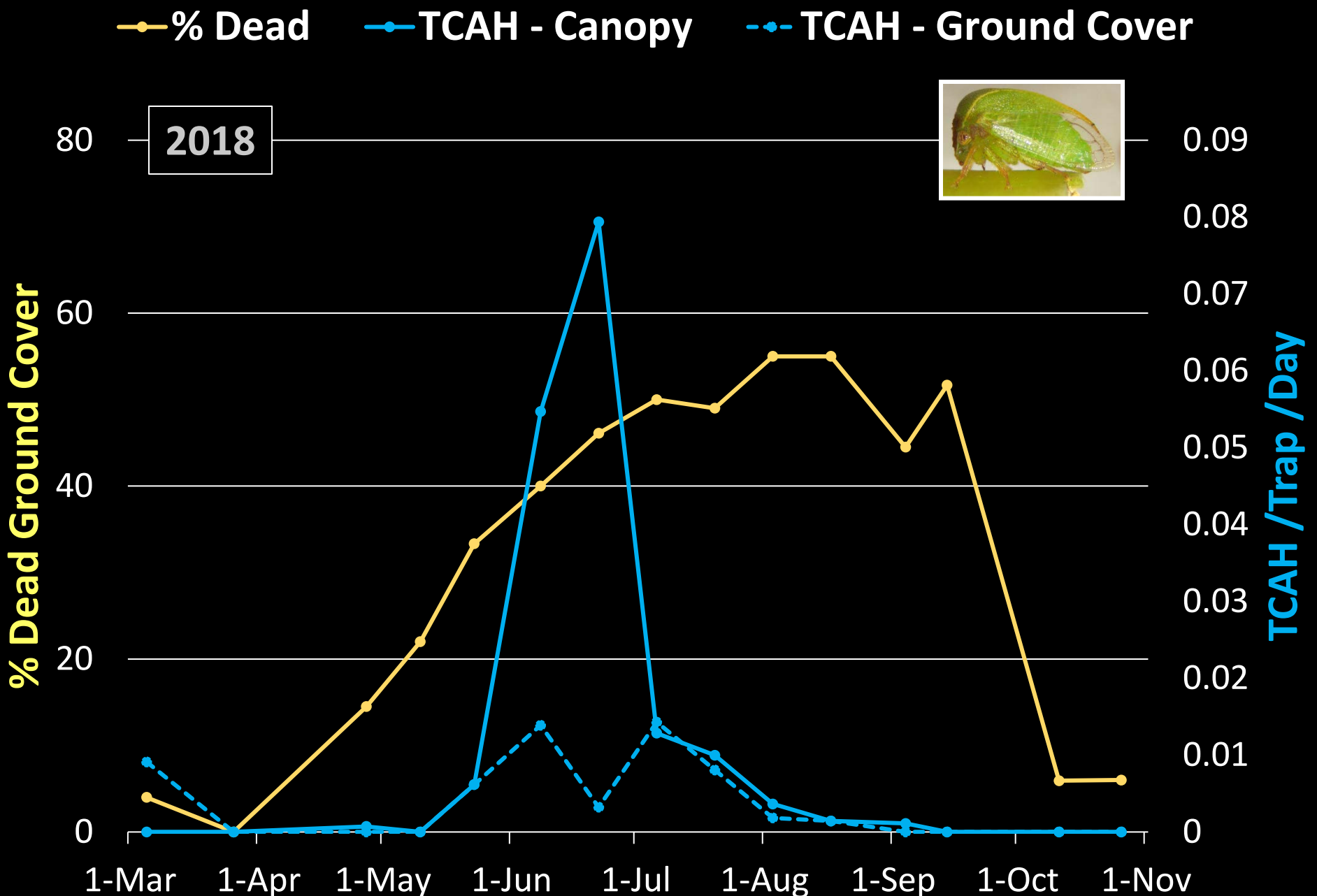
No consistent differences between canopy + ground covers



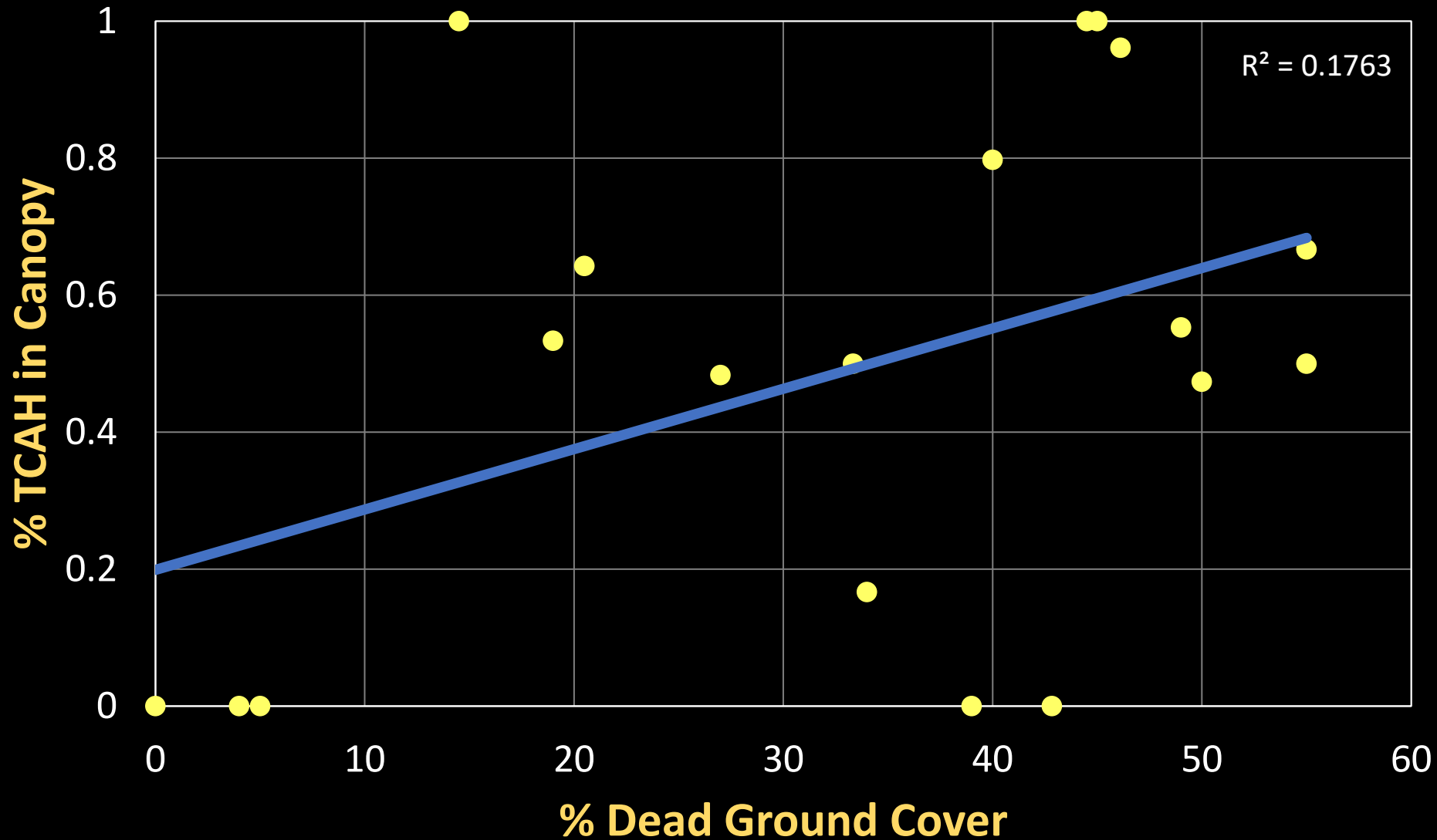
Ground cover quality decrease leads to vine canopy increase



Ground cover quality decrease leads to vine canopy increase



Ground cover quality decrease leads to vine canopy increase

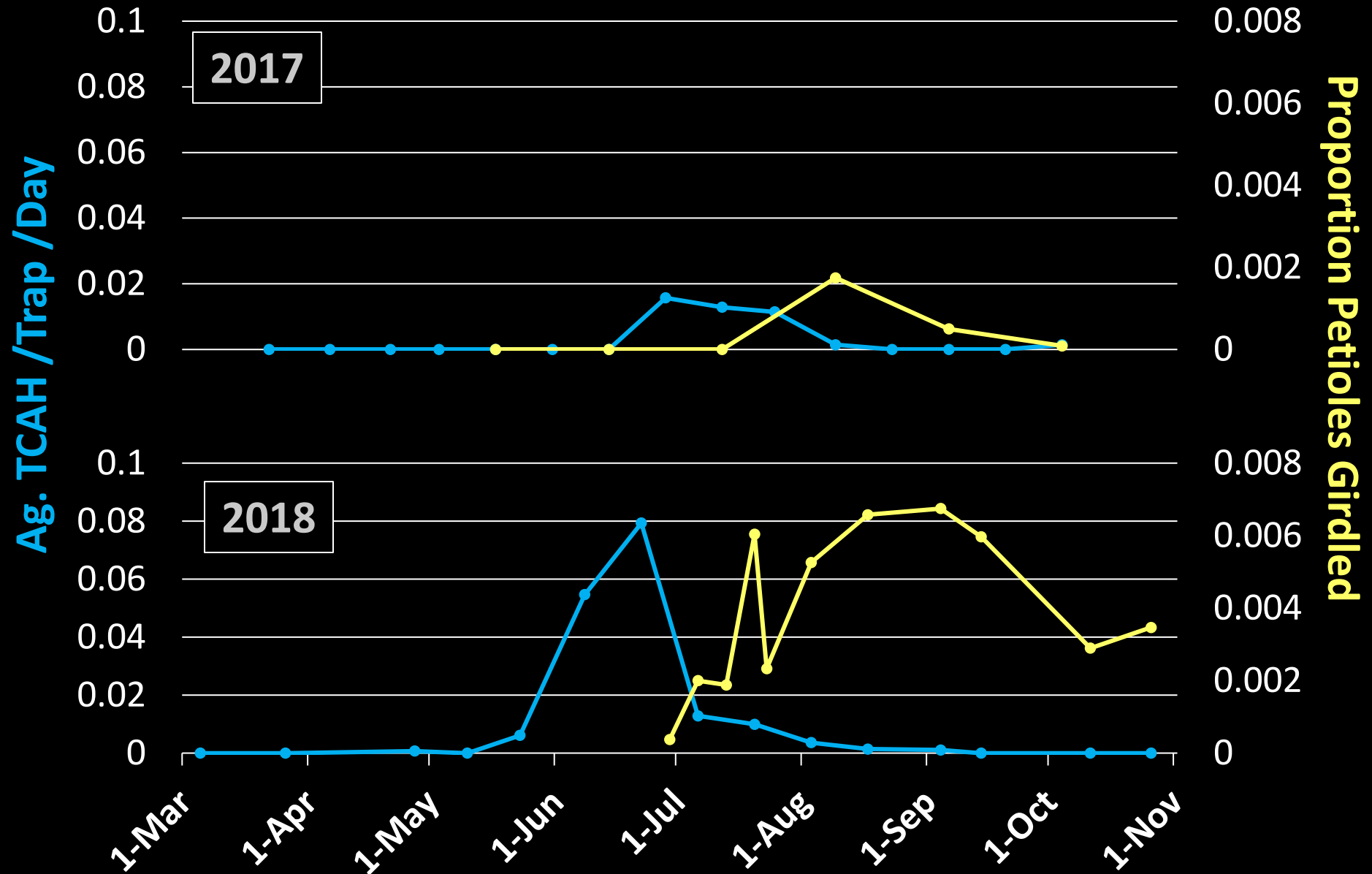


TCAH activity in vine canopy followed by girdling



TCAH - Canopy

Petiole Girdling





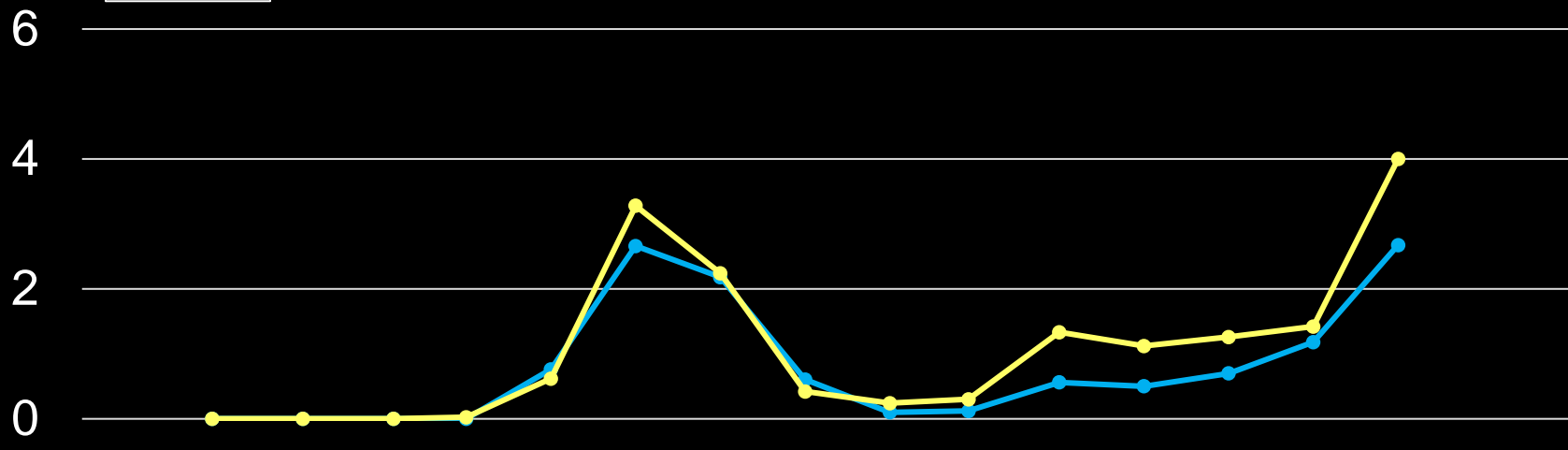
Scaphytopius sp.
Sharp-nosed Leafhopper
“Scaphy”

Vine Canopy – Densities slightly elevated at interior

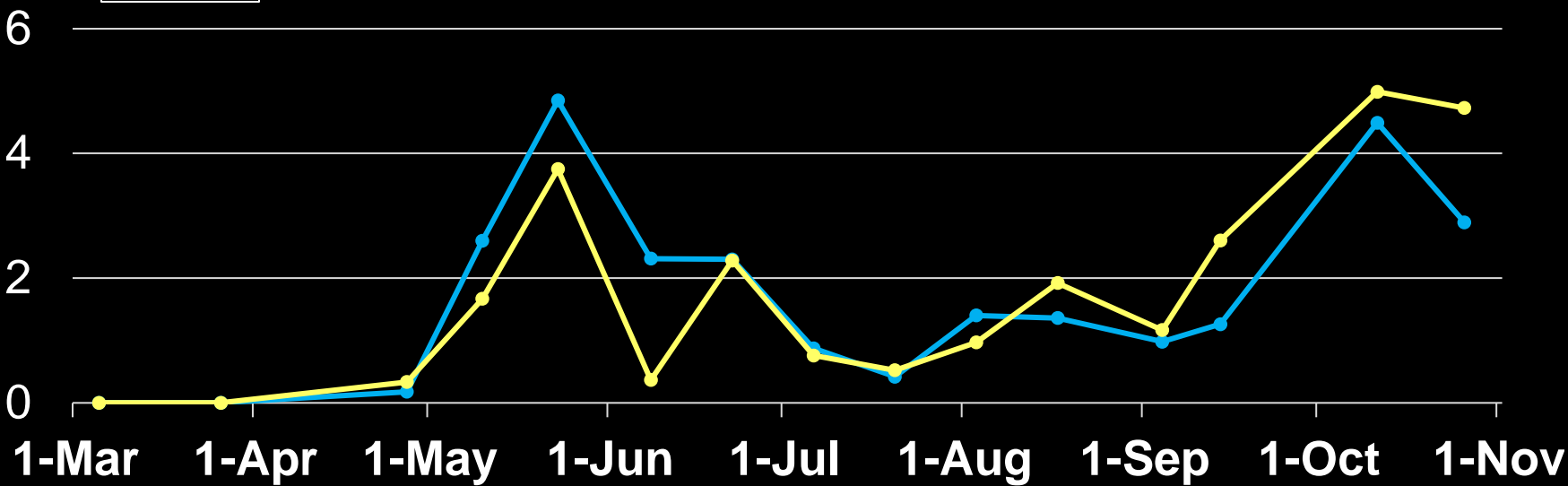
—●— Edge —●— Interior



2017



2018



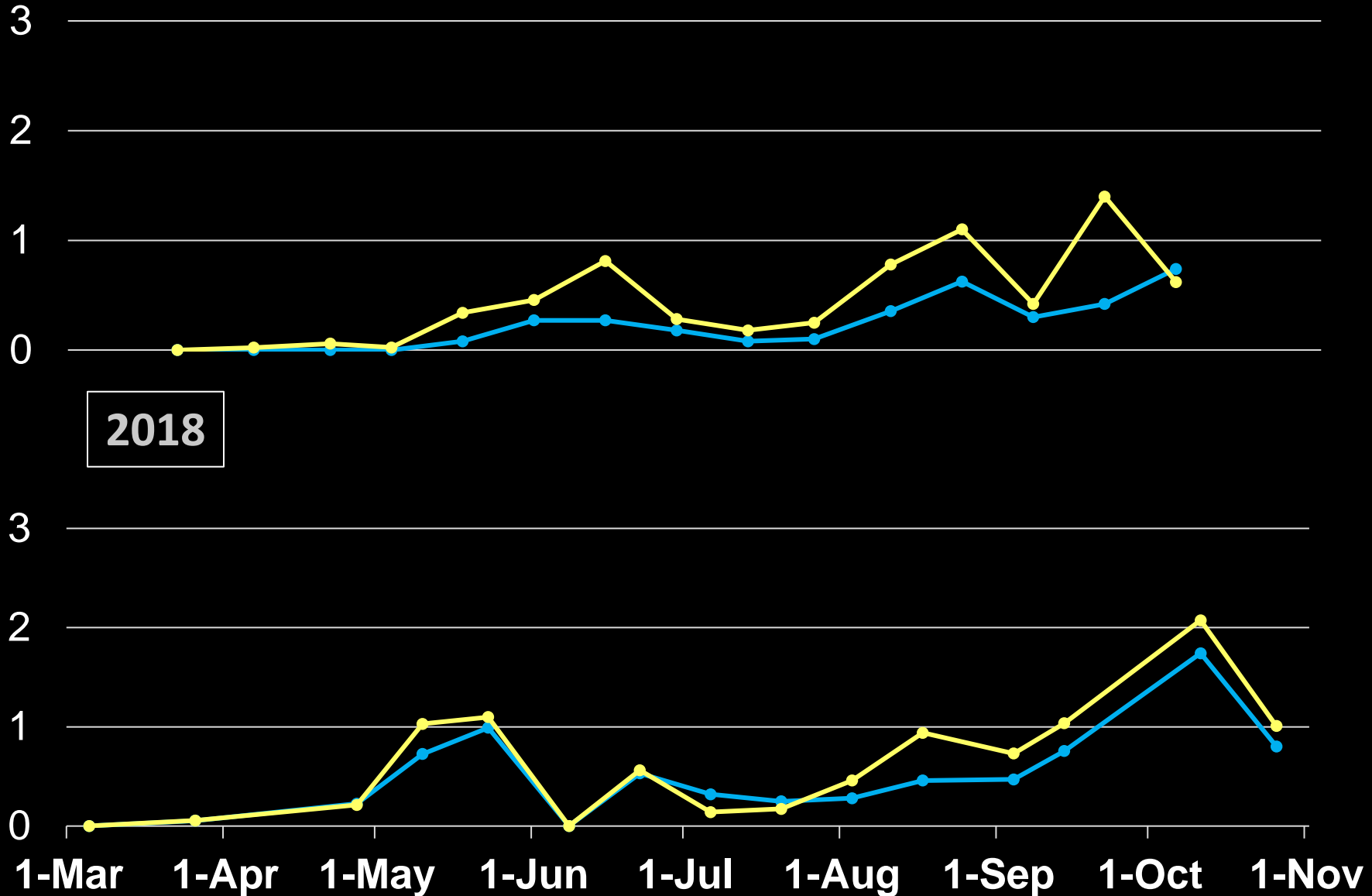
Ground Cover – Densities slightly elevated at interior

2017

Edge Interior



Avg. Scaphy / Trap / Week



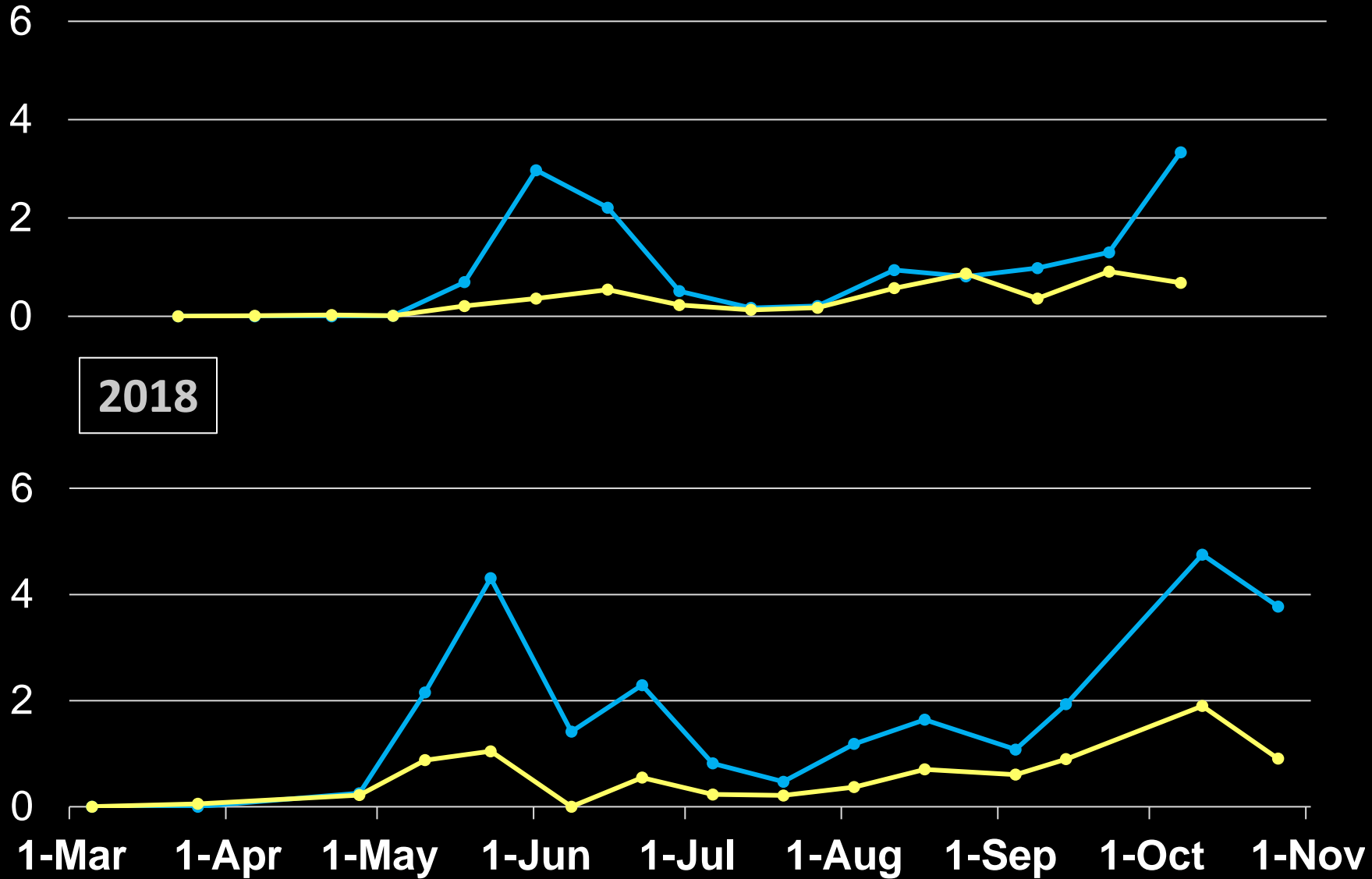
Consistently higher densities in the vine canopy

2017

Canopy Ground Cover



Avg. Scaphy / Trap / Week

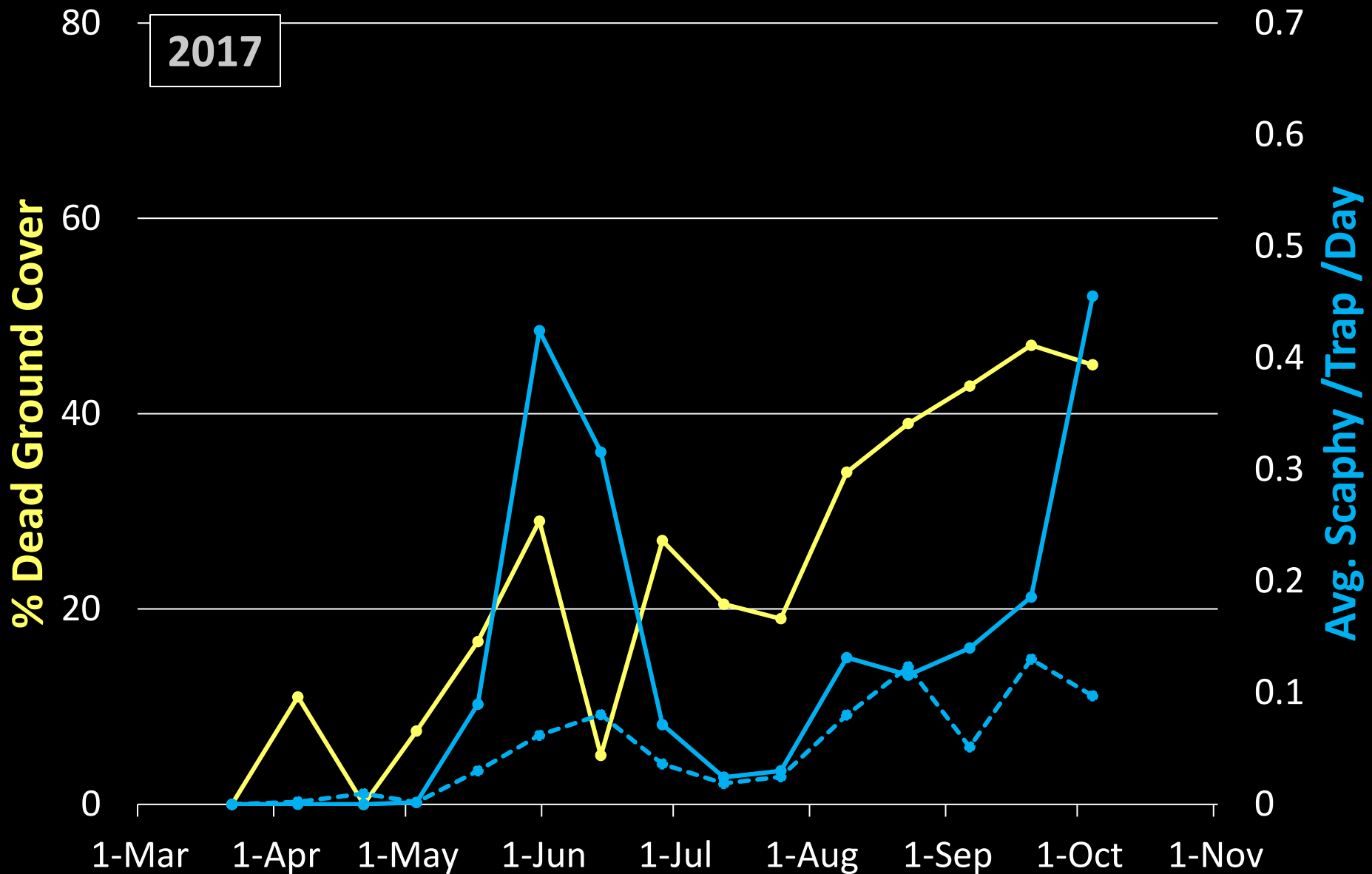


No response to ground cover quality



—●— % Dead —●— Scaphy - Canopy - -●- - Scaphy - Lower

2017

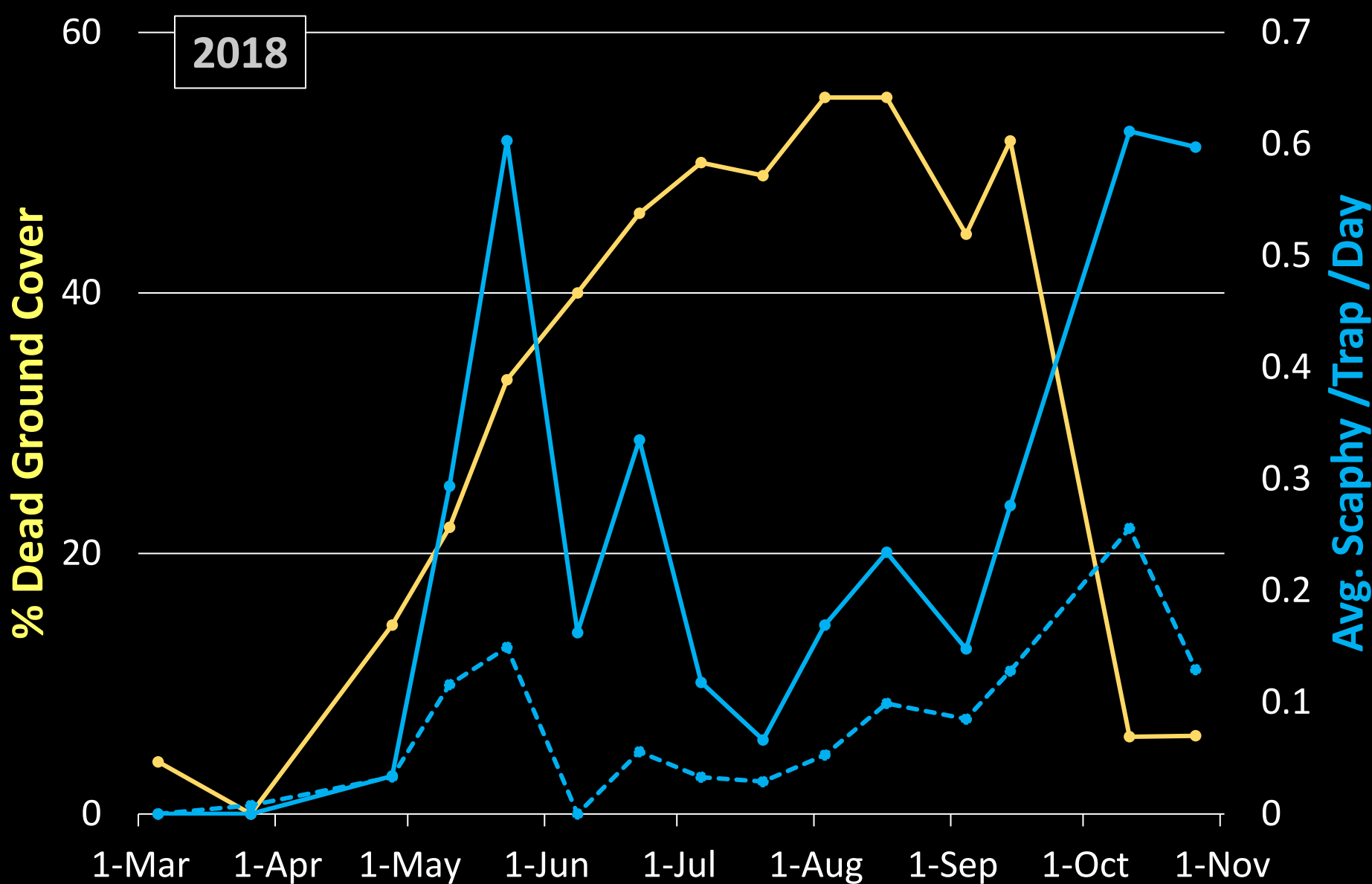


No response to ground cover quality

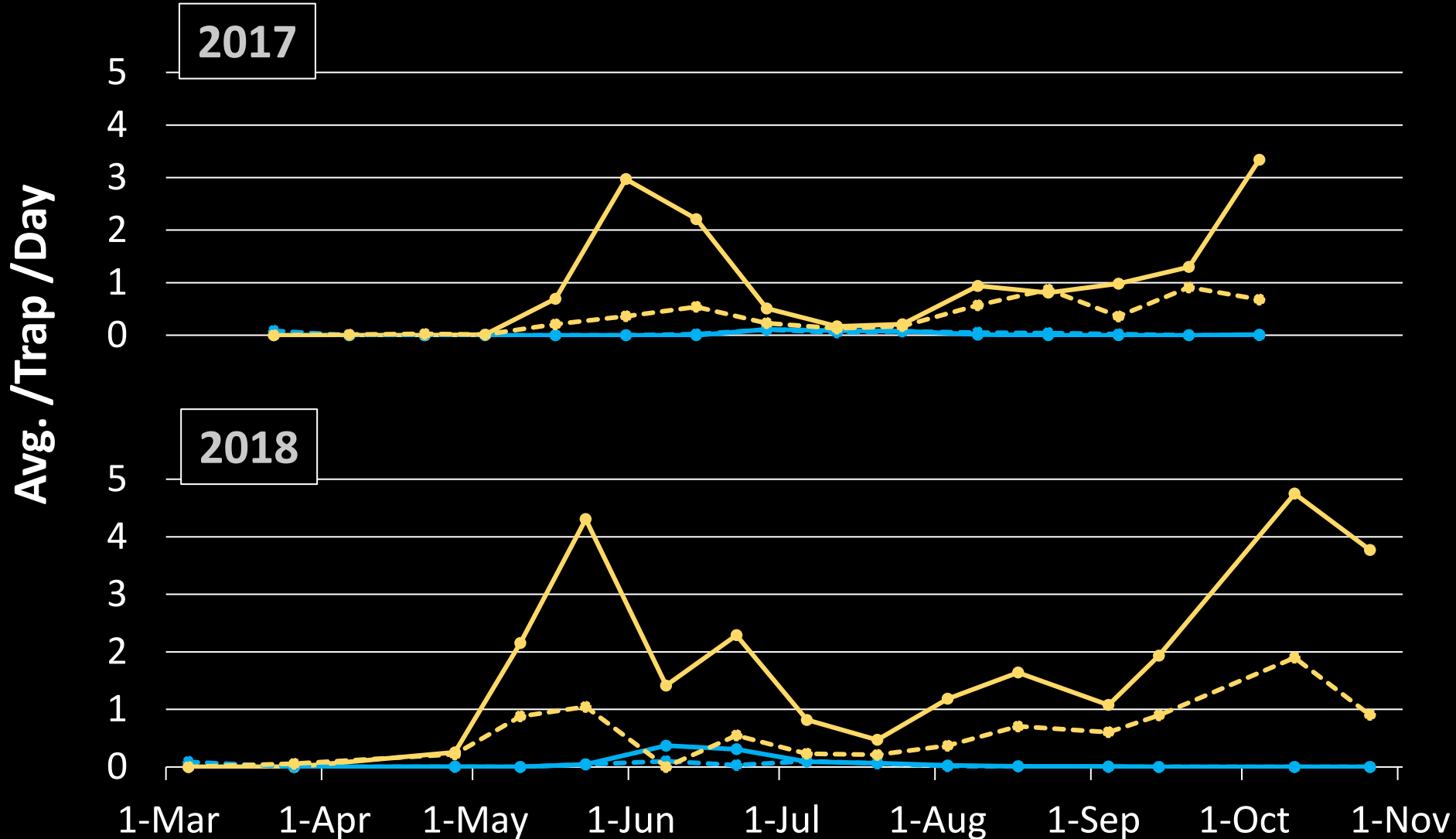


—●— % Dead —●— Scaphy - Canopy - - -●- - Scaphy - Lower

2018



Much higher densities of *Scaphytopius* relative to TCAH



Grapevine Red Blotch Virus (GRBV)

Summary and Conclusions

Grapevine Red Blotch Virus (GRBV)

Summary and Conclusions

Surveyed potential plant reservoirs and insect vectors

- *Vitis* is sole positive plant host
- Cicadellids, TCAH and a few others tested positive
- Some species seem more promising than others



Scaphytopius spp.



Colladonus montanus



Spissistilus festinus



Colladonus coquilletti

Grapevine Red Blotch Virus (GRBV)

Summary and Conclusions

Transmission experiments with common vineyard insects

- Lots of experiments, lots of insects
- Some acquisition, but no transmission observed
- Rules out a lot of the most common insects

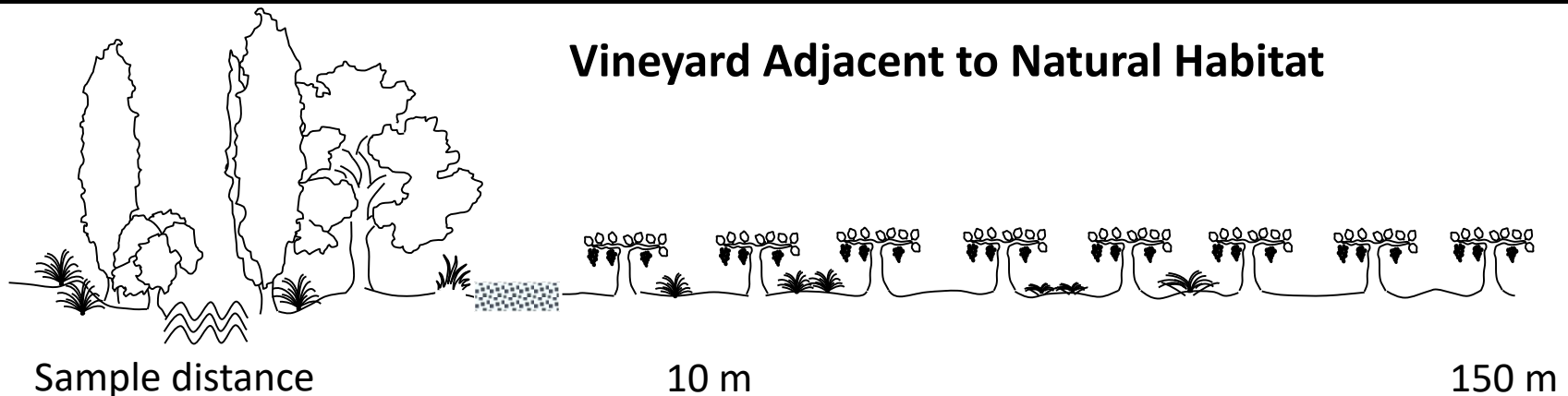
Family	Species Tested	Transmits
Cicadellidae	<i>Erythroneura elegantula</i>	No
	<i>Erythroneura variabilis</i>	No
	<i>Erythroneura ziczac</i>	No
	<i>Graphocephala atropunctata</i>	No
Aleyrodidae	<i>Trialeurodes vittatus</i>	No
Pseudococcidae	<i>Pseudococcus maritimus</i>	No
	<i>Planococcus ficus</i>	No
Phylloxeridae	<i>Daktulosphaira vitifoliae</i>	No
Membracidae	<i>Spissistilus festinus</i>	No

Grapevine Red Blotch Virus (GRBV)

Summary and Conclusions

TCAH Seasonal Ecology

- Confirm 2 adult peaks/year
- Natural habitats don't seem to be a major source of TCAH
- Leguminous ground covers are important for TCAH
- Vine canopy activity Jun.-Aug. followed by girdling
- Canopy activity related to changes in ground cover quality

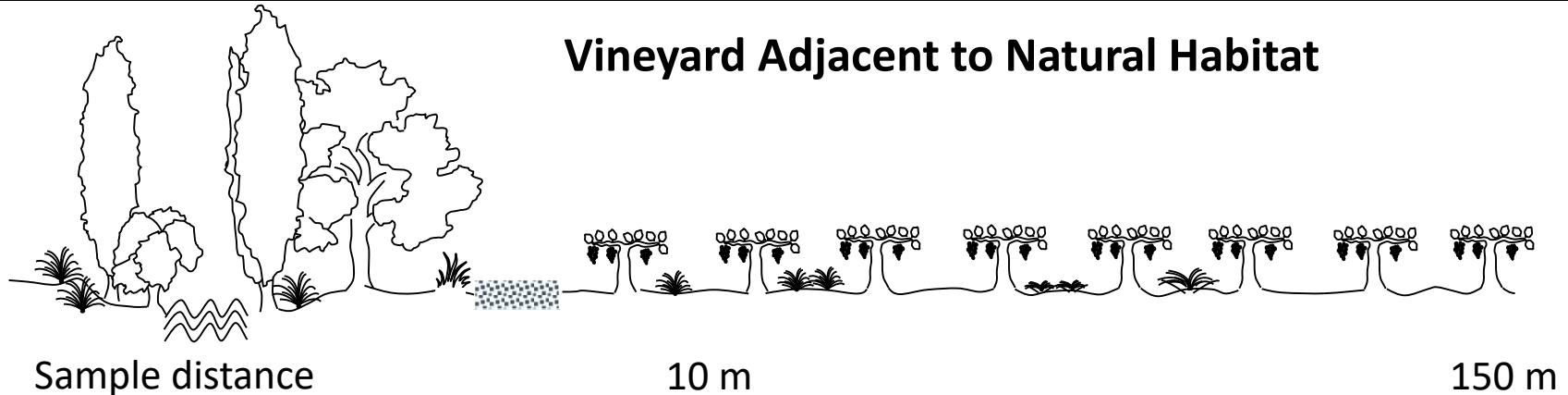


Grapevine Red Blotch Virus (GRBV)

Summary and Conclusions

What about *Scaphytopius*?

- Promising candidate vector
- Higher populations and affinity for grape vines
- Transmission studies currently in progress



Grapevine Red Blotch Virus (GRBV)

TCAH Management?

- **No treatment thresholds or guidelines**
 - **No support for sprays**
 - **No support for ground cover management**
 - **Tradeoffs = soil quality, compaction, erosion**
- **Circumstantial evidence unconvincing**
 - **Generally low abundance**
 - **Low affinity for grape vine**
 - **Transmission efficiency unclear**
 - **Field transmission unconfirmed**
- **Broader candidate insect testing still incomplete**



Grapevine Red Blotch Virus (GRBV)



Is Scaphytopius a Vector?

- We don't know yet!
 - Transmission unconfirmed
 - Experiments are in progress
 - Even if transmits, efficiency is still key
- Circumstantial evidence more convincing
 - Much higher densities
 - Strong affinity for grape vines
 - Paso Robles surveys – no TCAH, lots of Scaphy

Grapevine Red Blotch Virus (GRBV)

Research in 2020 and Beyond

Daane / Wilson Research Group

1. Test more novel candidate vectors
2. Field transmission studies
3. Identify TCAH overwintering sites
4. TCAH transmission efficiency
5. Manipulate ground covers



Scaphytopius spp.



Caladonus coquilletti



Colladonus montanus

Thank You!

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McCalla, Kristen Flores, Alonzo Ledesma, Jessica Maccaro

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