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AGENDA

California Olive Committee

Research Subcommittee Meeting
ZOOM/Conference Call

NOVEMBER 13, 2024
3:30 PM

Zoom Meeting
https://us02web.zoom.us/j/82779827437

Telephone:
+1669-900-6833
Meeting ID: 827 7982 7437

I. **Call to Order**
   a. Roll Call page 2
   b. Research Subcommittee Chairman's comments
   c. Approval of August 7, 2024 Minutes (action item) page 3

II. **Guest Speaker-Suterra**
   a. Olive Fruit Fly Attract and Kill Trap page 9

III. **Discussion and Review of 2024 Projects** page 10

IV. **Presentation of 2025 Proposals** page 44

V. **Approval of Authority to the Executive Director and Chairman to approve No-Cost Extensions** (action item) page 199

VI. **Approval of 2025 Budget** (action item) page 203

VII. **Approval of Authority to the Executive Director and Chairman for Inter-Item Transfer of the Research Subcommittee Budget** (action item) page 205

VIII. **Other Business**

IX. **Adjournment**

2023-2025 Research Subcommittee:

Producer Members:

Carolina Burreson
Michael Silveira
Kevin Neeley
Andy Weinrich
Michael Stokes
Vito DeLeonardis
Giulio Zavolta
Pat Ricchiuti
Galen Pfeiffer
Mark Heuer

Handler Members:

Dennis Burreson-Chairman
John Pieretti
Tomas Masanes Autard
Julia Tinsley
Phil Quigley
Vacant

California Olive Committee

Research Subcommittee Meeting

AUGUST 7, 2024
9:00 am

Zoom/Conference Call
http://US02web.zoom.us/j/88631846244

MINUTES

I. CALL TO ORDER

Chairman Dennis Burreson called the meeting of the Research Subcommittee to order at 9:02 a.m. and the following members were present:

Members	**Affiliation:**
Carolina Burreson	Producer
Michael Silveira	Producer
Kevin Neeley	Producer
Andy Weinrich	Producer
Michael Stokes	Producer
Giulio Zavolta	Producer
Pat Ricchiuti	Producer
Galen Pfeiffer	Producer
Dennis Burreson - Chairman	Handler
John Pieretti	Handler
Tomas Masanes Autard	Handler
Julia Tinsley	Handler
Phil Quigley	Handler

STAFF	
Todd Sanders	California Olive Committee
Elise Oliver	California Olive Committee
Jackie Nakashian	California Olive Committee
Janette Ramos	California Olive Committee

With the appropriate number of members from producers and handlers in, a quorum was established.

- **MOVED by RICCHIUTI, duly seconded by PFEIFFER and carried THAT the minutes for December 7, 2023, be approved as presented. (MOTION 8-7-24 #1)**

Chairman's Comments:

Chairman Dennis Burreson addressed the committee, and thanked everyone for attending and continuing to work through various research projects.

II. REVIEW OF 2023 RESEARCH FINAL PROJECTS

2023 RESEARCH PROJECTS FOR THE CALIFORNIA OLIVE COMMITTEE

- Projects in red had No Cost Extensions and are now complete. Final Reports can be found in the following pages of the packet.

Researcher	Project	Amount	No Cost Extension
Dr. Jim Adaskaveg	Management of Foliar Diseases-A. Olive Knot and B. Evaluation of New Fungicides For Control of Olive Leaf Spot	$10,000	
Dr. Jim Adaskaveg	Epidemiology and Management of Olive Knot Caused by Pseudomonas Savastanoi pv. Savastanoi	$21,150	
Carol Lovatt and Elizabeth Fichtner	Integrating Alternate Bearing Mitigation Strategies in a Commercial Table Olive Orchard	$36,511	6/30/2024
Georgia Drakakaki and Louise Ferguson	Evaluating Accede for Ability to Decrease 'Manzanillo' Fruit Detachment Force and Increase Efficiency of Commercial Trunk Shaking and Experimental Canopy of Contact Harvesters	$115,186	6/30/2024
Jim Stewart	Southern San Joaquin Valley Olive Fruit Fly Monitoring	$11,000	
Ernie Simpson	Sacramento Valley Olive Fruit Monitor Project	$9,250	
	Contingency Fund	$0	
	Total	**$203,097**	

III. UPDATE ON CA SPECIALTTY CROP BLOCK GRANT PROJECTS

The 2025 Specialty Crop Block Grant Program will be opening its application submission period in the Fall; as a result, the COC is currently looking for potential grant ideas from the Committee.

- Funding Areas include:
  - Market Enhancement
  - Access, Education, and Training
  - Research

IV. PRODUCT REGISTRATION UPDATE

GWSS:

COC staff is currently looking into products registered to combat GWSS, as GWSS is a vector for Xf.

Staff has reached out to Valent regarding their products PyGanic and Danitol.

Product	Active Ingredient	Registrant	Status
Actara	Thiamethoxam	Syngenta	Not registered on olives
Assail 70 WP	Acetamiprid	United Phosphorous, Inc.	Not registered on olives
Assail 30 SG	Acetamiprid	United Phosphorous, Inc.	Not registered on olives
Baythroid XL	Beta-cyfluthrin	Bayer	Not registered on olives
Tombstone	Beta-cyfluthrin	Loveland Products, Inc.	Not registered on olives
Mustang	Zeta-cypermethrin	FMC Corporation	Not registered on olives
Danitol 2.4 EC	Fenpropathrin	Valent	Registered on Olives for Olive Fruit Fly, etc.
Lannate SP	Methomyl	TKI (Novasource)	Not registered on olives or in CA
Platinum 75 SG	Thiamethoxam	Syngenta	Not registered on olives
Sevin XLR Plus	Carbaryl	TKI (Novasource)	Registered on Olives for Black Scale
Sivanto Prime	Flupyradifurone	Bayer	Not registered on olives
PyGanic Crop Protection EC 5.0	Pyrethrins	Valent	Registered on olives for GWSS

Accede:

Valent will be pitching in $8,500 towards the research project currently underway!

COC Research Subcommittee Chairman, COC Chairman, and COC staff participated on a IR-4 call expressing support for Accede to become an IR-4 program project. We are waiting to hear back from the IR-4 program.

Syllit and Polyoxin-D (Ph-D):

UPL has submitted applications to EPA. Syllit and Ph-D will be used for managing Peacock spot and Syllit will be used for managing olive knot in mixtures with copper and other new bactericides such as Kasumi

Approvals are pending at the EPA level until the Endangered Species Act (ESA) review is Completed.

V. DISCUSSION AND APPROVAL OF 2025 RESEARCH PRIORITIES

- Each year the Research Subcommittee sets priorities of research they would like executed on their behalf for the following year. These efforts are to fund more specific and calculated research to enhance the benefits to the industry. Once the priorities are set, they are provided to the University of California liaisons to request proposals from researchers. Additionally, priorities are distributed to land grant universities across the nation and to private research facilities. Proposals will be reviewed for funding in November by the subcommittee.
- On the following page are the 2024 Research Priorities.

2024 RESEARCH PROJECTS FOR THE CALIFORNIA OLIVE COMMITTEE

Researcher	Project	Amount
Dr. Jim Adaskaveg*	Management of Foliar Diseases-A. Olive Knot and B. Evaluation of New Fungicides for control of olive leaf spot	$13,715
Dr. Jim Adaskaveg*	Epidemiology and Management of Olive Knot Caused by Pseudomonas Savastanoi pv. Savastanoi	$21,150
Carol Lovatt and Elizabeth Fichtner	Integrating Alternate Bearing Mitigation Strategies in a Commercial Table Olive Orchard	$33,825
Georgia Drakakaki and Becky Wheeler-Dykes	Evaluation of effects of Accede (ACC), tree architecture, and harvester type on enhancing horticultural maturity and abscission zone development and commercial trunk shaking efficiency in table olives	$131,380
Rodrigo Almeida	Survey of Xylella fastidiosa diversity within California olive trees	$29,752
Jim Stewart	Southern San Joaquin Valley Olive Fruit Fly Monitoring	$12,000
Ernie Simpson	Sacramento Valley Olive Fruit Monitor Project	$9,250
	Total:	**$251,072**

*Projects are co-funded by the California Olive Oil Commission.

California Olive Committee Research Priorities for 2024

- Olive Fruit Fly Trapping
- OFF management techniques with an emphasis on examining new traps that attract flies. Also, OFF management techniques focused on needing a membrane and a new delivery system.
- Management of Olive Knot with an emphasis on developing new methods of control via soil applications. Management of Olive Knot focused on systemic rather than topical treatments.
- Management of Peacock Spot
- Evaluation of drone technology and satellite mapping pertaining to moisture evaluation and crop load estimates (approach the Ag Tech companies that we have spoken with Aerorobotics and other, Matt Koball
- Mechanical harvesting on existing and new high density orchards
- Development of Loosening Agents

- Olive DNA evaluation to distinguish between different varieties in the market place
- Chemical control of Glassy-winged sharpshooter/Leaf Hoppers
- Determine costs of preventative measures relating to the spread of xylella-fastidiosa (xf)
- Pollination assist techniques focused on Manzanillo /Field measurement tool to determine optimal pollination timing
- Investigation of Urea as a thinning agent. What is the cost and optimal application rate? Where it is currently being used?
- Field measurement tool to determine optimal pollination timing
- Determine what research has already been conducted regarding sterilization of OFF
- Attract and Kill trap for Olive Fruit Fly
- Black Scale Control from a Natural Insect pest
- GWSS research on mass trapping

VI. OTHER BUSINESS

None

VII. ADJOURNMENT

Chairman Dennis Burreson adjourned the Research Subcommittee meeting at 10:03 a.m.

Todd W. Sanders
Executive Director
California Olive Committee

SUMMARY OF MOTIONS FOR AUGUST 7, 2024

Motion 8-7-2024 #1 **APPROVED**

MOVED by RICCHIUTI, duly seconded by PFEIFFER and carried THAT the minutes for December 7, 2023, be approved as presented.

****INFORMATION ONLY****

FROM: COC RESEARCH SUBCOMMITTEE

SUBJECT: GUEST SPEAKER-SUTERRA

BACKGROUND:

Suterra is present to speak about their product BioMagnet Oro, an Attract and Kill Trap for the Olive Fruit Fly. Please see below link to their website! This product is not yet commercially available, but Suterra would like to know more about current industry practices related to the Olive Fruit Fly.

Suterra | BioMagnet™ Attract and Kill System

They are seeking to learn more about the industry and current needs:

- What are the current control methods for OFF?
- Are growers seeing a resistance to current controls?
- What is the severity of the fly and how difficult is it to control?
- Inputs in 2022 were cut in half. Was this due to weather?
- What is the economic damage to the grower?

****INFORMATION ONLY****

FROM: COC RESEARCH SUBCOMMITTEE

SUBJECT: DISCUSSION AND REVIEW OF 2024 PROJECTS

BACKGROUND: Each year, the Subcommittee funds research projects and requests progress reports from researchers. Provided in your packet are the current research progress reports for five projects and the final olive fruit fly reports for the northern and southern regions.

2024 Project Titles

TOPIC	LEADERS	AMOUNT
Management of Foliar Diseases-A. Olive Knot and B. Evaluation of New Fungicides For Control of Olive Leaf Spot—**page 12**	J.E. Adaskaveg	$13,715*
Epidemiology and Management of Olive Knot Caused by Pseudomonas Savastanoi pv. Savastanoi—**page 11**	J.E. Adaskaveg	$21,150*
Integrating Alternate Bearing Mitigation Strategies in a Commercial Table Olive Orchard—**page 13**	Carol Lovatt Elizabeth Fichtner	$33,825
Evaluation of effects of Acceede (ACC) on tree architecture, and harvester type on enhancing horticultural maturity and abcission zone development and commercial trunk shaking efficiency in table olives—**page 27**	Georgia Drakakaki Becky Wheeler-Dykes	$131,380**
Survey of Xylella fastidiosa diversity within California olive trees—**page 34**	Rodrigo Almeida	$29,752
Southern San Joaquin Valley Olive Fruit Fly Monitoring—**page 42**	Jim Stewart	$12,000
Sacramento Valley Olive Fruit Monitory Project—**page 43**	Ernie Simpson	$9,250
Total		**$251,072**

*Projects co-funded by the Olive Oil Commission of California.
**Project that Valent has contributed $8,500 to, reducing the fiscal impact to $122,880.

California Olive Committee (COC) and California Olive Oil Committee (COOC)

I. PROJECT TITLE: Epidemiology and management of olive knot caused by *Pseudomonas savastanoi*

PI: Dr. J. E. Adaskaveg **Cooperating:** H. Förster and D. Thompson

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Research objectives and results –

1. **Evaluate new bactericides: GRAS food additives, sanitizers, and other experimentals against *Psv.***

Based on our laboratory screenings of the sensitivity of the olive knot pathogen *Pseudomonas savastanoi* pv. *savastanoi* (*Psv*) to alternative conventional and natural toxicants, Ɛ-poly-L-lysine (EPL) in mixture with cinnamaldehyde was highly toxic. We are currently working with a registrant who is interested in this mixture and willing to design an agricultural formulation with UV-protection and adjuvants to improve residual activity. Several studies at two locations were initiated to evaluate efficacy of this mixture under field conditions, and results are pending. In a preliminary greenhouse study, this mixture significantly reduced the development of olive knot on inoculated leaf scar wounds but was not as effective as Kasumin or copper treatments. We are now evaluating higher rates.

Other compounds being evaluated in field studies on olive knot include Thymox and Thyme Guard (thyme oil), QAM (extract of an *Acacia* sp.), an 8L formulation of Kasumin by itself or mixed with Syllit or Vacciplant (laminarin), and a mixture of Instill (chelated copper sulfate pentahydrate) with ProBlad (extract of *Lupinus* sp.). In 2023, we conducted a field study on the efficacy of foliar or soil applications with potassium phosphite against olive knot, but no disease developed. Therefore, greenhouse studies were set up, where potted olive trees were soil-treated with phosphite, and twigs were inoculated after selected times. We hope to be able to provide results of some of these studies in our 2024 Annual Report.

In previous studies, among new treatments, the two antibiotics kasugamycin (Kasumin) and oxytetracycline (FireLine) by themselves or mixed with copper or Syllit were highly effective, and copper-Syllit outperformed copper alone. We are continuing our efforts in developing kasugamycin, oxytetracycline, and the antibacterial food preservatives that all have high in vitro toxicity and are effective in field assays. Dodine will be an important mixture component with copper, kasugamycin, and oxytetracycline. Field studies on olive knot are difficult to do. Development of the disease is dependent on environmental conditions, but even more so on the physiology of the host. Development of knots requires an active metabolism where the host responds with cell division to the influx of the plant hormone IAA that is produced by the pathogen. Therefore, knot production is most rapid in the greenhouse environment, where nutrients and water are continuously supplied.

2. **Continue to support the registration of the antibiotics kasugamycin and oxytetracycline, dodine, and the food preservatives EPL and nisin.**

Registration of oxytetracycline (Fireline) is proceeding with EPA, and the registrant (AgroSource) was expecting full registration. EPA, however, delayed the review of oxytetracycline most recently due to the Endangered Species Act being a high priority for the agency. The PRIA date for Kasumin and FireLine was again changed with no PRIA provided. Syllit is being federally registered on olive based on IR-4's submission to EPA through the Chemistry Science Advisory Council (ChemSAC) program in early 2021, and olive is being added to the label federally and later for the California label. The first PRIA date was November 2022. UPL also requested a concurrent review of kasugamycin and dodine in California last summer 2021 but EPA continues to delay the registration of kasugamycin until new policies are developed. At this date, dodine is still pending, and I requested an update from UPL. I envision that dodine can and will be mixed with copper products similar to copper-mancozeb treatments on walnut with higher efficacy and will prevent the selection of pathogen resistance to copper or dodine.

II. PROJECT TITLE: Evaluation of new fungicides for control of olive leaf spot (peacock spot)

PI: Dr. J. E. Adaskaveg **Cooperating:** H. Förster and D. Thompson

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Research objectives and results -

1. **Evaluate the performance of new and older fungicides in field trials and application timing of selected treatments.** In late-April 2024 evaluations of our field trials, disease incidence was low at all three

locations. Ziram (dithiocarbamate - FRAC Code - FC M3), Ph-D (polyoxin-D - FC 19), Syllit (dodine - FC U12), Abound (azoxystrobin - FC 11), the pre-mixtures Inspire Super (difenoconazole-cyprodinil - FC 3/9) and Quadris Top (difenoconazole/azoxystrobin - FC 3/11), and the tank mixture of Ph-D and Syllit were compared to copper as a standard treatment in reducing olive leaf spot in a trial on Manzanillo olive in Glenn Co. Using a single application in November 2023, all treatments significantly reduced the incidence of disease from the control, and there was no significant difference among treatments. In two trials on Arbequina olive in Sutter Co., biological treatments (Botector - *Aureobasidium pullulans*, YSY – *Papiliotrema terrestris*, Serifel – *Bacillus amyloliquifaciens*, ProBlad Verde - extract of *Lupinus*, BTS EXP 100 – bark extract), were evaluated and compared to polyoxin-D and Quadris Top. In the first trial, Ph-D was significantly more effective than Serifel, and the other treatments were intermediate in performance. Ph-D lowered the disease from 14.8% in the control to 2.8%. In the second study, ProBlad Verde, Botector, YSY, and BTS EXP 100 were similarly effective and significantly reduced peacock spot from the control. In two Yolo Co. trials, disease developed at very low levels, and no data could be obtained. Polyoxin-D (Ph-D) was also effective in our previous trials, and UPL has agreed to add olive to the fungicide label. This research is still ongoing.

2. **Evaluate application timing and adjuvants of selected treatments.** In a trial on Manzanillo olive in Glenn Co., single applications of Abound or Quadris Top (March 2024) were compared to two applications (Nov. 2023 and March 2024). Two applications were numerically more effective, but there was no significant difference between the two programs for both fungicides.

3. **Evaluate new fungicides for their in vitro activity**. We are attempting to determine the in vitro activity of selected fungicides that are effective in field trials. This is very challenging because of the difficulty in getting isolates to grow on laboratory media and because the fungus has an extremely slow growth rate. Additionally, the fungus does not sporulate continuously on leaves and only at specific times coinciding with rain in the fall, winter, and early spring seasons. This difficulty in isolating the pathogen is also experienced by researchers from other countries that we talked to at a national meeting in 2024.

4. **IR-4 GLP Studies (Registration update).** In 2022, we have reviewed protocols and proposed labels with IR-4 and the registrants, and we assisted in field studies that have been completed. Three fungicides, ziram, cyprodinil-difenoconazole (Inspire Super), and azoxystrobin-difenoconazole (Quadris Top) are currently in the system, and GLP field residue studies and laboratory residue studies have been completed for all three fungicides in 2021. Inspire Super and Quadris Top are moving forward through the registration process with support from Syngenta.

   Because EPA is proposing cancellation of ziram in the spring of 2022, the registration of ziram on olive will most likely be cancelled within the IR-4 program. Currently, EPA has decided to cancel the dithiocarbamates (e.g., ziram, thiram, ferbam). This past spring, I prepared letters to the EPA protesting the cancellation of ziram and noted that the IR-4 GLP studies for ziram use on olives were initially approved by EPA. I am hoping that the EPA will allow selected uses, but this is doubtful.

   Polyoxin-D (Ph-D) is a biopesticide, it is exempt from tolerance, and thus, no GLP residue studies are required. UPL is amending the label to include olives, and our efficacy data will be used to support the registration in California. UPL obtained approval for the old label without olives and will have to re-submit the federal and the state labels with olive added to the label.

   Dodine (Syllit) has European tolerances, and IR-4 petitioned EPA through the ChemSAC process to register the fungicide on olive using existing residue data. We prepared the request and justified the need. This label amendment was submitted by IR-4 on July 2, 2021. In the summer of 2021, the registrant has submitted for a concurrent review with the state of California. The first PRIA due date was November 2022. Because of ESA, EPA has not provided a new PRIA date, and this registration remains as pending. Syllit is currently registered on pome fruit and almond in California.

Department of Botany and Plant Sciences
Relevant AES/CE Project
No.: 4556

University of California
Division of Agricultural Sciences

PROJECT PLAN/RESEARCH GRANT PROPOSAL INTERIM PROGRESS REPORT

Project Year: 2024 Anticipated Period of Performance: year 2 of 3 requested

Project Leaders:

Carol Lovatt, Ph.D.
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ejfichtner@ucdavis.edu

Project Title: Integrating Alternate Bearing Mitigation Strategies in a Commercial Table Olive Orchard

Collaborator:
Kent Daane, Professor and Cooperative Extension Specialist, Department of Environmental Science, Policy, and Management, UC-Berkeley, CA, and Kearney Agricultural Research and Education Center, 9240 S. Riverbend Ave., Parlier, CA 93648: Phone: 559-646-6522; Fax: 559-646-6593; E-mail: kdaane@ucanr.edu

KD volunteered his time *gratis* to train the Co-PIs and their technical staff (1) to quantify black scale populations at important stages in their life cycle, honeydew produced by black scale, and sooty mold that grow on the honeydew and rate the impact on fruit quality, (2) to analyze the collected data, and (3) to assist the PIs in interpreting the results.

Cooperators:
Ismael Gutierrez, I.G. Harvesting, 113 E. Hickory St., Lindsay, CA 93247; Phone: 559-805-8181; E-mail: Gutierrezismael559@gmail.com

Donald Cleek, Agricultural Supervisor, UC Lindcove Research & Extension Center, 22963 Carson Avenue, Exeter, CA 93221; Phone: 559-592-2408, Ext 1157; Email: dlcleek@ucanr.edu

IG provided us with a 'Manzanillo' olive orchard in the Woodlake area through the 3-year project period. The orchard meets our research requirements, including having black scale infestation. IG is responsible for pruning and harvesting the research orchards.

DC coordinated the application of treatments, applied the foliar treatments (approval for DC to spray off the Lindcove REC and on the cooperators property has been secured), and coordinated the harvests. DC also provided us with a 'Manzanillo' olive orchard at the Lindcove REC, Exeter, CA, to determine whether pruning one side of the tree 28 DAFB and then the other side *biennially* is superior to pruning both sides of the trees in every other row every other year (there was no room to conduct this research at Woodlake).

Overview of the problem and summary of our research results to date: ***The Problem.*** Alternate bearing (AB), production of a heavy, high yield "on crop" followed by a light, low yield "off crop", is a significant economic problem. In ON-crop years, trees produce numerous small size fruit with reduced commercial value. In OFF-crop years, trees produce large fruit, often too large, and there are too few fruit to provide growers with a good income. In addition, our research also documented that fruit quality is reduced in some OFF-crop years, e.g., the large fruit of OFF-crop trees tend to turn black earlier in the season, which can further exacerbate the problem of too few commercially valuable fruit. In the ON-crop year, fruit take longer to mature, attain size, and accumulate oil, which delays harvest and further reduces floral intensity the following spring. Alternate bearing often occurs beyond the tree or orchard level, synchronizing across geographic regions, particularly when initiated by environmental conditions that affect crop load (i.e., heat at bloom in 'Manzanillo' orchards). An industry-wide shortage of fruit in the OFF-crop year has a negative economic impact on every step in the production chain from farm to consumer, including orchard management, harvesting, packinghouse and processor operations, manufacture of value-added products, marketing, and consumer prices, which taken together jeopardize the stability and sustainability of the table olive industry.

Climate is the major factor initiating alternate bearing. Adverse climate events, such as high or low temperatures, water-deficit stress or excessive winter rain causing soil hypoxia etc., that significantly reduce yield result in an OFF crop that is followed by an ON crop. Conversely, optimal climate conditions during flowering and fruit set, such that natural fruit thinning fails to occur result in an ON crop that is followed by an OFF crop. Adverse climate events also reduce bloom of the pollinizer trees in an orchard, reducing out-crossing and contributing to an OFF-crop. Climate events repeat in a random manner, creating a reoccurring need for a strategy to mitigate alternate bearing and the negative economic impact of alternate bearing on table olive growers and the industry.

Results of our prior COC-funded research demonstrated that the young developing fruit of the ON crop of 'Manzanillo' olive trees inhibit summer vegetative shoot growth and thereby reduce the number of nodes that can produce floral (inflorescence) buds the following spring (Fichtner and Lovatt, 2018; Sibbett, 2000). The developing ON crop also inhibits the transcription of key genes required for inflorescence development and flower formation. The maturing fruit of the ON-crop significantly increase abscission of floral buds for next year's bloom starting in September, as well as inhibit spring bud break (Chao, 2014; Fichtner and Lovatt, 2018; Fitchner et al., 2021). These latter effects are consistent with reports that late-harvesting of ON-crop trees further reduces return bloom. For ON-crop olive trees, the negative effects of fruit set on a shoot (localized effect) are stronger than the effects of the total number of fruit (crop load) on the tree (whole tree effect). As a result, bearing shoots on ON-crop trees, which are subjected to both effects produce virtually no flowers the following spring (0.8 inflorescences/shoot) (Fichtner and Lovatt, 2018). Thus, it is the nonbearing shoots on ON-crop trees, which are in the minority, that produce the inflorescences at spring bloom following the ON-crop year (13.8 inflorescences/shoot) (Fichtner and Lovatt, 2018). Further, a cytokinin-based strategy was only effective in stimulating summer vegetative shoot growth and increasing return bloom on nonbearing shoots of ON-crop trees (Fichtner and Lovatt, 2018; Fichtner et al., 2021). Thus, a crop reduction strategy that increases the number of nonbearing shoots is necessary to mitigate the negative effects of the ON-crop in an alternate bearing olive orchard. Results of our COC-funded research identified two effective crop reduction strategies. Foliar-applied NAA at full bloom (FB) and pruning (hedging and topping) 28 days after full bloom (DAFB) to one side of the tree and then the other side of the tree every other year, *biennially, not annually,* evened out total annual yield, but more importantly increased the 3-year and 4-year cumulative yield of medium + large size fruit (Fichtner and Lovatt, 2023). However, it is important to note that these results were obtained in a single field experiment. Additionally, the success of these two treatments was due in part to not carrying out the scheduled crop reduction treatments in year 5 (the final project year) due the low number of inflorescences at bloom based on the sum of the estimated floral intensity at bloom on the east and west sides of the trees in north-south running rows. Thus, knowing when to carry out crop reduction and when not to is important for maintaining high yields of medium + large size fruit. Before this information is recommended for implementation by table olive growers, the results need to be validated in a second experiment conducted in a new commercial table olive orchard.

Crop reduction strategies reduce yield and are thus, economically viable in high yield ON-crop years because they increase the yield of commercially valuable size fruit. Foliar application of NAA or pruning to low or medium bloom trees can reduce total yield and yield of medium + large size fruit below the profit margin. Pruning is critical to tree crop production to open the canopy for light penetration (no light, no flowers, no fruit), to increase canopy complexity to create new fruiting shoots and to balance the proportion of bearing vs. nonbearing shoots to maintain yield and fruit size on an annual basis, in addition to keeping rows open for orchard management, e.g., canopy spraying, harvesting. Pruning both sides of the tree in winter has the disadvantage that nonbearing and bearing shoots from the previous year, which have a high and low potential to flower the following spring, respectively, are not as easy to distinguish. Given the random reoccurrence of climate conditions that result in OFF or ON blooms, decisions about when, how many sides of the tree, and how frequently to prune or apply NAA in an orchard are critical and are best made based on a visual inspection of bloom, with knowledge of the potential impact that each decision has on the yield of medium + large size fruit not just in the ON year, but in subsequent years also. Additionally, it is important to know whether the strategy selected to mitigate alternate bearing will impact table olive orchard pest management. Choices for crop reduction starting in an ON-crop year include pruning both sides of the tree annually, just one side of the tree annually, one side of the tree every other year, or eliminating pruning in favor of using foliar-applied NAA. Thus, the crop reduction strategy selected to mitigate alternate bearing regulates the degree to which the canopy is open or closed at different times of the year and would thereby affect black scale survival, associated honeydew production and sooty mold growth, fruit quality and pesticide use.

Black scale population densities and damage have long been associated with temperatures in the olive tree canopy. More open canopies and more frequent pruning result in higher temperatures and drier conditions – both of which result in greater scale mortality. This relationship is also clearly associated with summer temperatures, but traditional pruning strategies are timed to post-harvest winter periods, when the scale population has already established and reached the second to third instar stages. Proposed herein are pruning strategies timed closer to June when the scale first instars have hatched and are most vulnerable to hot, dry conditions. By increasing scale mortality at this critical period, damage resulting from the scale's associated honeydew production and growth of sooty mold may be lowered, resulting in reduced insecticide use targeting black scale.

Objectives for 2024 (Year 2). There are three research objectives and goals:

(1) to test the results of our prior COC-funded research in a second commercial 'Manzanillo' table olive orchard to confirm that foliar-applied NAA at full bloom (FB) or pruning (hedging and topping) 28 days after full bloom (DAFB) to one side of the tree and then the other side of the tree every other year (*biennially*) are the best crop reduction strategies for mitigating the severity of alternate bearing and increasing annual yields of medium + large size fruit over multiple years compared to the ON-/OFF-yields of alternate bearing (untreated) control trees, trees treated with NAA or pruned on one side of the tree 28 DAFB and the other side *annually*, and trees pruned on two sides of the tree in winter annually (grower standard practice). Additionally, in a second 'Manzanillo' olive orchard located at the Lindcove REC in Exeter, objective 1 is to compare the efficacy of pruning (hedging and topping) 28 days after full bloom (DAFB) to one side of the tree and then the other side of the tree every other year (*biennially*) with pruning both sides of the trees in every other row every other year. Note: all pruning treatments include topping at the time of pruning. The goal is to determine the management strategy that maximizes yield of commercially valuable size fruit both during and after mitigation of alternate bearing;

(2) to use the sum of the bloom (floral intensity) estimates on two sides of each olive tree to decide when to prune or not prune a set of trees in order to maintain high yields of medium + large size fruit and thereby, test how well the relationships among estimated bloom, total yield and yield of medium + large size fruit from our previous COC-funded research hold up in a second orchard and to make any needed adjustments. The goal is to develop a decision support tool that growers will find easy, rapid and valuable to use annually across multiple acres of table olive trees to maintain yields of medium + large size fruit;

(3) to quantify the effects of the crop reduction strategies, which range from pruning both sides of the tree annually, one side annually, one side every other year, to no pruning for three years (NAA treated trees and the untreated alternate bearing control trees) on the resurgence of black scale populations, honeydew, associated sooty mold, and fruit quality in a 'Manzanillo' table olive orchard. The goal is to determine, to the degree climatic conditions during the 3-year experiment permit, whether the integration of specific alternate bearing mitigation strategies in a commercial table olive orchard has the potential to positively or negatively affect black scale pest management, e.g., pesticide use, and table olive fruit quality.

Research Accomplishments for 2024.

To meet the objective 1, we treated the trees in the commercial 'Manzanillo' olive orchard in Woodlake, which was going into an ON bloom/ON-crop year in 2024 because of the low yield OFF-crop in 2023, with the seven treatments listed below. The experiment was designed as a randomized complete block design with 17 individual tree replications per treatment and seven treatments that specifically meet the three objectives of the proposed research:

1) Untreated ON-crop control (last pruned and topped in winter 2021), e.g., alternate bearing control
2) Foliar applied NAA @ full bloom (FB) to one side of the tree in 2023 then the other side of the tree every other year (2025) (*biennially*)
3) Pruning (hedging and topping) @ 28 days after full bloom (DAFB) to one side of the tree (2023) then the other side every other year (2025) (*biennially*)
4) Foliar-applied cytokinin biosynthesis stimulator applied two weeks after a set of trees are pruned (hedging and topping) @ 28 DAFB on one side of the tree then the other side every other year (*biennially*). The cytokinin biosynthesis stimulator is applied *annually* to increase yield of medium + large size fruit in the current year and to stimulate summer vegetative shoot growth to increase return bloom
5) Pruning (hedging and topping) @ 28 DAFB to one side of the tree (2023) then the other side every other year (2025) (*biennially*) on a flexible schedule, using a decision support tool based on estimated sum of the bloom on the two opposing sides of the tree. This orchard runs east-west, bloom estimates are made on the north and south sides of the tree
6) Pruning 28 DAFB to one side of the tree (2023) and then the other side of the tree the next year (2024) *annually*
7) Control – grower standard practice of pruning two sides of the tree and topping in winter *annually*

Pruning in the winter was carried out by our cooperator I.G. Harvesting. Liqui-Stik Concentrate® NAA (Loveland Products), which is identical to and has the exact same label and application rate as AMVAC's Olive Stop®, which we used in our previous experiment, was applied according to the label directions by our cooperator D. Cleek.

In our second pruning trial in a 'Manzanillo' olive orchard in Exeter, CA, we had an excellent bloom and fruit set in 2024 and the research was conducted as planned. The objective of this research is to test the efficacy of pruning 28 DAFB applied to one side of the tree and then the other side *biennially* (e.g., east side in year 1 and west side in year 3 in north-south running rows) *versus* pruning both sides of the trees in every other row every other year (e. g., both sides of trees in rows 1, 3 and 5 pruned in years 1 and 3, whereas both sides of trees in rows 2, 4 and 6 are pruned in years 2 and 4). There were not enough trees to include this comparison at our Woodlake site. Moreover, this orchard was added to serve as a control for testing our estimated bloom model. We know our bloom model works in the Lindcove REC orchard. If it doesn't work at Woodlake and continues to work at Lindcove, then we learn that modification is required to expand its use across orchards. If the model doesn't work at either site this year, then we learn that the model's efficacy is influenced by climate, e.g., effects on fruit set.

Due to the loss of the crop at fruit set, the trees at Woodlake were not harvested. The trees at Exeter are scheduled to be harvested by I.G. in mid-October 2024. Total yield and fruit size distribution as kg/tree will then be determined and these data will be used to calculate yield and fruit size distribution as kg and number of fruit per tree. This year, we will have two years of yield data in Exeter to use in calculating the alternate bearing index (ABI) for total yield and yield of medium + large size fruit: ABI = (year 1 yield – year 2 yield)/ (year 1 yield + year 2 yield), in which yield is in kilograms of fruit per tree and the difference in yield between years 1 and 2 is expressed as an absolute number. An ABI of zero means no alternate bearing, whereas an ABI of one is complete alternate bearing, i.e., crop one year, no crop the other year (Pearce and Dobersek-Urbanc, 1967). Analysis of variance (ANOVA) will be used to test for treatment effects on bloom estimates, yield and fruit quality parameters, and ABI using the General Linear Model procedure of SAS (version 9.3; SAS Institute, Cary, NC). When ANOVA testing indicates significant differences, post-hoc comparisons will be run utilizing Fisher's protected least significant difference (LSD) test. Pearson's product moment correlation coefficients will be calculated to identify significant relationships ($r > 0.5$, $P \leq 0.05$). Significant correlations will be subjected to regression analyses, using the least squares method for the generalized linear model. The experiment is designed to determine the management strategy that maximizes yield of commercially valuable size fruit both during and after mitigation of alternate bearing, the goal of objective 1.

To meet objective 2, we estimated the floral intensity at full bloom at our research orchards in Woodlake and Exeter on two opposing sides of the tree on a scale from 0 to 3 (0 = no bloom, 1 = low bloom intensity, 2 = medium intensity bloom and 3 = high intensity bloom) and calculated the sum of the estimated bloom. Bloom estimates were made on the south and north sides of the trees (east-west rows) in the Woodlake orchard and east and west sides (north-south rows) in the Exeter orchard. The estimated sum of the bloom, based on the relationships with total yield and yield of medium + larges size fruit observed in our previous COC-funded research, was used to determine when and when not to prune the trees in treatment 5. Application of the sum of the estimated bloom in treatment 5, resulted in three trees not being pruned in 2023 and no trees being pruned in 2024. In addition, data collected in Years 1 and 2 and subsequent years will be used to analyze the relationships among bloom estimates, total yield and yield of medium + large size fruit by calculating Pearson's product moment correlation coefficients to identify significant relationships ($r > 0.5$, $P \leq 0.05$) to determine how well the new data fit our earlier results. Significant correlations will be subjected to regression analyses, using the least squares method for the generalized linear model, and more sophisticated analyses as warranted. Prior results suggest that the variability in the relationship between the range in total yields that result in high yields of medium + large size fruit is limited. If the yield data from this second experiment prove this to be the case, the sum of bloom estimates on two opposing sides of the tree should be able to predict when and when not to impose a crop reduction strategy to better maintain yields of medium + large size fruit from one year to the next across orchards of similar size trees pruned according to our prescribed strategy (Fichtner and Lovatt, 2023). Importantly, the model will indicate poor crop years when the yield of commercially valuable size fruit can only be maintained by eliminating the use of a crop reduction strategy.

Bloom estimates were determined in the second pruning experiment at the Lindcove REC in Exeter to determine whether pruning performed 28 DAFB on one side of the tree and then the other side every other year (*biennially*), e.g., east side in year 1 and west side in year 3 in a north-south running row, is superior to the proposed alternative of pruning both sides of the trees in every other row every other year, e. g., rows 1, 3 and 5 pruned on both sides in years 1 and 3 and rows 2, 4 and 6 pruned on both sides in years 2 and 4. These bloom estimates will be used to analyze the relationships among bloom estimates, total yield and yield of medium + large size fruit in the Exeter orchard. Thus, this orchard also serves as a control for testing our estimated bloom model as described above to meet objective 2.

<u>To meet objective 3</u>, in addition to going into an ON-crop year, the 'Manzanillo' table olive orchard we selected was documented in year 1 to have black scale. The orchard was not treated for black scale. Black scale population numbers, presence of honey dew and level of sooty mold were monitored as follows. The presence of honeydew droplets on olive leaves in spring, which corresponds to a rapid increase in scale size, is often the earliest signal of increased scale density in the orchard. On April 30, 2024, just a few days before full bloom on May 6, 2024, the terminal ends [20 inches long (about 50 cm)] of four branches were monitored on one tree per each of the 7 treatments in each of 6 replication and honeydew and sooty mold accumulation were rated on a scale of 0-3 (0 = no honeydew or no sooty mold, 1 = presence of honeydew or sooty mold, 2 = honeydew or sooty mold on < 30% of the branch, and 3 = honeydew or sooty mold on > 30% of the branch. Also in April, the scale density was evaluated by counting the number of mature scales (third instar to adult) on the terminal ends of the four branches on each tree per 7 treatments for six replications. For categorical ratings of scale number, honeydew presence and levels of sooty mold, treatment effects will be compared by ANOVA with treatments separated by Fisher's Least Significant Difference (LSD) and in a 2 by 2 contingency table with treatments separated using Pearson's Chi-square test. Scale densities will be compared using the General Linear Model function, with treatments separated using Tukey or Dunnett Pairwise comparison. This procedure was not carried out at harvest 2024 because there was an average of less than one black scale detected per tree in April across treatments.

To the degree climatic conditions during the 3-year experiment permit, the treatments in our experiment provide a range in canopy openness and closure and pruning times, which combined with our detailed analyses of black scale at two periods of the life cycle, plus honeydew and sooty mold ratings, will enable us to determine whether the integration of specific alternate bearing mitigation strategies in a commercial table olive orchard have the potential to positively or negatively affect black scale pest management, e.g., pesticide use, and table olive fruit quality (goal 3).

<u>Results for 2024.</u>

<u>Bloom estimates.</u> <u>Woodlake.</u> Estimates of floral intensity determined on April 30, 2024, just a few days before full bloom on May 6, 2024, for the north and south sides of the trees in the orchard in Woodlake, indicated significant differences in floral intensity across treatments that were not related to the sides of the tree treated in 2023 (Table 1). All trees had a sum of the estimated bloom for the south and north sides of the trees greater than 4.5 ± 0.25, the threshold value above which trees need to be treated with a crop reduction strategy in order to maximize yield of medium plus large (M+L) size fruit, with the exception of trees pruned on both sides of the tree in winter, the grower standard practice (Table 1). In 2023, at Woodlake, trees in Treatment 5, for which pruning was carried out on a flexible schedule based on bloom estimate, had three trees with bloom

estimates for the sum of the south and north sides of the tree that were below 4.5 ± 0.25. These trees were not pruned and the sum of the bloom for trees in Treatment 5 was restored to a high level greater than 4.5 ± 0.25 in 2024. We noted that at Woodlake, the trees in all treatments had lower bloom estimates on the north sides of the trees than the south sides of the trees in both 2023 and 2024, which suggests a negative impact on bloom independent of treatment but consistent with a negative effect due to shading.

Exeter. Estimates of floral intensity were determined on April 29, 2024, just a few days before full bloom on May 3, 2024, for the east and west sides of the trees in the orchard in Exeter. The results indicated significant differences in floral intensity across treatments that were not related to the sides of the trees treated in 2023 (Table 2). For examples, in 2023, trees in treatments 1 and 2 were pruned on the east side of the trees, with the untreated ON-bloom west side of the tree having a classic OFF-bloom in 2024, whereas trees pruned on both sides (treatment 3) or not pruned on either side of the tree (treatment 4) had blooms on both sides of the tree that were not significantly different.

Yield. There was no fruit to harvest in Woodlake in 2024; harvest in Exeter is planned by I.G. Harvesting for the week of October 14, 2024.

Honeydew and sooty mold levels and black scale number at Woodlake. The amount of honeydew and sooty mold present and average number of black scales per shoot per tree was determined on April 30, 2024, just a few days before full bloom on May 6, 2024. There was no honeydew visible on shoots in the 'Manzanillo' olive trees in Woodlake (Table 3). Sooty mold present per tree was less than 1 on a scale from 0 to 3, with 1 meaning that sooty mold was present at a low level on each tree, except trees in treatments 2 and 5 (Table 3). The number of individual black scales per tree was also less than 1 per tree, with no black scales on trees in treatments 1, 2, 5 and 7. Presence of honeydew and sooty mold are putative predictors of black scale populations. Whereas there was no honeydew observed in the 'Manzanillo' olive orchard in Woodlake at any of the three sampling dates, there was a significant relationship between the level of sooty mold present on the shoots of a tree and the number of individual black scales on the shoots of the tree. However, the strength of the relationship decreased from bloom 2023 ($r = 0.88$, $P < 0.0001$) to harvest 2023 ($r = 0.75$, $P < 0.0001$) to bloom 2024 ($r = 0.61$, $P < 0.0001$). Due to the fact that there was no honeydew at Woodlake, that the presence of sooty mold and number of black scales were low, and that we planned to replace this orchard with a new one, these data were not collected again at harvest 2024.

Future goals.

The future (remaining) goals for Year 2 are to harvest the trees in the 'Manzanillo' olive orchard in Exeter to determine total yield (kg/tree). These data will be used to determine fruit size distribution per tree and to analyze the relationships among estimated floral intensity, total yield and yield of M+L size fruit and to test the validity of our model at our Exeter 'Manzanillo' olive orchard. The goals for year 3 are to replace the Woodlake orchard with a new orchard going into an ON-bloom and having a history of black scale populations with no use of insecticide and a significant black scale population present throughout the orchard.

Outreach during Year 2 (2024).

During this period, Dr. Elizabeth Fichtner and Dr. Carol Lovatt have participated in outreach programs that have contributed significantly toward educating table and oil olive growers about factors that initiate alternate bearing, mechanisms that perpetuate the repeating cycles of ON/OFF crops, and strategies that mitigate alternate bearing.

1) California Olive Oil Commission Annual Member Meeting, March 2024, Monterey, CA, oral presentation, entitled "Mechanisms and Mitigation of Alternate Bearing in Olive."

2) North Coast Olive Field Day, July 2024, St. Helena, CA, oral presentation, poster and handouts, entitled "Alternate Bearing in Olive - *Initiation, perpetuation of OFF/ON yield cycles and mitigation strategies that reduce the severity of alternate bearing and increase yield of commercially valuable size fruit*."

3) Trade article: Andrews, E. and Fichtner, E.J. 2024. Pre-bloom foliar boron application on olive may improve yield. Progressive Crop Consultant, Topics in Subtropics newsletter.

4) Trade article: Fichtner, E. and Lovatt, C. 2024. Yield benefits from a new strategy using naphthaleneacetic acid to manage olive crop load. Progressive Crop Consultant, Topics in Subtropics newsletter; UCCE SJV Website.

<u>References</u>:

Chao, Y.Y. 2014. Alternate Bearing in Olive (*Olea europaea* L.). MS Thesis. University of California, Riverside, CA.

Fichtner, E., Lovatt, C.J. 2018. Alternate bearing in olive. Acta Hortic. 1199:103-108. doi:10.17660/ActaHortic.2018.1199.17

Fichtner, E., Lovatt, C.J. 2023. Alternate bearing in olive - *Mitigation with properly timed foliar-applied naphthaleneacetic acid or pruning.* Acta Hortic. In review.

Fichtner, E.J., Y.Y. Chao, L. Ferguson, J.S. Verreynne, L. Tang and C.J. Lovatt. 2021. Repeating cycles of ON and OFF yields in alternate bearing olive, pistachio, and citrus trees — *Different mechanisms, common solutions*. Acta Hortic. 1315. DOI 10.17660.

Pearce, S.C. and S. Dobersek-Urbanc. 1967. The measurements of irregularity in growth and cropping. J. Hort. Sci. 42(3):295–305.

Sibbett, S. (2000). Alternate bearing in olive trees. California Olive Oil News. 3(12),1

Table 1. Estimated floral intensity (bloom) of 'Manzanillo' olive trees in Woodlake, CA, prior to the application of the 2024 fruit thinning treatments, including foliar-applied naphthaleneacetic acid at approximately full bloom (May 6), pruning 28 days after full bloom (June 26) to one side of the tree and then the other side *annually* or *biennially*, with a set of trees pruned *biennially* treated 14 days later (July 5) with a foliar-applied cytokinin biosynthesis stimulator to increase yield of M+L size fruit in the current year and return bloom the following year, grower standard practice of pruning trees on both sides *annually* in winter, and untreated ON-crop control trees and the effect of these treatments on total yield (kg/tree) in October.

	2024 Bloom estimates[y] (Before 2024 treatments applied)			
2023 Treatment	South side of tree	North side of tree	Sum per tree	2024 total yield (kg/tree)
1) Untreated ON-crop control (last pruned and topped in winter 2021–2022) (alternate-bearing control)	2.9 a	2.1 ab	5.0 ab	
2) Foliar-applied NAA at FB to the south side of tree in 2023 and the other (north) side of the tree every other year (2025)	2.6 a	2.1 ab	4.8 ab	
3) Pruning (hedging and topping) 28 DAFB on the north side of tree in 2023 and then the other (south) side every other year (2025)	2.8 a	1.93 bc	4.7 b	
4) Foliar-applied cytokinin biosynthesis stimulator annually to the whole tree 14 days after pruning as in Treatment #3 was completed at 28 DAFB	2.8 a	2.0 ab	4.8 ab	
5) Pruning (hedging and topping) 28 DAFB to the north side of tree in 2023 and then the other (south) side every other year (2025) on a flexible schedule based on estimated floral intensity	2.7 a	2.1 ab	4.8 ab	

6) Pruning (hedging and topping) 28 DAFB on the north side of the tree in 2023 and then the other (south) side of tree annually (2024 and 2025)	2.9 a	2.3 a	5.2 a
7) Grower standard practice of pruning both sides of tree and topping in winter 2023 and annually (2024 and 2025)	2.3 b	1.7 c	3.9 c
P-value	< 0.0001	0.0084	< 0.0001

[z] Bloom was evaluated on the following scale: 0, no inflorescences; 1, low floral intensity; 2, medium floral intensity; and 3, high floral intensity.

[y] Full bloom was May 6, 2024. Bloom estimates were made April 30, 2024. NAA was applied May 6, 2024. Winter pruning was March 27, 2024; no other pruning treatments were required in 2024. The cytokinin synthesis biostimulator was not applied in 2024, because there were no fruit set and we plan to select a new orchard for 2025.

[x] Mean values within a vertical column followed by different letters are significantly different at the specified *P* level by Fisher's Protected LSD test.

Table 2. Estimated floral intensity (bloom) of 'Manzanillo' olive trees in Exeter, CA, prior to the application of the 2024 fruit thinning treatments, which included pruning (hedging and topping to 14 feet) 28 days after full bloom (DAFB) (June 3) to one side of the tree and then the other side *biennially versus* pruning 28 (DAFB) (June 3) on both sides of the trees in every other row every other year and the effect of these treatments on total yield (kg/tree) to be harvested in October.

Treatment	2024 Bloom estimates[y] (Before 2024 treatments applied) East side of tree	West side of tree	Sum per tree	2024 yield (kg/tree)
1) Pruned 28 DAFB on east side of the tree in 2023; pruned on the west side in 2025	2.5 a	1.4 bc	3.9 ab	
2) Pruned 28 DAFB on east side of the tree in 2023; pruned on the west side in 2025	2.1 a	1.0 c	3.1 b	
3) Pruned 28 DAFB on both sides of the tree in years 2023 and 2025	2.0 a	2.0 ab	4.0 ab	
4) Pruned 28 DAFB on both sides of the tree in 2024	2.1 a	2.3 a	4.4 a	
P-value	0.5072	< 0.0001	0.1732	

[z] Bloom was evaluated on April 29, 2024, using the following scale: 0, no inflorescences; 1, low floral intensity; 2, medium floral intensity; and 3, high floral intensity.
[y] Full bloom was May 3, 2024. All trees were pruned on June 3, 2024, and all trees were topped on June 6, 2024.
[x] Mean values within a vertical column followed by different letters are significantly different at the specified *P* level by Fisher's Protected LSD test.

Table 3. Honeydew and sooty mold ratings and number of black scale per shoot of 'Manzanillo' olive trees in Woodlake, CA, on April 30, 2024, prior to the application of the 2024 fruit thinning treatments, including foliar-applied naphthaleneacetic acid at approximately full bloom (May 6, 2024), winter pruning 28 days after full bloom (DAFB) (June 26) to one side of the tree and then the other side *annually* or *biennially*, with a set of trees pruned *biennially* treated 14 days later (July 5) with foliar-applied cytokinin biosynthesis stimulator to increase the yield of M+L size fruit in the current year and increase return bloom the following year, grower standard practice of pruning trees on both sides *annually* in winter, and untreated ON-crop control trees.

	2024 honeydew and sooty mold ratings and number of black scales[z]			
	Data collected 1 week prior to 2024 full bloom treatment applications			
2024 Treatment	Honeydew	Sooty mold	Black scale	
1) Untreated ON-crop control (last pruned and topped in winter 2021–2022) (alternate-bearing control)	0 a	0.21 b	0.00 b	
2) Foliar-applied NAA at FB to the south side of tree in 2023 and the other (north) side of the tree every other year (2025)	0 a	0.00 b	0.00 b	
3) Pruning (hedging and topping) 28 DAFB on the south side of tree in 2023 and then the other (north) side every other year (2025)	0 a	0.04 b	0.04 b	
4) Foliar-applied cytokinin biosynthesis stimulator annually to the whole tree 14 days after pruning, as in Treatment #3, was completed at 28 DAFB	0 a	0.50 a	0.37 ab	
5) Pruning (hedging and topping) 28 DAFB to the south side of tree in 2023 and then the other (north) side every	0 a	0.00 b	0.00 b	

other year (2025) on a flexible schedule based on estimated floral intensity			
6) Pruning (hedging and topping) 28 DAFB on the south side of the tree in 2023 and then the other (north) side of tree *annually* (2024 and 2025)	0 a	0.17 b	0.67 a
7) Grower standard practice of pruning both sides of tree and topping in winter 2023 and *annually* (2024 and 2025)	0 a	0.08 b	0.00 a
P-value	NA	0.0820	0.4847

[z] Honeydew and sooty mold were evaluated on the following scale: 0, not present; 1, present; 2, present on < 30% of the shoot; and 3, present on > 30% of the shoot and reported as average per shoot; individual black scales were counted and reported as number/shoot.
[y] Full bloom was May 6, 2024. Honey dew and sooty mold presence and black scale numbers were determined on April 30, 2024. NAA was applied May 6, 2024. Winter pruning was March 27, 2024; no other pruning treatments were required in 2024. The cytokinin synthesis biostimulator was not applied in 2024, because there were no fruit set and we plan to select a new orchard for 2025.
[x] Mean values within a vertical column followed by different letters are significantly different at the specified *P* level by Fisher's Protected LSD test; NA = not applicable.

Workgroup/Department: Olive / Plant Sciences, UC Davis

Project Year: April 1, 2024 – March 31, 2025. Anticipated Duration of Project: 1 year
(UC Davis Sponsored Programs Proposal # xxxxx- submitted)

Project Title:
Evaluation of effects of Accede® (ACC), tree architecture, and harvester type on enhancing horticultural maturity and abscission zone development and commercial trunk shaking efficiency in table olives

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2024 Season Summary

During the 2024 growing season, we tested the combination of skirt pruning and foliar spray of Accede^{TM} (ACC, 1-aminocyclopropane-1-carboxylic acid) at 1500 ppm, 100 GPA rate in promoting mechanical harvest efficiency. ACC was applied on September 30th (1369 GDD) and the mechanical harvest was performed after 7 days on Oct 7th (1442 GDD). Spring skirt pruning does not generate a yield penalty. We followed last year's protocol that we timed the ACC application again at the point where the fruit removal force was around 0.3 kg, and we observed significant fruit removal force decrease compared to control in both 3 days and 7 days after ACC application. Combination of skirt pruning and ACC application induced 23.4% increase of trunk shaker mechanical harvest efficiency, compared to the unskirted control.

Through this season's work, we have established a recommendation of mechanical harvesting protocol for high efficiency

1. Skirting pruning during May - June

2. During pre-harvest (early September or starting at GDD 1000°C), Monitor fruit removal force, until it drops below 0.3 kg.
3. Appy 1500 ppm ACC afterwards, and harvest after 7-10 days post application.

This year's results showed that skirting low branches in spring does not affect the total yields of the trees. This year's results confirmed that ACC application after fruit removal force dropping below 0.3 kg can increase the mechanical harvesting efficiency by ~15%, similar to last year's results.

Cellular analyses confirmed that ACC promotes the formation of fruit abscission zone (FAZ). In FAZ cell layer of ACC treated fruits, alkalization of cytosol and decrease of plasmodesmata callose were observed as in ripe fruits. This underlines natural cellular mechanisms of FAZ formation that can be accelerated with ACC application.

We have initiated the IR-4 application process. Next year we will obtain the third year's data to fulfill the IR-4 application requirements.

Material and Methods

As indicated in the plot map on the next page. Individual rows of 1-13 were sprayed with 1500 ppm ACC or the control spray + 0.025% non-ionic surfactant (Activator 90). Half of each row is skirted. Except rows 1, 3, 12, and 13, all the rows are harvested by trunk shaker and hand gleaned afterwards. Harvester performed a 15-tree run and we performed timing experiment on 10/7/24 during 8 am to 2 pm, and hand-gleaning was performed on 10/15/24 and 10/16/24 during 7 am to 2:30 pm, to compare the efficiency of mechanical harvester and hand gleaning method.

At harvest, the mechanically harvested yield of 15-tree plots was weighed, and a subsample was submitted for grading and determination of price per ton for each plot. Grading samples were submitted to Musco Orland Receiving Station for quality grading. We will report the grades among ACC treated fruits and control samples in the final report.

Harvester efficiency of each treatment are calculated as:

$$Efficiency = \frac{Mechanically\ harvested\ (lb)}{Manually\ harvested\ (lb) +\ Mechanically\ harvested\ (lb)} \times 100$$

Pull tests will be performed by 12 rows x 4 trees per row x 5 olives per tree = 240 olives per pull test on 8/23, 8/30, 9/9, 9/18, 9/23, 9/29, 10/3, and 10/7.

Nickels Olive Accede Trial 2024 Map

N

row/tree	1	2	3	4	5	6	7	8	9	10	11	12	13
1	#1	#3	#5	#7	#4	#6		#2	#8	#4	#8	#2	#6
2													
3													
4													
5													
6													
7	X	X	X	X	X	X		X	X	X	X	X	X
8													
9													
10													
11													
12													
13													
14													
15													
16	#2	#4	#6	#8	#3	#5		#1	#7	#3	#7	#1	#5
17													
18													
19													
20													
21													
22	X	X	X	X	X	X		X	X	X	X	X	X
23													
24													
25													
26													
27													
28													
29													
30													
	1	2	3	4	5	6	7	8	9	10	11	12	13

Note: Planted 7-8-01. Tree spacing =12'x18' or 202 trees/ac

Treatments: :

Treatment 1	ACC + Skirting + TS
Treatment 2	ACC + No Skirting +TS
Treatment 3	ACC + Skirting + TS
Treatment 4	ACC + No Skirting + TS
Treatment 5	Control + skirting +TS
Treatment 6	Control + no skirting + TS
Treatment 7	Control + skirting + TS
Treatment 8	Control + no skirting + TS

Results of 2024 season

Fruit removal force reaches ~0.3 kg in late September

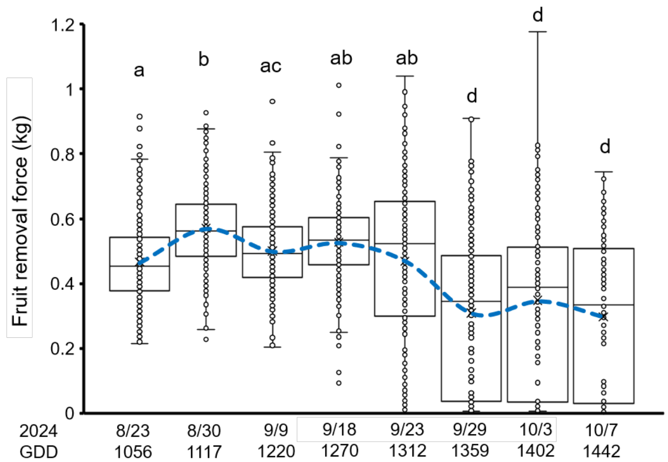


Figure 1. Fruit removal force during pre-harvest stages of olive fruit development. Each biological replicate is one fruit. Blue dashed line indicates the change of average during the sampling period. Different letter indicates significant difference between groups (n = 120 or 240, one-way ANOVA, Scheffe's Test, $p < 0.05$). Growing degree day (GGD), is shown on the x axis and is calculated using 15°C as base value and May 10th, 2024 as half bloom day, and mechanical harvest was performed on 10/7/24.

We continued to monitor fruit removal force in relation to growing degree days (GDD) and calendar dates. A similar trend was observed throughout the past three years and a major drop of fruit removal force was also observed near the end of September this year, at around 1350 GDD (**Figure 1**). We followed the criteria of 0.3 kg fruit removal force and sprayed with ACC at 1500 pm, with a 100 gallons per acre (GPA) rate on September 30th, 2024. The mechanical harvest was performed on October 7th, 2024, 7 days after the spraying the orchard.

ACC application significantly reduces FRF

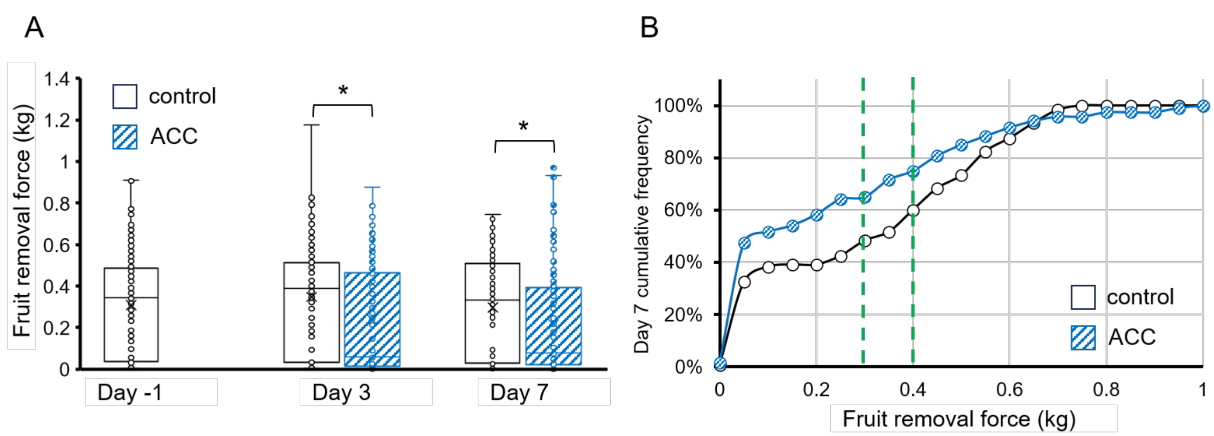


Figure 2. (A) Fruit removal force before and after ACC treatment. Each biological replicate is one fruit. Asterisk indicates significant difference between control and ACC treated group (n = 120, t-test, $p<0.05$). **(B) Cumulative frequency of fruit removal force of fruits sampled on the harvest day.** Green dashed lines suggest fruit removal force range corresponding to mechanical harvest removal rates.

ACC treatment (1500 ppm, 100 GPA) was performed on 9/30/24 (GDD 1369°C), and no significant FRF changes were observed during the 8 days in the control group (**Figure** 2) in the course of the ACC application period (9/30-10/7). However, in the ACC treated group, a significant lower FRF was observed on both the 3rd-day and 7th -day after treatment.

Cumulative frequency of FRF on 7th-day after treatment (**Figure 2B**) suggest that the fruits with lower than 0.4 kg FRF, represent 75% and 60% of the population in ACC treated and control group, which approximately matches the mechanical harvest efficiency of ACC treated and control group. Our data of 2024 (**Figure 2B**) suggested that this year the threshold for using the trunk shaker to remove a fruit from the tree is between 0.3 - 0.4 kg.

Spring skirting pruning does not affect yield

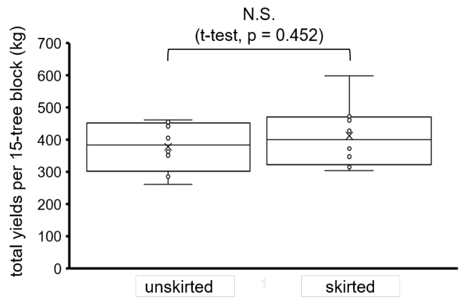
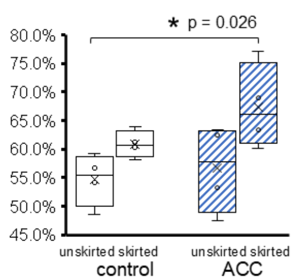


Figure 3. Total yields of two pruning treatment (unskirted vs skirted). Each biological replicate is a block of 15 trees (n = 8).

We found that low branches dangling on the receiver of trunk shaker cannot be efficiently harvested by the trunk shaker. Sometimes lower branches even directly block the receiver and get knocked off by the receiver. We performed a skirting pruning test (4 feet above the ground) this spring. The total yield of 15-trees, including the weight of mechanical harvest and hand gleaned fruits, are not significantly different between the unskirted and skirted groups (**Figure 3**). These results suggest that skirting improves mechanical harvesting efficiency without the penalty of yield. Skirting pruning should be the practice to follow, preparing the orchard in the spring for the upcoming fall mechanical harvesting.

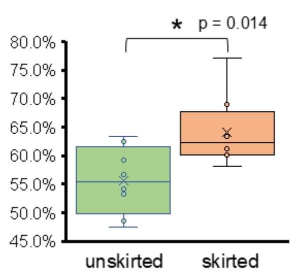
Both skirting pruning and ACC application increase mechanical harvest efficiency

A



	control	ACC	efficiency increase by ACC
unskirted	54.7%±2.3%	56.6%±3.8%	*3.5%*
skirted	60.9%±1.2%	67.4%±3.7%	*10.7%*
efficiency increase by skirting	*11.3%*	*19.1%*	*Combinational increase: 23.4%**

B



	combined
unskirted	55.6%±2.1%
skirted	64.2%±2.2%*
efficiency increase by skirting	*15.5%*

Figure 4. Efficiency comparison (**A**) of pruning and ACC treatments. Asterisk indicates significant higher efficiency (23.4%) observed in the group treated by both ACC spray and skirt pruning, compared to the no treatment control (n = 4, t-test, p = 0.026). (**B**) by combining spray treatment groups. Asterisk indicates significant higher efficiency is observed in skirted group (n = 8, t-test, p = 0.014). Each biological replicate is a block of 15-tree.

We tested effect of skirting and ACC spray on mechanical harvest this year. Both induced an increase in harvesting efficiency. Combinational treatment of ACC spray and skirting induced a **23.4% increase of harvesting efficiency** (**Figure 4A**), while improved efficiency was also observed by skirting alone (15%) (**Figure 4B**).

This year's result confirmed the efficacy of ACC implemented using current established orchard management practices. Further our 2024 results provide 2nd year supporting evidence for IR-4 application of ACC in the table olive mechanical harvesting.

Comparison of growing degree days during the 2022 - 2024 growing seasons

We are working to build the growing degree day (GDD) model for the table olive mechanical harvest and general olive industry (**Figure 5**). Using the **formula (1)** with T_{base} as 15°C, which is a basic growing degree day model formula, a very similar trend of 2022 and 2024 was observed, deviating far from 2023. We will continue to monitor the GDD and will establish a better model accompanied to the fruit removal force to better predict the ACC application and mechanical harvest.

$$GDD = \frac{(T_{max} + T_{min})}{2} - T_{base} \qquad (1)$$

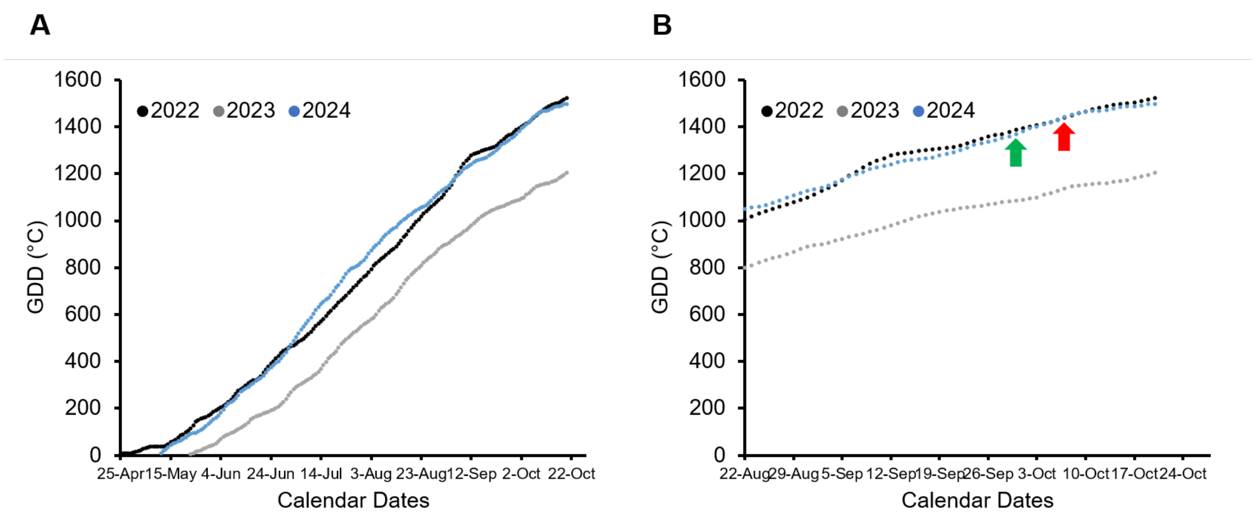


Figure 5. Growing degree days comparison for 2022 – 2024 growing season. (A) Full season heat unit accumulation starting from half bloom day. **(B)** Zoom-in of pre-harvest period. Green arrow indicates 2024 ACC application date (9/30/24, 1359 GDD) and red arrow indicates 2024 harvest date (10/7/24, 1442 GDD).

Live staining confirms the efficacy of ACC to promote FAZ formation at the cellular level

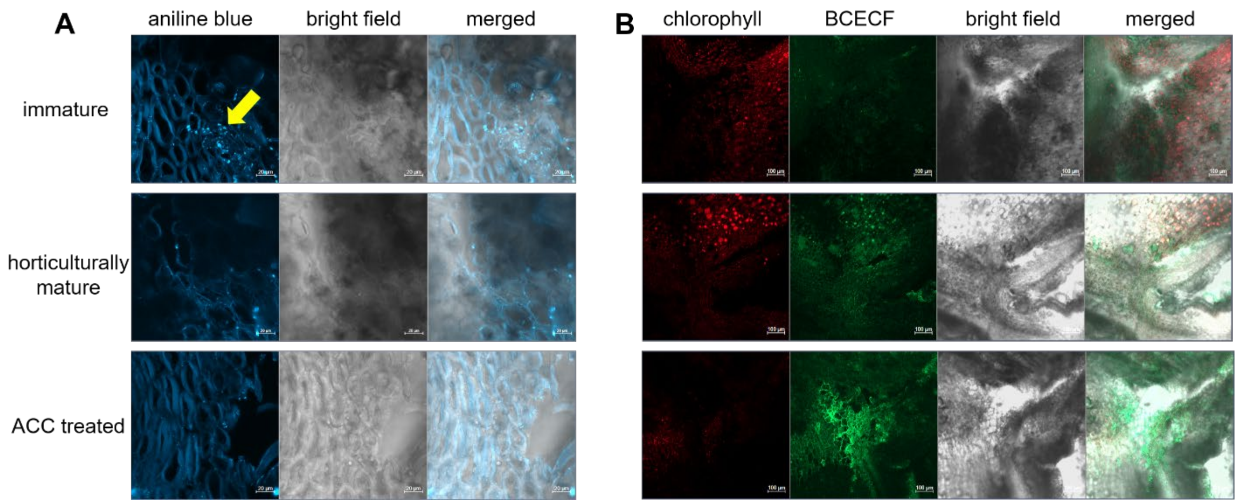


Figure 6. Live staining of callose (A) and cytosolic pH (B) at olive FAZ. (**A**) Fluorochrome aniline blue staining of callose at plasmodesmata. Yellow arrow points to blue-fluorescent spots that indicate the presence plasmodesmatal callose at FAZ. Scale bar = 20 µm. (**B**) BCECF cytosolic staining of FAZ. Higher green signal indicates higher pH. Scale bar = 100 µm.

Less plasmodesmata callose staining signals were observed in horticulturally mature samples compared to immature samples at FAZ, suggesting that symplasmic trafficking is promoted during the maturation process. ACC treated samples also showed a higher rate of FAZs with low plasmodesmata callose (**Figure 6A**), suggesting ACC application advances the maturation process. We observed the same effect in ethephon treated samples in previous years' study.

Alkalization of abscission zone cytosolic pH was previously suggested as an indicator of fruit abscission zone formation (Sundaresan et al., 2015). We now observed this phenomenon in a tree crop, olive. More cells with alkalized cytosol were observed in horticulturally mature and ACC treated samples (**Figure 6B**).

Our manuscript on small-trial ethephon application is in progress. We observe similar abscission zone changes in 1500 ppm ACC-treated and 1500 ppm ethephon-treated samples, in live staining.

In summary, live staining of plasmodesmata callose and cytosol pH confirms the efficacy of ACC in promoting the formation of fruit abscission zone at cellular level.

Survey of *Xylella fastidiosa* diversity within California olive trees

-Interim Report October 2024-

Principal Investigator:
Professor Rodrigo Almeida
Department of Environmental Science, Policy, and Management
University of California, Berkeley

Cooperator:
Patrick Lee

In this interim report we summarize activities on this project. Because the survey of the pathogen *Xylella fastidiosa* in olive trees requires some disease symptom expression for the selection of samples, field work started in the summer of 2024. Research is ongoing, as described below.

Aims:

1. Determine the genetic identity of *Xylella fastidiosa* (*Xf*) strains causing disease in ornamental olive trees.
2. Identify environmental factors associated with high strain diversity to inform future management practices.

Methods:

Field collections:

Maps generated with data from Love et al. (2022) provide geographically explicit locations of all olive trees throughout Los Angeles and San Diego counties. Areas with high densities of olive trees were chosen for sampling. In San Diego, this process was repeated until a representative area of the county was sampled.

Only olive trees showing symptoms of *Xf* infection (canopy dieback, leaf scorch, etc.) were sampled. Initial sampling protocol was based on standard protocols used by European agencies and scientists studying *X. fastidiosa* (EPPO 2018). Approximately 20 cm of olive branch tissue was collected from each symptomatic olive tree using a pole pruner or hand shears.

Diagnostic testing:

All samples were screened for *Xf* using a robust qPCR assay (Harper et al. 2010); a DNA-based molecular method for pathogen detection. This test determines whether *Xf* cells are present in the sample, and allows for approximation of bacterial population size.

Bacterial Isolation:

Attempts to isolate *Xf* cells in pure culture were made for a subset of samples. Media preparation, tissue homogenization, and growing conditions were all chosen from an established and widely used protocol (Hill & Purcell 1995). Isolated *Xf* cells were then stored in -80C for future experiments.

Genetic typing:

Multi-locus sequence typing (MLST) is a genetic typing scheme that can be used to broadly categorize the genetic identity of bacterial isolates. MLST protocols developed for *Xf* will be used to determine the sequence type (ST) for positive infections detected initially by qPCR (Yuan et al. 2010). Whole genome sequencing (WGS) provides a much higher coverage of the bacterial genome, but requires cells isolated in pure culture. All bacterial isolates obtained from the field will be sequenced using WGS platforms.

<u>Summary of work completed:</u>

Overview:

Three collection trips were conducted from June to September 2024, when we expect to find more disease symptoms and higher pathogen loads in olive trees. Two hundred-forty sex symptomatic olive trees were sampled across San Diego County (Figure 1) and the greater Los Angeles area (Figure 2).

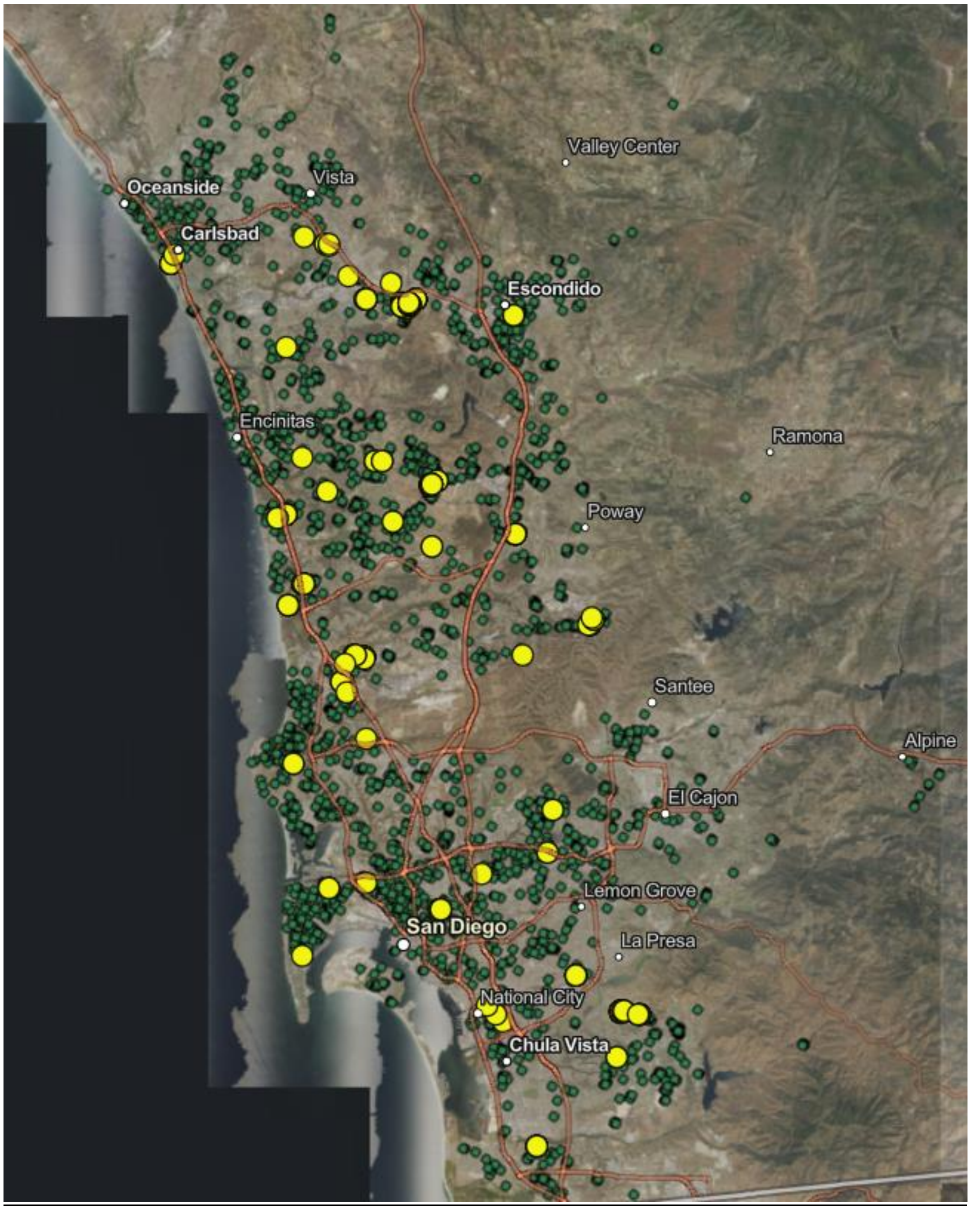


Figure 1: Distribution of olive trees in San Diego County. Green dots represent the distribution of all olive trees. Yellow dots indicate trees that were sampled for *Xf* in June - September 2024.

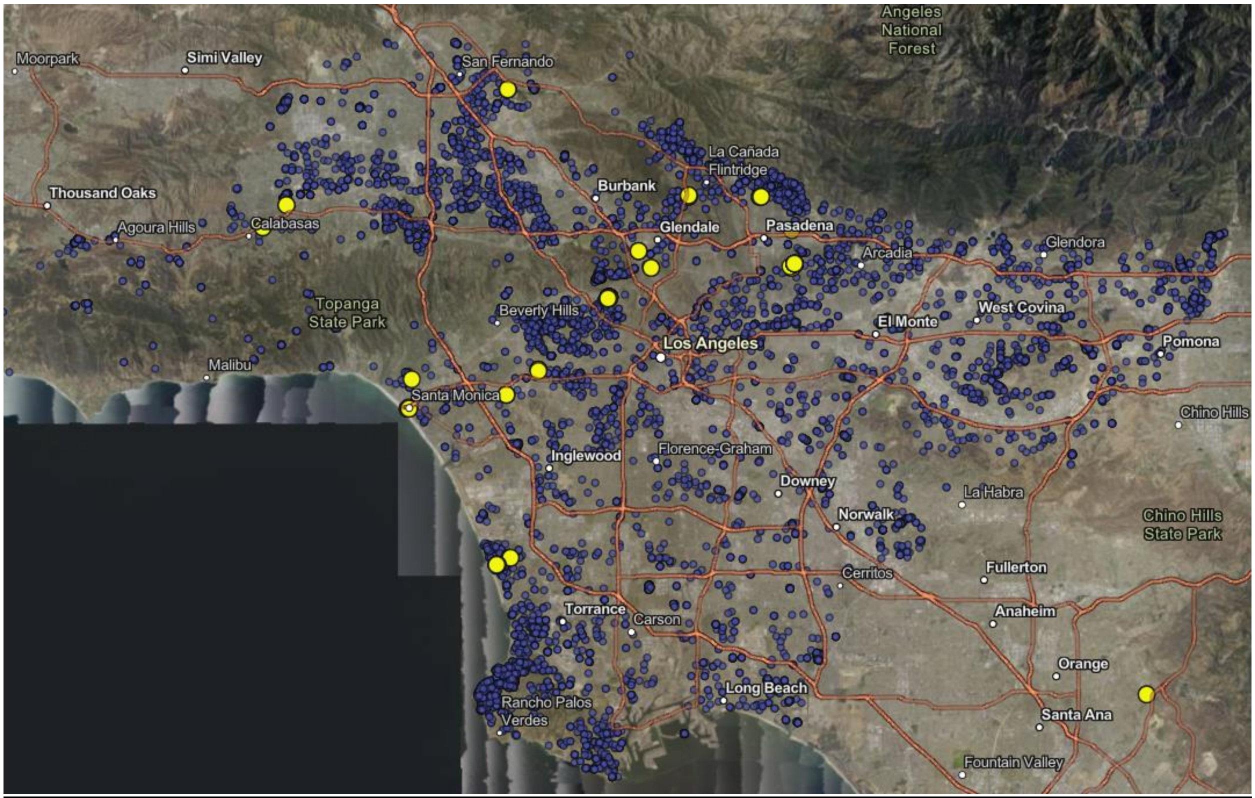


Figure 2: Distribution of olive trees in the greater Los Angeles area. Blue dots represent the distribution of all olive trees. Yellow dots indicate trees that were sampled for *Xf* in September 2024.

Common symptoms observed included canopy dieback (Figure 3) and leaf scorching (Figure 4).

Figure 3: Symptomatic olive trees in Manhattan Beach, CA (left) and San Marcos, CA (right). Note severe dieback throughout the canopy.

Figure 4: Leaf scorch on olive sample.

While the effort to process these samples is still ongoing, preliminary results indicate that approximately <u>33% of all samples</u> have tested positive for *Xf* via qPCR, indicating that the pathogen is prevalent in Southern California olive populations.

Isolation attempts:

Attempts to isolate *Xf* in pure culture were unsuccessful across samples from 77 olive trees. Obtaining *Xf* isolates from olive is notoriously difficult, but further optimization of culturing protocols is possible. Our lab group is soliciting feedback from colleagues in both the United States and Europe to improve our chances of isolation success in the future. Burbank et al. successfully isolated strains of *Xf.* from olive trees in California (O'Leary et al. 2020), and the group lead by Maria Saponari has developed protocols to successfully isolate *Xf* subsp. *pauca* from olive trees in Italy. *Xf* subsp. *pauca* is very distinct from North American *Xf* strains, but we work closely with the Italian group leading research on the olive disease in that region. Their feedback will generate critical improvements to our protocol that will increase our likelihood of obtaining more *Xf* isolates from olive trees next season.

Genetic typing:

We are processing samples to be genotyped using this approach, which will be helpful in generating insights into the genetic diversity of the pathogen infesting olive trees in our samples. Results are pending as the sampling season is finalizing and the focus on lab work has been initiated.

Successful isolation of *Xf* from non-olive tree species provides an insight into potential risks to the industry, as *Xf* from one host plant species may infect another, leading to disease emergence. Two isolates were collected from symptomatic New Zealand Christmas tree (*Metrosideros excelsa*) and purple leaf plum (*Prunus cerasifera*) trees encountered incidentally while searching for olives. Preliminary MLST sequencing reveals they are *Xylella fastidiosa* subsp. *multiplex*, a subspecies previously isolated from olive in California. These strains belong to a larger genetic subgroup that is most associated with almond leaf scorch (ALS) in multiple countries in Europe (ST6). Strains in ST6 have not been reported in either of these plant host species before, suggesting that urban centers are indeed incubators for novel strains of *Xf*.

<u>Future research:</u>

We plan to expand the scope and success of this work in 2025. Symptomatic olive trees were abundant and widespread throughout Los Angeles County in a preliminary collection effort, suggesting the need to expand sampling in the future. We are also leveraging our network of collaborators to improve our existing protocols and address relevant questions that require additional expertise. *Xf* is clearly abundant in the urban forests of Southern California. Continued work will lead to the identification of prolific strains of *Xf* and assess the risk they pose for California olive growers.

Works cited:

Love, Natalie L.R., Viet Nguyen, Camille Pawlak, Andrew Pineda, Jeff L. Reimer, Jennifer M. Yost, G. Andrew Fricker, et al. "Diversity and Structure in California's Urban Forest: What over Six Million Data Points Tell Us about One of the World's Largest Urban Forests." *Urban Forestry & Urban Greening* 74 (August 2022): 127679. https://doi.org/10.1016/j.ufug.2022.127679.

"PM 7/76 (5) Use of EPPO Diagnostic Standards." *EPPO Bulletin* 48, no. 3 (2018): 373–77. https://doi.org/10.1111/epp.12506.

Harper, S. J., L. I. Ward, and G. R. G. Clover. "Development of LAMP and Real-Time PCR Methods for the Rapid Detection of Xylella Fastidiosa for Quarantine and Field Applications." *Phytopathology*® 100, no. 12 (December 2010): 1282–88. https://doi.org/10.1094/PHYTO-06-10-0168.

Hill. B.L., and Purcell, A.H. (1995). "Acquisition and retention of *Xylella fastidiosa* by an efficient vector, *Graphocephala atropunctata*. *Phytopathology*® 85, 209-212.

Yuan, Xiaoli, Lisa Morano, Robin Bromley, Senanu Spring-Pearson, Richard Stouthamer, and Leonard Nunney. "Multilocus Sequence Typing of *Xylella Fastidiosa* Causing Pierce's Disease and Oleander Leaf Scorch in the United States." *Phytopathology*® 100, no. 6 (June 2010): 601–11. https://doi.org/10.1094/PHYTO-100-6-0601.

O'Leary, Michael L., Lindsey P. Burbank, Rodrigo Krugner, and Drake C. Stenger. "Complete Genome Sequence Data of Three *Xylella Fastidiosa* Subsp. *Multiplex* Strains Isolated from Olive Trees in California, U.S.A." *Phytopathology*® 110, no. 11 (November 2020): 1759–62. https://doi.org/10.1094/PHYTO-05-20-0167-A.

Southern Region Olive Fruit Fly Project

Sponsored by: California Olive Committee and AgIPM Consultants

2024 Total OLFF for the Week Ending

	Mar.27		Apr. 05		Apr.12		Apr. 19		Apr.26		May. 03		May.10		May. 17		May.24		May. 31		Jun.07		Jun. 14		Jun.21		TOT/YR	
Block	M	F	M	F	M	F	M	F	M	F	M	F	M	F	M	F	M	F	M	F	M	F	M	F	M	F	**M**	**F**
Woodlake	set	set	21	1	6	1	31	2	2	1	6	0	0	0	4	3	16	1	1	7	1	0	26	0	1	0	115	16
Ivanhoe	set	set	4	2	4	0	0	0	1	0	0	0	0	0	5	1	2	0	5	0	2	0	0	0	0	0	23	3
Exeter	set	set	8	1	3	0	1	0	7	1	4	0	1	0	1	0	0	0	0	0	0	0	0	0	0	0	25	2
South Exeter	set	set	1	1	0	0	4	2	2	0	0	0	1	0	1	0	0	0	0	0	1	1	2	0	0	0	12	4
Tonyville	set	set	8	1	4	1	6	2	8	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	26	4
W. Lindsay	set	set	7	5	3	0	8	3	1	0	7	0	0	0	1	0	0	0	3	0	0	0	0	0	1	0	31	8
Strathmore	set	set	0	0	3	0	8	0	1	0	1	0	0	0	0	0	0	0	1	0	0	1	0	0	0	0	14	1
Porterville	set	set	21	1	5	1	10	1	7	1	0	0	1	0	1	0	0	0	3	0	1	0	0	0	2	0	51	4
Terra Bella	set	set	4	0	1	0	1	0	0	0	0	0	0	0	1	0	1	0	0	0	1	0	0	0	0	0	9	0
Total			74	12	29	3	69	10	29	3	18	0	3	0	14	4	19	1	13	7	6	2	28	0	4	0	306	42
City of Visalia	set	set	0	0	0	0	0	0	0	0	0	0	0	0	0	0	1	0	3	0	1	1	0	0	8	1	13	2
	Jun.28		**Jul. 05**		**Jul.12**		**Jul. 19**		**Jul.26**		**Aug. 02**		**Aug.09**		**Aug. 16**		**Aug.23**		**Aug. 30**		**Sep.06**		**Sep. 13**		**Sep.20**		**TOT/YR**	
Block	M	F	M	F	M	F	M	F	M	F	M	F	M	F	M	F	M	F	M	F	M	F	M	F	M	F	**M**	**F**
Woodlake	4	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	119	16
Ivanhoe	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	23	3
Exeter	0	0	0	0	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	26	2
South Exeter	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	1	0	0	0	13	4
Tonyville	0	0	0	0	0	0	0	0	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	27	4
W. Lindsay	3	2	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	34	10
Strathmore	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	14	1
Porterville	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	51	4
Terra Bella	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	9	0
Total	7	2	0	0	0	0	1	0	0	0	1	0	0	0	0	0	0	0	0	0	0	0	1	0	0	0	316	44
City of Visalia	3	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	16	3
	Sep.27		**Oct. 04**		**Oct. 11**		**Oct. 18**		**Oct. 25**		**Nov..01**																**TOT/YR**	
Block	M	F	M	F	M	F	M	F	M	F	M	F	M	F	M	F	M	F	M	F	M	F	M	F	M	F	**M**	**F**
Woodlake	0	0	0	0	0	0	1	0	1	0	0	0															121	16
Ivanhoe	0	0	0	0	0	0	0	0	0	0	0	0															23	3
Exeter	0	0	0	0	0	0	0	0	0	0	0	0															26	2
South Exeter	0	0	1	0	0	0	0	0	0	0	0	0															14	4
Tonyville	0	0	0	0	0	0	0	0	0	0	0	0															27	4
W. Lindsay	0	0	0	0	0	0	0	0	0	0	0	0															34	10
Strathmore	0	0	0	0	0	0	0	0	0	0	0	0															14	1
Porterville	1	0	0	0	0	0	0	0	0	0	0	0															52	4
Terra Bella	0	0	0	0	0	0	0	0	0	0	0	0															#REF!	#REF!
Total	1	0	1	0	0	0	1	0	1	0	0	0															320	44
City of Visalia	0	0	0	0	0	0	0	0	0	0	0	0															16	3

2024 Olive Fruit Fly Trapping Report

Date	11-Sep		18-Sep		25-Sep		2-Oct		9-Oct		*16-Oct*		22-Oct		30-Oct		6-Nov						Previous		Total	
Sex Of Fly	M	F	M	F	M	F	M	F	M	F	M	F	M	F	M	F	M	F	M	F	M	F	M	F		
Orland 1																										
County Road 15 and Papst Ave.	1	0	0	0	1	0	0	0	0	2	0	0	0	1	2	0	0	0					103	32	107	34
Orland 2																										
Road 200 and E	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0					49	19	49	19
Orland 3																										
SE of N and 16	0	0	0	1	0	0	0	0	0	1	0	1	1	0	0	0	0	0					23	10	24	13
Orland 4																										
NE of N and 12	0	0	0	0	0	0	0	0	0	0	0	0	0	0	1	0	0	0					8	7	9	7
Orland 5																										
Road 21 and M	0	0	0	0	0	0	0	0	1	0	0	1	2	1	0	0	1	0					25	3	29	5
Orland 6																										
Hwy 99W and Road 18	0	0	0	0	0	0	0	0	0	0	0	0	0	0	1	0	0	0					15	33	16	33
Corning 1																										
Northbound I-5 Rest Area	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0					99	37	99	37
Corning 2																										
Fig Lane and Houghton	0	0	2	0	4	0	4	0	5	1	1	0	3	0	0	0	0	0					392	110	411	111
Corning 3																										
Barham and Samson	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0					4	4	4	4
Corning 4																										
Sac River and Kopta Road	0	0	0	0	0	0	0	0	0	0	0	0	4	3	4	5	5	6					37	21	50	35
Corning 5																										
Viola and Orchard Ave.	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0					4	3	4	3
Corning 6																										
Dora and Marguerite	0	0	1	0	0	0	0	0	1	0	0	1	1	1	0	0	0	0					20	6	23	8
Totals	1	0	3	1	5	0	4	0	7	4	1	3	11	6	8	5	6	6					779	285	799	309

****INFORMATION ONLY****

FROM: COC RESEARCH SUBCOMMITTEE

SUBJECT: PRESENTATION OF 2025 PROPOSALS

BACKGROUND:

As a reminder, please see below 2025 research priorities that were established at the August 7, 2024 meeting.

California Olive Committee Research Priorities for 2025

- Olive Fruit Fly Trapping
- OFF management techniques with an emphasis on examining new traps that attract flies. Also, OFF management techniques focused on needing a membrane and a new delivery system.
- Management of Olive Knot with an emphasis on developing new methods of control via soil applications. Management of Olive Knot focused on systemic rather than topical treatments.
- Management of Peacock Spot
- Evaluation of drone technology and satellite mapping pertaining to moisture evaluation and crop load estimates
- Mechanical harvesting on existing and new high density orchards
- Development of Loosening Agents
- Olive DNA evaluation to distinguish between different varieties in the market place
- Chemical control of Glassy-winged sharpshooter/Leaf Hoppers
- Determine costs of preventative measures relating to the spread of xylella-fastidiosa (xf)
- Pollination assist techniques focused on Manzanillo/ Field measurement tool to determine optimal pollination timing
- Investigation of Urea as a thinning agent. What is the cost and optimal application rate? Where is it currently being used?
- Determine what research has already been conducted regarding sterilization of OFF
- Attract and Kill trap for Olive Fruit Fly
- Black Scale Control from a Natural Insect Pest
- GWSS research on mass trapping

The above highlighted priorities were discussed at length in August of 2024. Staff has the below updates.

1. Evaluation of Drone Technology and Satellite Mapping Pertaining to Moisture Evaluation and Crop Load Estimates:

Pat Ricchiuti alerted staff that this technology already exists with a company called AgMonitor. Below is a link to their website and the email for Olivier Jerphagnon with AgMonitor.

olivier@agmonitor.com

2. Attract and Kill trap for Olive Fruit Fly:

Information was provided earlier from Suterra.

3. Black Scale Control from a Natural Insect Pest:

Mike Silveira mentioned he had previously worked with a scientist at USDA-ARS Parlier who worked on this. Dr. Victoria (Vicky) Yokoyama retired a few years ago. COC Staff coordinated with USDA-ARS Parlier to receive a proposal pertaining to black scale.

4. GWSS research on mass trapping:

COC Staff reached out to David Haviland, who is extremely familiar with GWSS due to table grapes. He let us know that mass trapping has never been used nor is there any thought it would work. The yellow traps that are used are considered blunder traps. The yellow gives them a slight attractiveness, but they aren't really attractive (like a pheromone-based trap would be). As a result, the traps are not really effective and aren't good for anything more than general monitoring. The reality is that if you get 10 or 20 GWSS on a trap, there are hundreds, and possibly thousands, in the trees or vines around it. His estimate is that the numbers of traps that you would need to have an effective mass-trapping program would be astronomical and cost prohibitive.

Product Registrations

Product	Active Ingredient	Registrant	Status
Assail 70 WP	Acetamiprid	UPL	Subcommittee instructed staff to look into registering this for OFF. UPL is looking into it.
Assail 30 SG	Acetamiprid	UPL	Subcommittee instructed staff to look into registering this for OFF. UPL is looking into it.
Danitol 2.4 EC	Fenpropathrin	Valent	COC staff worked with Mike and Valent to put together a Section 2ee for GWSS control. This is scheduled for processing in Jan 2025. They will inform us when the label is live. Draft label is on the following page.
Kasugamycin	Kasumin	UPL	Staff submitted a letter requesting a concurrent review to the CDPR. This letter can be seen in the following pages.
Syllit	Dodine	UPL	Currently at CDPR.
Ph-D	Polyoxin D Zinc Salt	UPL	Currently at CDPR.

Other Projects

Reclassifying Olives as a Stone Fruit:

COC Staff reached out to the IR-4 program to determine the process and pros and cons of reclassification. IR-4 expressed that this would be a susbstantial undertaking and they were not sure of the feasibility. Olives currently exist as the rep crop for a crop group, so there would be significant efforts on the crop groups as a whole that they are not sure EPA would be willing to restructure. It is a rep crop for a US tropical fruit crop group and it is also a rep crop for Codex subgroup "Subgroup 005A, Assorted tropical and sub-tropical, edible peel-small." One of the goals of revising US crop groups was to harmonize the revised groups with the Codex Classification as much as possible. Since Codex is directly related to export, changing olives to a stone fruit would likely impact export markets for both olives and stone fruit, and likely not in a good way. Finally, they noted that since olives is currently a rep crop that is more likely to be associated with a residue study than as a member crop and thus implementation of the change to a stone fruit could adversely impact existing tolerances.

The IR-4 feedback did not shed light onto the process itself, so COC staff has reached out to the EPA to gain clarification.

Olive Fruit Fly-Elizabeth Fichtner:

Elizabeth assembled the historic olive fruit fly trapping data from the San Joaquin Valley into models. She concluded that the highest populations were observed in 2018. She provided her findings in the following pages. She also submitted a research proposal expanding upon this work.

FIFRA Section 2(ee) Recommendation

THIS RECOMMENDATION IS ONLY FOR USE IN THE STATES OF CALIFORNIA

DANITOL® 2.4 EC SPRAY

EPA Reg. No. 59639-35

DANITOL INSECTICIDE FOR USE ON GLASSY-WINGED SHARPSHOOTER IN OLIVE

This FIFRA Section 2(ee) Recommendation Expires December 31, 2028

KEEP OUT OF REACH OF CHILDREN
WARNING

DIRECTIONS FOR USE

It is a violation of Federal law to use this product in a manner inconsistent with its labeling.

This recommendation is made as permitted by Section 2(ee) of FIFRA, as amended, and has not been submitted to or approved by the U.S. Environmental Protection Agency. This 2(ee) recommendation contains directions for use which do not appear on the package label and must be in the possession of the user at the time of pesticide application. Follow all applicable directions, restrictions, Worker Protection Standard requirements, and precautions on the EPA registered label when using Belay Insecticide. Always read and follow all label directions when using any pesticide alone or in tank mix combinations. The most restrictive labeling applied when using a tank mix.

DANITOL INSECTICIDE APPLICATION TO OLIVE

<table>
<tr><th colspan="3">FOLIAR APPLICATION</th></tr>
<tr><th>PEST</th><th>APPLICATION RATE</th><th>APPLICATION INSTRUCTIONS</th></tr>
<tr><td>Glassy-Winged Sharpshooter</td><td>10-2/3 - 16
(0.2 - 0.3 lb ai/A)</td><td>Apply as a ground application in a minimum of 100 gallons of water per acre or apply by air in a minimum of 10 gallons of water per acre.
Make applications beginning when first pest activity is noticed and repeat as needed but not more often than every 14 days.
Under severe pest pressure use the higher recommended rate.</td></tr>
<tr><td colspan="3">• Do not apply within seven (7) days of harvest.
• Do not exceed 2-2/3 pt (42-2/3 fl oz, 0.8 lb ai) total application of Danitol 2.4 EC Spray per acre per season.</td></tr>
</table>

Refer to *Danitol* Insecticide label for additional insects controlled.

Registered and Manufactured by: Valent U.S.A. LLC P.O. Box 5075 San Ramon, CA 94583
Belay is a registered trademark of Sumitomo Chemical Company, Ltd..

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November 8, 2024

Mr. Tulio Macedo, Branch Chief
Pesticide Registration Branch
Department of Pesticide Regulation
1001 I Street, P.O. Box 4015
Sacramento, CA 95812-4015

Dear Branch Chief Macedo:

This letter is to request a concurrent review by the California Department of Pesticide Regulation (CDPR) for the UPL bactericide kasugamycin (commercially known as Kasumin Reg No. 66330-404), for use on olives in California. The California Olive Committee (COC) is comprised of two canneries and nearly a thousand growers who are responsible for producing 95% of olives grown in the United States. One of the primary objectives of the COC is to facilitate research projects and product registrations that benefit the industry. The California ripe table olive industry produces on average, 50,000 tons of olives per year with the majority of acreage located in Glenn, Tehama, and Tulare counties. The industry continues to expand as "modern acreage" that can be mechanically harvested is planted into the ground. This transition and addition in acreage will not only help the industry be more efficient but will also increase sustainable practices.

The United States Environmental Protection agency has completed the new 'framework' for registering bactericides this fall and will begin the federal registration process for Kasumin on olives. This is a critical registration for the olive industry because alternative bactericides are needed for the management of olive knot caused by *Pseudomonas syringae* pv. *savastanoi*. The detection of copper-resistance in localized populations of the pathogen makes the registration of alternatives imperative. This allows for mixtures or rotations of products with copper to prevent further selection of copper resistance in different geographical areas and/or the increase of resistance levels. Both types of olives are susceptible to olive knot. The pathogen enters through wounds that are especially created by mechanical harvesters. The proposed use pattern of kasugamycin is for applications immediately after harvest in the fall, during the dormant period (winter), and early spring when leaf drop occurs. Thus, the crop will not be treated during the growing season from bloom to harvest, and fruit residues are expected to be undetectable as determined by IR-4 GLP studies completed in May 2020. The registration petition was submitted to EPA in late 2020 after 6 years through the IR-4 program. Currently, no other effective alternatives for copper are available.

Kasugamycin is registered as Kasumin in California on pome fruits, sweet cherry, and walnut, and there is an ongoing emergency registration (Section 18) for use on almonds. Thus, CDPR has evaluated Kasumin for these registrations and should be familiar with this compound. Counting the IR-4 studies and the EPA delays due to COVID and ESA (Endangered Species Act), it has been over 10 years. Thus, our request for a concurrent review is justified in hopes of a registration by 2026.

In summary, because the bactericide Kasumin is federally registered on several crops (pome fruits, sweet cherry, and walnut) in California and resistant strains of the olive knot pathogen *P. syringae* pv. *savastanoi* have been detected, we are requesting a concurrent review by CA-DPR with the US-EPA for Kasumin use on olive. Registration of Kasumin for after harvest use and before bloom at leaf drop will prevent the selection of resistant sub-populations of the target pathogen by using mixtures or rotations with copper. As with fungicides, the availability of different modes of action is the best way to fight the selection of resistant sub-populations of pathogens in agriculture and to minimize the use of any one product in the environment. Kasumin is strictly a plant agriculture bactericide with a unique mode of action that is not used in animal or human medicine.

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Thank you for your time and attention to this request. If you have any questions, my e-mail address is eoliver@calolive.org and my cell phone no. is 559-575-4884.

Sincerely,

Elise Oliver
Director of Operations
California Olive Committee

Machine learning predictive model for estimating the future population dynamics of olive fruit fly, *Bactrocera oleae*

Santosh Bhandari[1], James Stewart[2], Bert Quezada[2], Elizabeth Fichtner[3]
[1]Assistant Specialist, UCANR, [2]Crop Consultant, Ag IPM Consultants, Inc., [3]Farm Advisor, UCANR

Abstract

In this study, we used Random Forest Classifier (RFC), a commonly used supervised machine-learning model, for training on population data of olive fruit fly, *Bactrocera* oleae, (OFF) collected in the southern San Joaquin Valley (SSJV) of California from 2019 to 2023. The model, when trained, detects specific patterns from the past and responds to new entries based on the developed trends. Eighty percent of the total data volume was used for training and 20% was used for testing the efficiency of the model. For the deployment, predictions were made on unseen data and Google Cloud Platform (GCP) was used to deploy the web application for convenient access to growers. Estimating the presence or absence of OFF based on forecasted weather parameters can be an effective tool for making data-driven decisions for pest management, thus reducing insecticide applications and improving the economic sustainability of olive cultivation in California.

Introduction

Olive fruit fly (OFF) was first reported in United States in 1998 and has since become an established, economic pest of olives. In response to the introduction of OFF, the California Olive Committee (COC) has funded pest monitoring programs for the pest in table olive growing regions of the Sacramento Valley and southern San Joaquin Valley since 2001. The goal of these OFF-monitoring programs was to provide growers with data of regional pest activity, thus facilitating population-based pest management (Johnson et al. 2006).

The goal of the current study was to utilize the historical OFF population data to create a model that can predict future OFF populations. Numerous studies have demonstrated that OFF infestation levels vary based upon weather parameters including mean daily temperature, precipitation, and relative humidity (Yokoyama et al. 2008, Wang et al. 2009 a, b). In this study, we created a preliminary model, based on 5 years of OFF population and weather data, that can predict presence or absence of OFF at any time based on the developed trends.

Figure 1. Olive fruit fly adult (Photo: M. Johnson)

Figure 2. Adult female olive fruit fly oviposits the fruit

Figure 3. Larvae of olive fruit fly (Photo: M. Johnson)

Research Objective

- Utilize historic OFF trapping data and corresponding weather data to develop a predictive model for determining future OFF populations.

Materials and Methods

Initial steps include framing the problem, collecting the data, and preprocessing the data. Data preprocessing prepares the data for the analysis and/or modeling through sequential steps such as data quality assessment, data cleaning, data transformation, and data reduction. After preprocessing, exploratory data analyses can be performed to derive insights from the data. Similarly, feature analysis is used to determine whether a variable is categorical or numerical, and which variables should be utilized for mathematical operations. Next, feature engineering is implemented to derive new features from the existing features and to improve accuracy on unseen data.

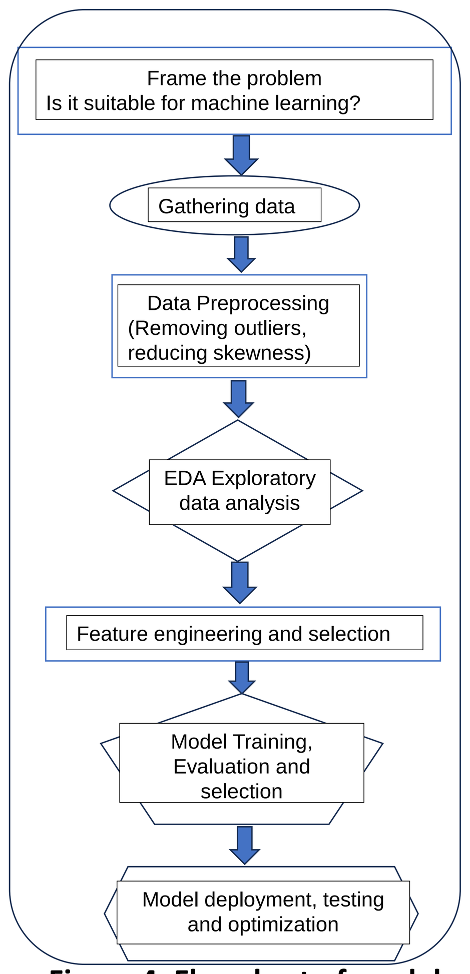


Figure 4. Flowchart of model development protocol

Software and packages

We used Python to develop the model, with Scikit-learn handling data preprocessing, model building, and evaluation. Flask was used to create an API for easy integration with other services. The application was deployed on Google Cloud Platform for scalability and reliability. JupyterLab from the Anaconda distribution was the development environment where we wrote, tested and debugged the code in a Jupyter notebook.

Feature Engineering and Selection

Features	Importance
Weekly avg. wind speed (mph)	0.2663
Weekly avg. soil temp (F)	0.2427
Weekly average temp (F)	0.2268
Weekly average RH (%)	0.2017
Weekly average prep (inch)	0.0625

Data preprocessing and cleaning

- No missing values were found in the dataset
- Twelve potential outliers were detected and removed
- The skewness of male and female OFF was reduced through log transformation and adjusted for the total population as final output prior to analysis

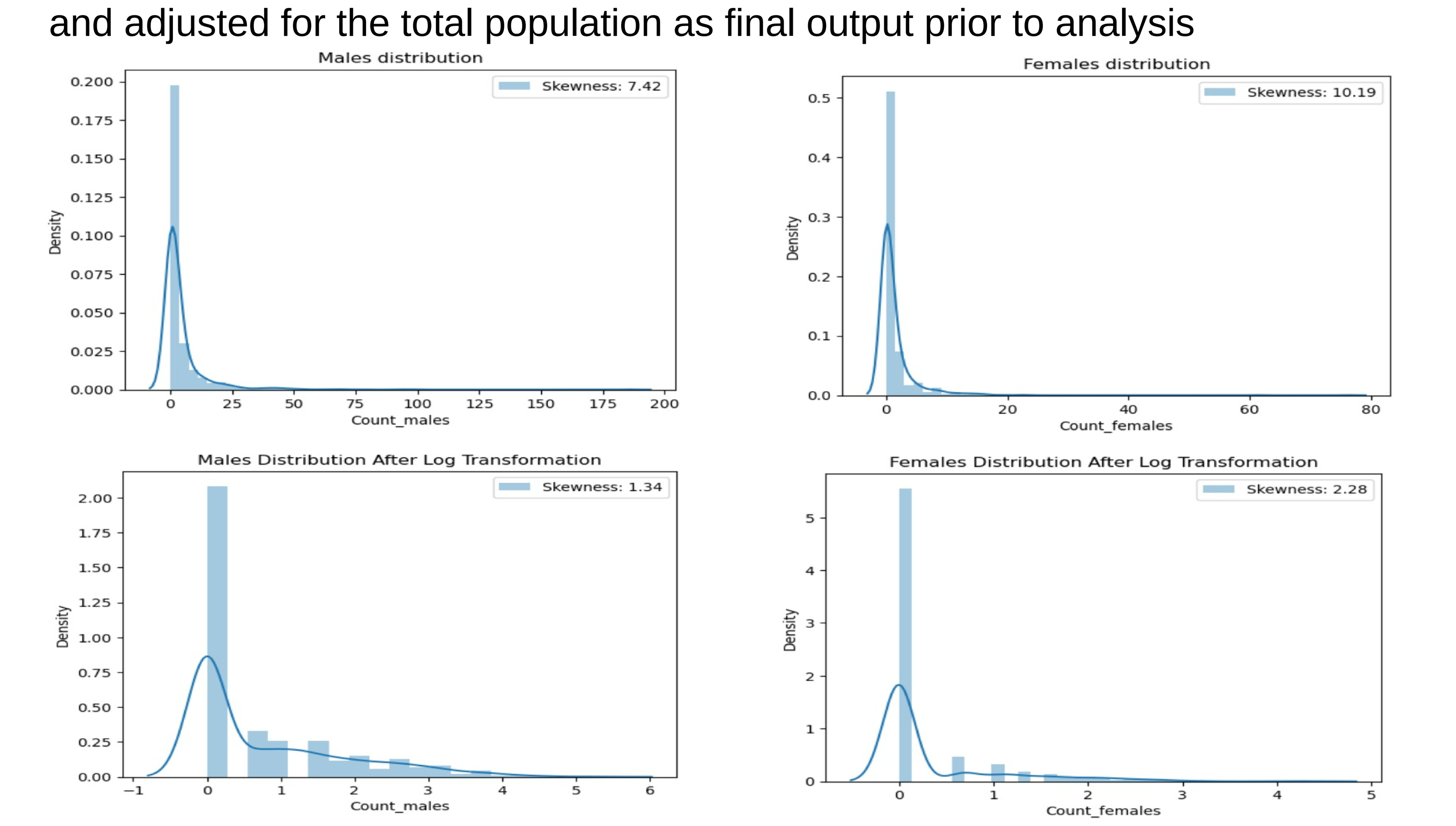


Fig 5. Data pre-processing (Log transformation to reduce the skewness of the data)

Exploratory Data Analysis

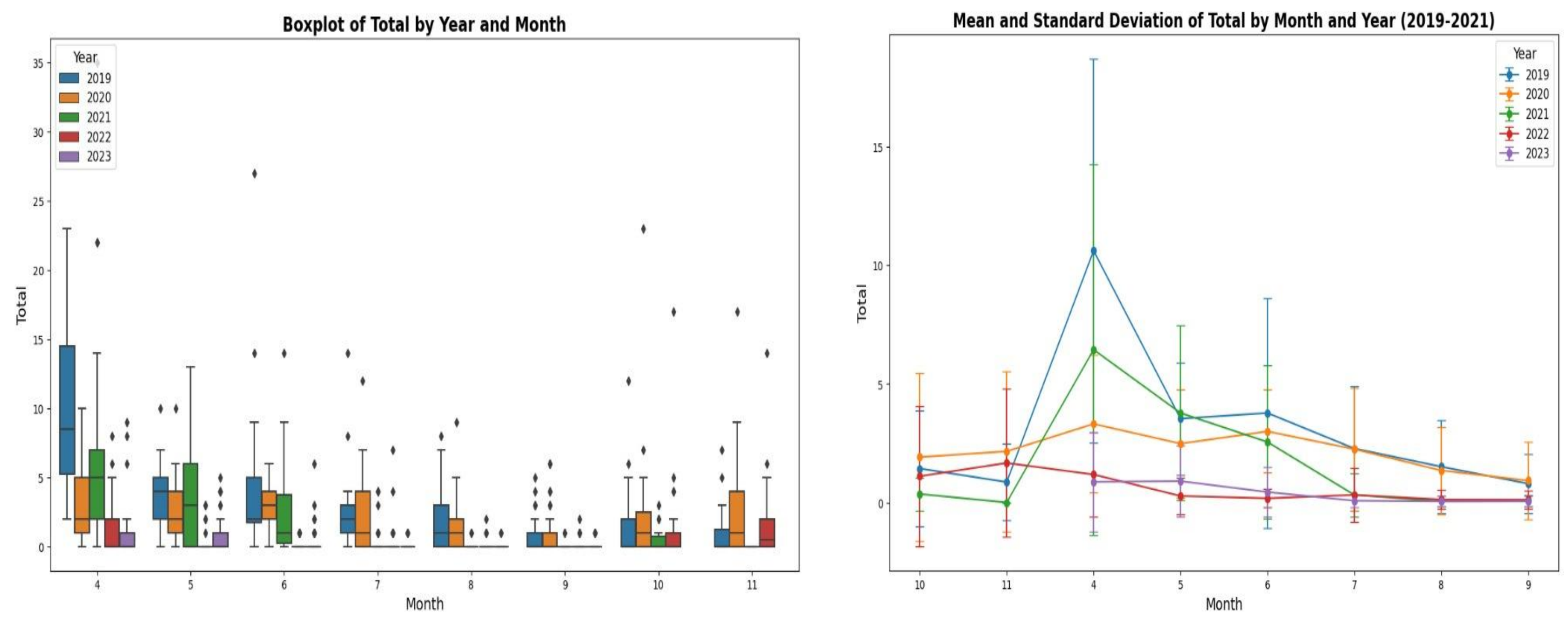


Fig.6. Boxplot and summary graph showing the distribution of total population of olive fruit fly in different months and years

Training, cross-validation, and testing dataset

- Different models (random forest, logistic regression, support vector machine, gradient booster, xGBooster) were tested for their efficiency in predicting the new values
- 80% of the dataset was used for training and 20% for testing and cross-validation
- Random Forest model showed the greatest efficiency with the following metrics

Metrics	Percentage
Accuracy	70.6%
Precision	74.8%
Recall	73.5%
F1 score	74.1%

Hyperparameter tuning

Hyperparameter tuning improves the efficiency of the model used for training

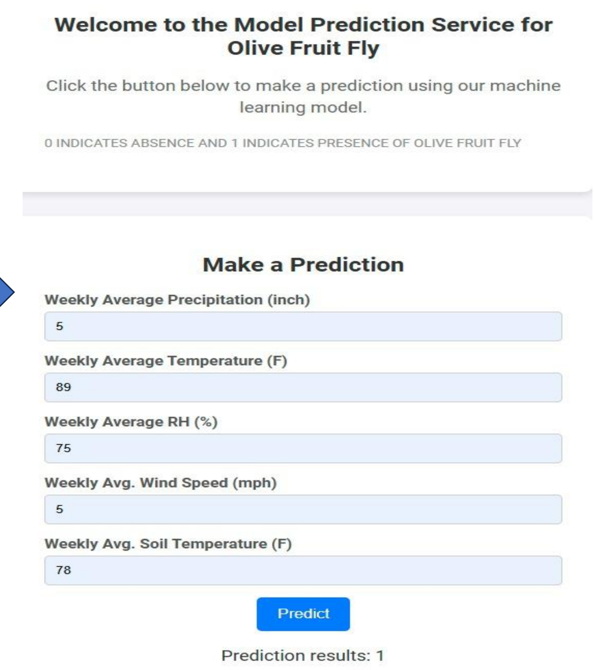
Parameter grids	Values
n_estimators	[100, 200, 300]
Max_depth	[None, 10, 20, 30]
Min_samples_split	[2, 5, 10]
Min_samples_leaf	[1, 2, 4]
Bootscrap	[True, False]

Metrics	Percentage
Accuracy	70.6%
Precision	73.9%
Recall	75.2%
F1 score	74.5%

Preliminary model deployment

Scan me for predicting olive fruit fly

Link:
https://off-prediction.wl.r.appspot.com



Note: The features selected for model deployment are not conclusive and are part of a grant proposal; this is merely a demonstration of how the model would function with the selected features under certain conditions.

Findings and Discussion

- The population trend shows that the number of OFF in traps peaks from April to June and then decreases.
- The feature selection indicates that wind speed and temperature (soil and air temperature) have high importance in developing the prediction model; precipitation had the least importance.
- The efficiency matrices highly depend on the total variance of the model (Lesser the variance, higher is the efficiency of the model)
- The weather data was retrieved from https://cimis.water.ca.gov to the nearest weather station from the olive sites, and does not represent the actual micro-weather conditions on those sites
- The number of OFF adults in traps indicate the field representation, however zero counts does not always indicate true absence of the pest
- Future research includes installation of data loggers at trapping sites to record real-time micro-climate data for use in the model.

References

Johnson, M., Zalom, F., Steenwyk, R., Kicenik Devarenne, A., Daane, K., Krueger, W., Connell, J., Yokoyama, V., Bisabri, B., Caprile, J., & Nelson, J. (2006). Olive Fruit Fly Management Guidelines for 2006. *UC Plant Protection Quarterly*, *16*, 1–7.

Yokoyama, V. Y., Rendón, P. A., & Sivinski, J. (2008). Psyttalia cf. c oncolor (Hymenoptera: Braconidae) for Biological Control of Olive Fruit Fly (Diptera: Tephritidae) in California. *Environmental Entomology*, *37*(3), 764–773. https://doi.org/10.1603/0046-225X(2008)37[764:PCHBF]2.0.CO;2

Wang, X.-G., Johnson, M. W., Daane, K. M., & Nadel, H. (2009a). High Summer Temperatures Affect the Survival and Reproduction of Olive Fruit Fly (Diptera: Tephritidae). *Environmental Entomology*, *38*(5), 1496–1504. https://doi.org/10.1603/022.038.0518

Wang, X.-G., Johnson, M. W., Daane, K. M., & Opp, S. (2009b). Combined Effects of Heat Stress and Food Supply on Flight Performance of Olive Fruit Fly (Diptera: Tephritidae). *Annals of the Entomological Society of America*, *102*(4), 727–734. https://doi.org/10.1603/008.102.0418

Anaconda Software Distribution. (2020). *Anaconda Documentation*. Anaconda Inc. Retrieved from https://docs.anaconda.com/

Google Cloud (console.cloud.google.com)

Acknowledgement

We thank the California Olive Committee for the support of the OFF trapping program and to California Irrigation Management System (CIMIS) for access to historic weather data. We also appreciate the growers who have provided orchard access for the OFF trapping program.

UNIVERSITY OF CALIFORNIA
Agriculture and Natural Resources

2025 Research Proposals

#	Researcher	Project	Amount
1	Dr. Jim Adaskaveg*	Epidemiology and management of olive knot caused by Pseudomonas savastanoi pv. Savastanoi (year 1)—**page 53**	$15,490
2	Rodrigo Almeida*	Survey of Xylella fastidiosa genetic diversity within California olive trees (year 2)—**page 58**	$30,361
3	Georgia Drakakaki and Becky Wheeler-Dykes*	Evaluation of effects of Accede® (ACC) at two different application rates on enhancing horticultural maturity and abscission zone development and commercial trunk shaking efficiency in table olives (year 3)—**page 71**	$115,129.20
4	Dr. Carol Lovatt and Elizabeth Fichtner*	Integrating Alternate Bearing Mitigation Strategies in a Commercial Table Olive Orchard (year 3)—**page 86**	$29,156
5	Elizabeth Fichtner*	Designing a web app for predicting risk of olive fruit fly—a tool for California olive growers and pest control advisors—**page 104**	$19,860
6	Franklin Lewis*	Development of screening tools to determine Xylella fastidiosa tolerance in olives—**page 114**	$10,443
7	Joel Kimmelshue-Land IQ	Olive Acreage Mapping—**page 121**	$30,700
8	Nicholas Manoukis*	New prospects for the control of black scale in California olive groves—**page 128**	$73,000
9	R.A. Van Steenwyk*	Improved monitoring of olive fruit fly through gamma-hexalactone lures and improved attract-and-kill control of olive fruit fly through the combination of gamma hexalactone and GF-120.—**page 189**	$66,596
10	Ernie Simpson	Sacramento Valley Olive Fruit Fly Monitoring Project—**page 195**	$12,500
11	Jim Stewart	Southern San Joaquin Valley Olive Fruit Fly Monitoring Project—**page 196**	$12,000
		Contingency Fund	
		Total	**$415,235.20**

*Presentations for all proposals can be downloaded here in the WeTransfer Link!
Land IQ, Ernie Simpson, and Jim Stewart did not provide video presentations for their projects as acreage mapping and trapping are very straight forward.

Downloadable WeTransfer Link:
https://we.tl/t-8I7JkobPVh

Written Proposals can be seen in the following pages of the packet.

AES/CE MAR 84 Workgroup: Olive

Department: Plant Pathology/UCR

University of California
Division of Agricultural Sciences
PROJECT PLAN/RESEARCH GRANT PROPOSAL

Project Year: 2025 Anticipated Duration of Project: 1st of 3 years

Principal Investigators: J. E. Adaskaveg

Cooperating: L. Solari and H. Förster

Project Title: Epidemiology and management of olive knot caused by *Pseudomonas savastanoi* pv. *savastanoi*

Keywords: Bactericides, copper enhancing compounds, antimicrobial natural products, biological controls

JUSTIFICATION/ BACKGROUND

Pseudomonas savastanoi pv. *savastanoi* (*Psv*), the causal agent of olive knot, is a serious disease of olives (*Olea europaea*) throughout all growing regions of the world (8). The pathogen enters through wounds causing outgrowths (knots, tumors, galls) mostly on trunks, branches, and twigs. Olive knot is the most economically important disease of olives. Infection may lead to tree defoliation, dieback, and reduced vigor, which ultimately lowers fruit yield and quality (6), and the disease may lead to orchard failure. *Psv* can survive epiphytically on olives, but the main source of inoculum is bacteria living within knots (7). Large quantities of bacterial ooze can be exuded when knots become wet (12). This exudate is disseminated by rain, wind, insects, birds, and human orchard activity. The opportunistic pathogen invades wounds caused by natural leaf abscission (4), frost, hail, or cultural practices such as pruning and harvesting. Sodium hypochlorite or quaternary ammonia that was registered under a special local need (Section 24c) registration based on efforts from this project can be used to sanitize field equipment and minimize the spread of the pathogen during harvest and pruning operations (10). After entering its woody host, the pathogen induces knot formation through the production of indoleacetic acid (IAA) and cytokinins (2). In California, infections occur mostly during the rainy season (late fall to spring) but knots do not develop until new growth starts in the spring. Infections can occur at low temperatures (-5°C) and thus, wetness is the main limiting factor for the disease. None of the currently grown olive cultivars is resistant to the pathogen (5).

Control of olive knot is difficult, and growers rely on applications of copper-based bactericides as the only effective foliar treatment. Manual application of cresol- and xylenol-based compounds (Gallex) to individual knots can eliminate the pathogen but is unfeasible on a commercial scale because this treatment is phytotoxic when applied as an air-blast foliar treatment and must be applied directly to the knots. Copper has been extensively used in olive production for many years for the control of peacock spot and olive knot. Reliance on a single active ingredient has led to our detection of copper resistance in *Psv* strains in some commercial olive orchards although the incidence of copper resistance is currently very low, accounting for only 2% of the total strains collected in surveys of olive growing regions of California. When resistant strains were inoculated to Arbequina and Manzanillo olive wounds, application of copper had reduced or provided no control as compared to inoculation with a sensitive strain. Therefore, there is a potential risk of copper resistance spreading with the continued and sole use of copper. Additionally, copper use is becoming more regulated due to accumulation in the soil and water sheds. This necessitates the development of new bactericides with different modes of action or of copper-activity-enhancing materials to overcome resistance. Copper products with lower MCE such as CS-2005, Cueva, or Instill are less effective. In walnut blight management where copper resistance in *Xanthomonas arboricola* pv. *juglandis* is common, copper-mancozeb mixtures have provided exceptional control for many years, but mancozeb can no longer be registered on new crops. Therefore, other alternatives need to be evaluated.

We have been instrumental in the development of the new agricultural antibiotic kasugamycin (Kasumin) for managing bacterial diseases of several agronomic crops in the United States. Kasugamycin has high activity against *Erwinia* (1) and *Pseudomonas* (11) and moderate activity against *Xanthomonas* species and other plant pathogenic bacteria. In our studies, we found it to be the most effective new treatment for preventing olive knot on naturally formed leaf scars and wounds created by hedging or harvesting (11).

Kasugamycin was first federally registered on pome fruits, followed by cherry and walnut. Registrations on almonds, peaches, and olives are pending. The olive submission of kasugamycin to the EPA was delayed because new GLP residue trials and laboratory analyses had to done that were completed in 2019. An IR-4 petition was prepared in 2020 and finally submitted in the summer of 2021. With EPA's new protocols revised in 2024, kasugamycin registrations are still pending, but we requested a concurrent review between CDPR and EPA. Kasugamycin would greatly complement current copper sprays and could be used in rotation or mixtures with copper and other bactericides that we recently identified. A second antibiotic, oxytetracycline, is also being pursued for registration on olives through the IR-4 program. The IR-4 petition for oxytetracycline was submitted to EPA in late summer 2020 and the registration was scheduled for Oct. 2021, but it is now still pending. Most likely new requirements will be required for registering bactericides and fungicides that are medically important under the new EPA protocols.

The fungicide dodine (commercial name Syllit) has bactericidal properties when used at labeled rates, and this was confirmed in our studies. In the last three years we demonstrated that the addition of dodine to kasugamycin or oxytetracycline improved the efficacy against olive knot. Since dodine is being registered for peacock spot where it shows high efficacy, it will be available for use in combination with other antimicrobials (i.e., copper, antibiotics, and food-grade bactericides) once registered. Copper-dodine mixtures are also being developed for walnut.

Another strategy is the use of natural products. We evaluated several of these over the last two years with inconsistent but high success in some trials. We attributed this to using unformulated active ingredients that do not have adjuvants to stabilize and prevent degradation. Nisin and ε-poly-L-lysine (EPL) are antibacterial food preservatives that are FDA 'generally recognized as safe' (GRAS) and are naturally produced by gram-positive *Streptomyces* species. Products based on natural oils such as Guarda (thyme oil) or Seican (cinnamaldehyde), organic acids including the commercial product Dart (capric/caprylic acids) have high in vitro toxicity and have also shown promise in our field evaluations but are inconsistent. Two potential registrants are working with us to develop agricultural formulations of nisin and EPL (KFD-622 and 623) or mixtures of EPL and cinnamaldehyde (Seican) that may be more stable in the environment. These registrants will support EPA biopesticide registrations. UPL is also supporting the dodine (and the polyoxin-D) registration on olive for peacock spot. Other products that we successfully evaluated in studies on walnut blight or fire blight are an extract of *Acacia mearnsii* (QAM 8921), Curezin (a copper-phosphite mixture), and a new sanitizer product (Virus Shield) that should be evaluated for managing olive knot. Integration of these alternative materials in rotation or in mixtures with copper or potentially with registered antibiotic treatments may provide consistent high disease control, reduce the risk of resistance development, and provide olive growers with more resources for managing olive knot.

Therefore, we are working to formulate nisin and EPL as part of an ongoing process to develop new GRAS bactericides for olive knot control, and we are identifying new bactericides and sanitizers that may be used for managing olive knot. Potential strategies for optimizing these compounds include reformulation with acids or adjuvants (9) to prevent photo- or UV-degradation, and mixtures with other natural products, bactericides (e.g., kasugamycin) or fungicides (e.g., dodine). UPL has provided us an organic formulation of kasugamycin that also needs to be evaluated for olive knot. This information is very valuable because rotational programs could be developed with different modes of action for different phases of the disease, i.e., leaf scars during leaf drop and twig wounds occurring during harvest and pruning. These materials are potentially registerable as conventional biopesticides and possibly as organic treatments. Treatments proposed to be evaluated in olive knot studies are listed in Table 1.

RESEARCH OBJECTIVES

1) **Evaluate new bactericides against *Psv*: GRAS food additives, natural products, and other experimentals** (see Table 1)
   a) Laboratory in vitro sensitivity studies: plant extracts, nisin and Ɛ-poly-L-lysine (EPL) formulations mixed with cinnamaldehyde or capric/caprylic acids (Dart).
   b) Field efficacy studies with new bactericides in comparison with kasugamycin - copper mixtures for the management of olive knot.
      i) Food preservatives (EPL, nisin) and formulated natural products (Seican-cinnamaldehyde)

ii) Sanitizers (VirusShield – a stable chlorine dioxide formulation) and Curezin (copper-phosphite) as potential novel strategies.

2) Continue to support the registration of the antibiotics kasugamycin and oxytetracycline

a) We continue to support these bactericides, but the date of registration is unknown.

Table 1. Treatments proposed for evaluation in olive knot studies

Compound	Active ingredient	Compound
Organic Kasumin	kasugamycin	Naturally produced antibiotic
Seican + EPL Seican + nisin	Cinnamaldehyde + EPL Cinnamaldehyde + nisin	Plant extract + food preservative Plant extract + food preservative
Seican+EPL + Manniplex-Zn	Cinnamaldehyde + EPL+Zn	Plant extract + food preservative + Zn
Guarda mixed with EPL	thyme oil	Plant extract
QAM 8921	*Acacia mearnsii*	Plant extract
Virus Shield*	Chlorine dioxide	Sanitizer
Curezin*	copper + phosphite	Metallic ion + acidifier
Champ + Syllit	Copper + dodine	Metallic ion + fungicide/bactericide

* - Pending receipt of product.

PLANS AND PROCEDURES

1) Evaluate new bactericides, food additives, GRAS sanitizers, and other experimentals against *Psv*.
a. To evaluate the in vitro toxicity of new bactericides, the spiral gradient endpoint (SGE) or agar dilution methods will be used. Hereby bacterial strains are exposed to a bactericide concentration gradient on a single agar plate or to selected single concentrations, respectively. This will allow the determination of minimal inhibitory values for *Psv* of products being evaluated that will help to optimize formulations and calculate appropriate field rates.

b. Field studies will be done on Arbequina and Manzanillo olives in established orchards at UC Davis and commercial sites. Lateral wounds on 1-2-year-old twigs will be made using a scalpel by removing the bark and exposing cambial tissue. Leaf scars will be made by pulling leaves off the same twigs. In addition, wounds from natural leaf drop in the spring will be used. Treatments as listed in Table 1 will be sprayed onto wounds, allowed to air-dry, and inoculations will be done with a suspension of copper-sensitive or -resistant *Psv* strains. The efficacy of treatments will be assessed as the percent incidence of knots forming on treated, inoculated wounds as compared to wounds that are treated with water and inoculated (i.e., controls).

2. Continue to support the registration of the antibiotics kasugamycin and oxytetracycline, as well as dodine. Registration of oxytetracycline (FireLine) is proceeding with EPA, and the registrant is expecting full registration. EPA, however, delayed the review of oxytetracycline due to COVID-19, new ESA requirements, retirements within the agency, and new protocols for medically important products (i.e., oxytetracycline) resulting in multiple PRIA date changes. Registration requests generally take 18 months to review and the federal review most likely will not be completed until 2026.

Benefits to the industry

For management of olive knot, in addition to cultural methods, sanitation practices, and the labor-intensive Gallex, only copper materials and the natural product Regalia are currently available. We determined that applications have to be made within 24 h of wounding events (e.g., harvesting, pruning, hail storms, freezing) for best efficacy, and we demonstrated that high labeled rates of copper often outperform low rates. We showed that copper resistance in *Psv* is currently found in only a few locations where copper has been used for many years. Because copper-resistant strains of *Psv* were found to be virulent and competitive, and because they were not genetically clonal, there is a risk of further development and spread of copper resistance. Therefore, alternatives are needed for a sustainable and effective management program for many years ahead. We initiated the registration of the agricultural antibiotic kasugamycin that was registered in 2014 on pome fruits, and in 2018 on cherry and walnuts in California. Registrations are pending on almonds, peaches, and olives. Oxytetracycline for use on olive went through the IR-4 program, has been submitted to the EPA in 2020, and registration is also pending. Kasugamycin and oxytetracycline showed high activity against olive knot especially in mixtures with copper and dodine. These are exciting developments for the olive industry.

We will continue to explore and evaluate other potential bactericide products that include the natural GRAS products nisin and EPL in mixtures with cinnamaldehyde (i.e., Seican), the sanitizer chlorine dioxide, and copper-potassium phosphite. Nisin and EPL are being commercially formulated in cooperation with UPL (the registrant of dodine and kasugamycin) and in combination with cinnamaldehyde in cooperation with Summit Agro. We will also continue to evaluate other new natural products such as thyme oil formulations for which we obtained preliminary promising but inconsistent results. The registration of several materials for olive knot management will allow the implementation of anti-resistance strategies and will prevent over-use of any single mode-of-action bactericide. Still, integrated practices will be critical for the successful management of the disease. Any bactericide treatment will be most effective when pathogen population levels are at a minimum, and the host is less susceptible.

References

1. Adaskaveg, J.E., Förster, H., and Wade, M.L. 2011. Effectiveness of kasugamycin against *Erwinia amylovora* and its potential use for managing fire blight of pear. Plant Dis. 95:448-454.
2. Comai, L., and Kosuge, T. 1980. Involvement of plasmid deoxyribonucleic acid in indoleacetic acid synthesis in *Pseudomonas savastanoi*. J. Bacteriol. 143: 950-957.
3. Edgecomb, D.W. and Manker, D. 2005. *Bacillus subtilis* strain QST 713, bacterial disease control in fruit, vegetable and ornamental production. Mitteilungen-Biologische Bundesanstalt für Land und Forstwirtschaft 408:167-169.
4. Hewitt, W. B. 1939. Leaf scar infection in relation to the olive knot disease. Hilgardia 12:41-66.
5. Penyalver, R., García, A., Ferrer, A., Bertolini, E., Quesada, J.M., Salcedo, C.I., Piquer, J., Pérez-Panadés, J., Carbonell, E.A., del Río, C., Caballero, J.M., López, M.M., 2006. Factors affecting *Pseudomonas savastanoi* pv. *savastanoi* plant inoculations and their use for evaluation of olive cultivar susceptibility. Phytopathology 96:313–319.
6. Schroth, M.N., 1973. Quantitative assessment of the effect of the olive knot disease on olive yield and quality. Phytopathology 63:1064.
7. Wilson, E. E. 1935. The olive knot disease: Its inception, development, and control. Hilgardia 9:233-264.
8. Young, J.M., 2004. Olive knot and its pathogens. Australasian Plant Pathology 33:33–39. doi:10.1071/AP03074
9. Chheda, A. H., and Vernekar, M. R. 2015. A natural preservative epsilon-poly-L-lysine: fermentative production and applications in food industry. Intern. Food Research J. 22:23-30.
10. Nguyen, K., Förster, H., and Adaskaveg, J.E. 2017. Quaternary ammonium compounds as new sanitizers for reducing the spread of the olive knot pathogen on orchard equipment. Plant Dis. 101:1188-1193.
11. Nguyen, K. A., Förster, H., Adaskaveg, J.E. 2018. Efficacy of copper and new bactericides for managing olive knot in California. Plant Disease 102:892-898.
12. Nguyen, K. A., Förster, H., Adaskaveg, J.E. 2018. Genetic diversity of *Pseudomonas savastanoi* pv. *savastanoi* in California and characterization of epidemiological factors for olive knot development. Plant Disease 102:1718-1724.

Budget Request:
Funding Source: California Olive Commission and California Olive Oil Commission

Budget Request with UC indirect costs:

Budget Year: 2025 Funding Source*:	**OOC**	**COC**	**Total Budget**
Salaries and Benefits: SRA/Postdoc	3,500	3,000	6,500
Lab/Field Ass't	0	0	0
Subtotal	3,500	3,000	6,500
Employees' Benefits**	1,500	1,500	3,000
Subtotal	5,000	4,500	9,500
Supplies and Expenses	0	0	0
University Land and Orchard charges	1,000	1,000	2,000
Operating Expenses/Equipment Travel	0	0	0
Travel	1,500	1,500	3,000
Direct Cost Totals Total	**$7,500**	**$7,000**	**$14,500**
Off Campus IDC @ 11%		$990	$990
Total Budget Requested	**$7,500**	**$7,990**	**$15,490**

J. E. Adaskaveg
________________________________ Date: Nov. 5, 2024
Originator's Signature (PI)

Dept. Chair Katherine Borkovich ________________________________ Date: Nov. 5, 2024
(Riverside Campus)

Liaison Officer ________________________________ Date:____________

UNIVERSITY OF CALIFORNIA, BERKELEY

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October 30, 2024

Elise Oliver
Director of Operations and Research
California Olive Committee
2565 Alluvial Ave., Ste. 152
Clovis, CA 93611

RE: Project Title: Survey of Xylella fastidiosa Genetic Diversity Within California Olive Trees
Principal Investigator: Rodrigo Almeida, Ph.D.
Amount of Proposed Funding: $30,361

Dear Elise:

Please find attached the proposal for the California Olive Committee titled "Survey of Xylella fastidiosa Genetic Diversity Within California Olive Trees" on behalf of Professor Rodrigo Almeida of UC Berkeley. I have reviewed and approved the work and budget outlined in the proposal. If you need any additional information, please feel free to contact me.

Best regards,

Anissa Jones
Industry Liaison and Contracts Manager
Industry Alliances Office

Project title
Survey of *Xylella fastidiosa* genetic diversity within California olive trees

Principal Investigator (PI)
Rodrigo P.P. Almeida
Professor & Hildebrand-Laumeister Chair in Plant Pathology
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Rausser College of Natural Resources, University of California, Berkeley; Berkeley, CA, 94720
e-mail: rodrigoalmeida@berkeley.edu

Objective(s) of Proposed Research:
This proposed project directly addresses the research priorities associated with the risks of *Xylella fastidiosa* disease to California olive production. Our objectives are as follows:

Objective 1 - Evaluate *X. fastidiosa* genetic diversity in olive trees in Southern California.

Objective 2 - Identify environmental variables associated with high *X. fastidiosa* diversity to inform future risk assessments.

This research was initiated in 2024 with COC funding for one year. Field work started in the summer as pathogen populations within plants are higher at that time of the year, and disease symptoms are more severe. We collected more samples from olive trees for testing than originally proposed, and over one-third of those were positive for *X. fastidiosa*. We are now working on genotyping the positive samples, as described in the original proposal; the project is moving forward as proposed. The goal for this second year is to expand the area sampled so that a representative sampling of the *X.* fastidiosa-olive disease problem in California is obtained, and to initiate work on possible biotic and abiotic correlates that may relate to disease.

Justification and Importance of Proposed Research:
Ornamental olive trees in Southern California urban forests may be incubators for novel pathogenic strains of *X. fastidiosa*. The goal of this study is to survey the genetic diversity of *X. fastidiosa* strains that are currently present in urban olive trees and assess their risk to California's commercial olive industry. Additionally, non-olive plant hosts house an extensive reservoir of *X. fastidiosa* genotypic and phenotypic diversity, which represent an unquantified threat to the emerging olive oil industry in California. Very little known about *X. fastidiosa* infecting plants outside of commercial agriculture settings, especially for trees in Southern California where the pathogen is abundant throughout the landscape (Lee & Almeida, unpublished data). Current *X. fastidiosa* infections in ornamental olives in urban and suburban landscapes may be the source of future spillover onto commercial olive orchards. In 2024 *X. fastidiosa* was frequently detected in olive trees throughout San Diego and Los Angeles counties, suggesting that the pathogen's distribution is broader than previously described. Pervasive infections of olive trees may select for more virulent genotypes of *X. fastidiosa*, posing an even greater risk to California's olive production. This project will be the first to provide a robust assessment of *X. fastidiosa* strains causing disease in ornamental olives. This work will critically evaluate the risk *X. fastidiosa* in urban forests poses to California olive production.

Previous work

Xylella fastidiosa is a vector-borne plant pathogenic bacterium that infects over 720 plant species. There are three well-established subspecies of *X. fastidiosa*, and within each subspecies there are many genotypes of the pathogen associated with different plant diseases (Kahn et al. 2022). *X. fastidiosa* outbreaks are threatening the health of olive trees across the globe. The emergence of olive quick decline syndrome (OQDS) in Italy serves as the most severe example of a *X. fastidiosa* epidemic seriously disrupting agricultural olive production. Olive orchards in the Apulia region have been devastated following the introduction of *X. fastidiosa* subspecies *pauca* and the first report of disease just one decade ago (Sicard et al. 2021). Subsequent detections and accounts of olive disease have also been reported in Spain and France (Amandine et al. 2022, Landa et al. 2018). Although the outbreak in Italy was caused by a single genotype of *X. fastidiosa* subsp. *pauca* and resulted in severe disease emergence, the detection of *X. fastidiosa* subsp. *multiplex* in Spain and France is associated with only mild symptoms (Amandine et al. 2022, Landa et al. 2018). The complex ecology of *X. fastidiosa* in Europe illustrates the need to identify potentially pathogenic strains of *X. fastidiosa* in the landscape before they generate devastating outbreaks.

The emergence of disease in South American olives also demonstrates the necessity of this approach. Phylogenetic analysis revealed genetically distinct *X. fastidiosa* subsp. *pauca* strains causing disease in olive in Argentina and Brazil (Haelterman et al. 2015, Tolocka et al. 2022, Donegan et al. 2023). Analysis in Argentina has revealed two distinct genetic varieties (ST78 and ST69), whereas only one has been described in Brazil so far (ST16; although more are known but have not been published yet) (Haelterman et al. 2015, Tolocka et al. 2022, Donegan et al. 2023). The symptoms caused by the strains isolated in Brazil also closely resemble those reported in Italy (Coletta-Filho et al. 2016), although the strains infecting olive in Italy (ST53) and in Brazil are not closely related. In summary, there are currently numerous genetic groups of *X. fastidiosa* causing disease in olive trees across the globe. Without timely monitoring of *X. fastidiosa* outbreaks and the knowledge of the genetic diversity present in a region, it will be impossible to assess the threat it poses to future olive production.

The effect of *X. fastidiosa* in California olive trees is a generally understudied phenomenon, particularly at the landscape scale. *X. fastidiosa* became abundant throughout Southern California as a result of the introduction of the glassy-winged sharpshooter (*Homalodisca vitripennis*) in the late 1990s (Blua & Morgan 2003). The glassy-winged sharpshooter is the main vector for Pierce's disease of grapevines in Southern California, oleander leaf scorch, and disease in a variety of ornamental hosts (Almeida & Purcell. 2003, Purcell et al. 1999, Wong et al. 2006). The meadow spittlebug (*Philaenus spumarius*) - a naturalized insect vector - has also been identified as an important vector in California (Beal et al. 2021). The broad proliferation of *X. fastidiosa* throughout Southern California now poses an unknown risk to California olive producers.

Olive trees remain as an understudied host species despite frequent reports of disease in the urban forests of major cities; ornamental olive trees used in urban and suburban landscaping have been known hosts of *X. fastidiosa* for years. Most of the isolates from olive in Southern California so far belong to *X. fastidiosa* subsp. *multiplex* (O'Leary et al. 2020, Krugner et al. 2014, Nunney et al. 2019), a different subspecies than what is responsible for the outbreak in Apulia, but the same as those in Spain and France. A second subspecies (subsp. *sandyi*) was also recovered from an olive tree in Irvine (O'Leary & Burbank 2022). In fact, the varieties of subsp. *multiplex* in Spain and France are closely related to those present in California, where they originated from (Landa et al. 2020, Kahn et al. 2022). Greenhouse inoculation studies have demonstrated that the established strains from Southern California only exhibit mild pathogenicity over a one-year study period (Krugner et al. 2014). However, our research group observed severe leaf scorch, stunting, and branch disease in *X. fastidiosa*-positive olives during both our 2023 pilot study and subsequent follow-up work in 2024. It is also clear that only a fraction of the *X. fastidiosa* strains in Southern California have been adequately described.

In addition to unknown levels of genetic diversity, it is unclear what role that climate conditions and landscape features play in the proliferation of *X. fastidiosa* in Southern California olive trees. *X. fastidiosa* appears to respond to a number of environmental variables. Increasing the temperature of insect vector enclosures in greenhouse conditions led to a higher proportion of diseased grapevines in one study (Daugherty et al. 2017). Our lab group has also demonstrated that some strains of *X. fastidiosa* are adapted to local climate conditions - specifically their tolerance to low winter temperatures (Almeida, unpublished data). The effect of environmental variables on *X. fastidiosa*'s disease progression in California olive trees has gone completely untested, and could be vital for predicting high-risk areas for olive production.

In Europe, remote sensing technologies (GIS, Google Earth Engine, etc.) are being used to quantify the incidence of *X. fastidiosa* infections on the landscape (Hornero et al. 2018, Maggiore et al. 2019). This approach could be expanded to elucidate environmental variables that are correlated with specific *X. fastidiosa* genotypes. At a minimum, remote sensing technologies can be used to annotate the landscape into relevant cover types (e.g. riparian, forest, desert). This can be overlaid with *X. fastidiosa* diversity data to identify correlations between specific ecosystem types and high strain diversity, which could help identify high-risk areas for olive growers in the future. Quantifying broad ecosystem variables using remote sensing technologies will help identify landscape features that increase *X. fastidiosa* strain diversity.

This study will be the first systematic evaluation of the genetic diversity of olive-associated *X. fastidiosa* strains in California. This work can be used to generate more accurate risk assessments for the California olive industry, as well as to guide future research on this problem.

Work completed 2024:

Although the genetic typing of these *X. fastidiosa* strains is still in progress, our data suggest that urban forests may be an even greater reservoir of olive-associated *X. fastidiosa* strains than previously thought. While diagnostic testing of this field season's samples has not concluded, over one-third of all olive trees screened for *X. fastidiosa* thus far have tested positive. This is approximately 2-3 times the incidence reported in similar studies with *X. fastidiosa* conducted in urban forests (Harris et al. 2014) and semi-rural landscapes (Olmo et al. 2021). Symptomatic olive trees were observed throughout wide ranging climates in San Diego County and were also abundant in a preliminary collection effort in Los Angeles in 2024 (Figure 1). The prolific incidence and widespread geographic range of *X. fastidiosa*-olive disease in Southern California reinforces the urgency of our proposed study.

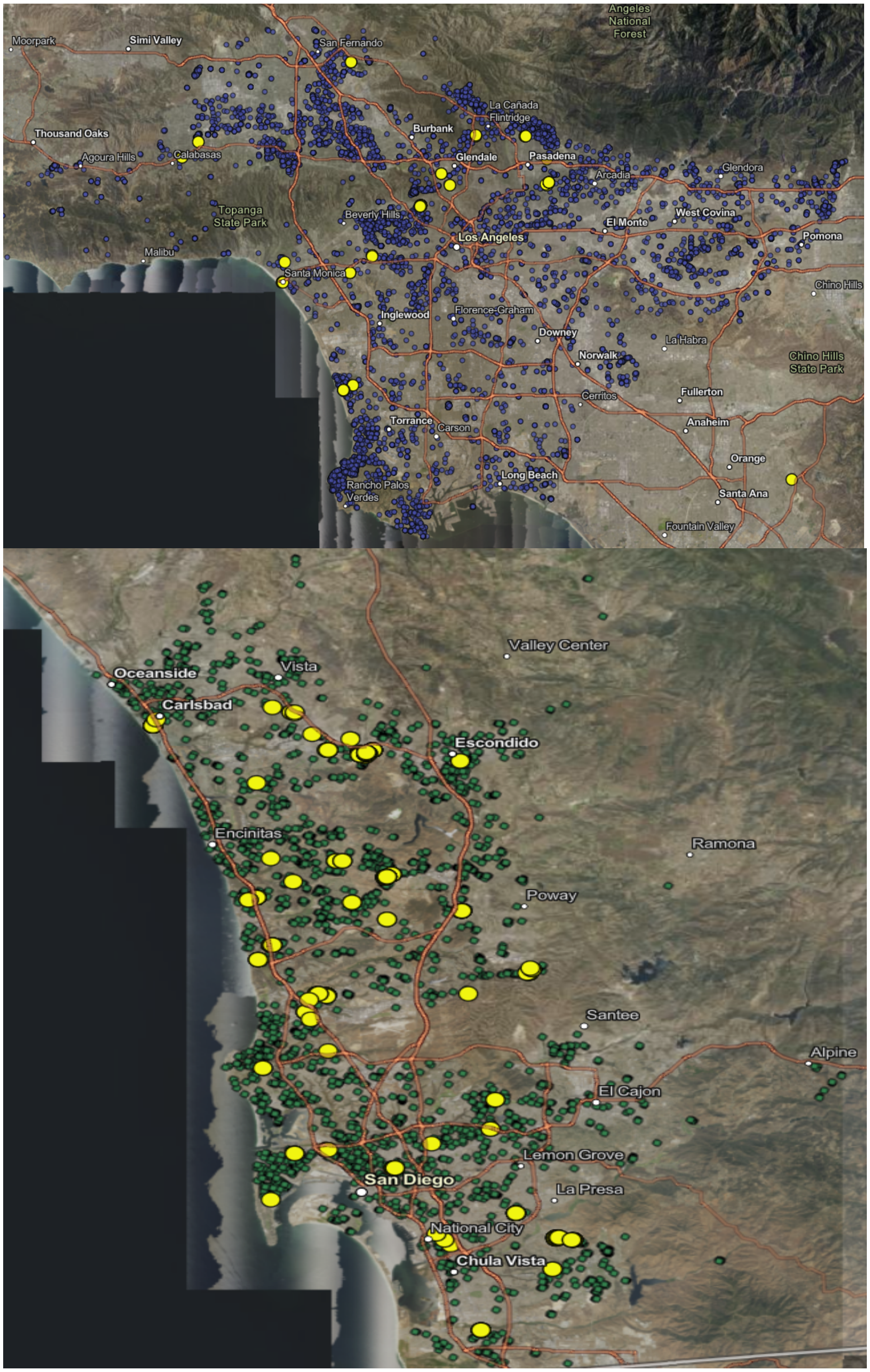

Figure 1: Distribution of samples collected in 2024. Yellow dots represent individual olive tree sampled. Blue dots represent the locations of olive trees in Los Angeles County (top) and green dots represent the locations of olive trees in San Diego County (bottom).

Procedures to Accomplish Objective(s)

Aim 1: Collect field samples from trees throughout Southern California

Symptomatic olive trees will be sampled following established protocols widely used in the surveillance of *X. fastidiosa* outbreaks in Europe (EPPO 2018). Additionally, precise location of each tree will be recorded through GIS platform Survey123 (ESRI). Site selection will be determined by expert knowledge of commercial arborists in Southern California, and UC Cooperative Extension specialists working locally. Sites will have a strong emphasis on areas with dense olive plantings and/or are adjacent to commercial olive orchards. Our lab group recently acquired the precise GPS location of all urban olive trees in Southern California (Love et al. 2022), which in turn has made sampling efforts more precise and efficient. We expect to collect over 300 samples in 2025.

Samples will be shipped to our lab facilities at UC Berkeley for their immediate processing. Non-olive tree species growing in proximity to diseased olives may be sampled to provide a more comprehensive understanding of the genetic diversity of *X. fastidiosa* throughout the landscape. Novel strains that could potentially infect olive trees may be detected by this approach.

Aim 2: Genetic typing of *X. fastidiosa* strains

Samples will undergo DNA extraction and purification from plant petioles using DNeasy plant mini kits (Qiagen) upon their arrival at UC Berkeley. A universal primer set as described in Harper et al. (2010) will be used to detect positive *X. fastidiosa* isolates via real-time quantitative PCR. Live *X. fastidiosa* colonies will also be cultured on PWG media following established protocols (Hill and Purcell 1995). We propose to use two methods to genotype *X. fastidiosa* from olive trees. If we are able to culture cells in the laboratory, we will sequence the complete genome of those strains. Our group has sequenced and analyzed the genomes of several hundred *X. fastidiosa* strains (e.g. Vanhove et al. 2020, Kahn et al. 2022, Donegan et al. 2023), and we do not expect issues, other than the fact that we do not know how many strains we will obtain in pure cultures. The alternative method is to use multi-locus sequence typing by obtaining *X. fastidiosa* genotype information from total DNA extractions, which is widely used for the identification and typing of this pathogen in quarantine situations (e.g. Loconsole et al. 2016). MLST uses the sequence information from seven house-keeping genes, and it will distinguish genotypes of *X. fastidiosa* successfully. We also have experience generating and analyzing MLST data (e.g. Almeida et al. 2008, Kahn et al. 2022). Whole-genome sequences are more informative, but are limited by the number of live *X. fastidiosa* isolates we are able to obtain. Either approach will allow us to untangle the evolutionary relationships between *X. fastidiosa* strains in Southern California with a high degree of specificity, and neither whole-genome sequencing or MLST has been systematically conducted for *X. fastidiosa* in olives in Southern California before this study.

Aim 3: Remote sensing analysis

Various remote sensing and spatial ecology methods will be used to map the distribution of positive *X. fastidiosa* cases and determine associations between environmental variables and pathogen genotype. Widely available satellite imagery datasets such as Landsat 8 platform use infrared sensors to directly measure surface temperature and other wavelengths of light that can be used to calculate key bioclimatic variables without the need to deploy individual weather stations. A series of raster images will be generated from this dataset using Google Earth Engine, a powerful open-source remote sensing analysis platform. These raster images will map the variation in key bioclimatic (annual mean temperature, precipitation, etc.) and spectral (greenness, amount of developed space) variables across Southern California. Remote sensing technologies have been used to explore the links between climate and *X. fastidiosa* infections even at fine spatial scales (Hornero et al. 2018).

The raster values at GPS coordinates for symptomatic trees will be extracted to determine the specific microclimatic conditions at each site. A robust series of R packages (Terra, Raster, etc.) are available to manipulate raster data and calculate these values. Statistical analysis will then be performed to examine if bioclimatic variables are highly correlated with pathogen incidence and specific *X. fastidiosa* genotypes. The climate conditions of areas where olive trees are currently affected by *X. fastidiosa* can then be compared to regions of California where olive production is projected to increase to generate risk assessments. GIS and other geospatial analysis software can also be used to divide regional maps up into relevant subclasses (farmland, riparian, developed, etc.). *X. fastidiosa* diversity data could be then overlaid with these maps to demonstrate associations between specific ecosystem types and higher *X. fastidiosa* strain diversity. Such an approach is already being used to infer the range and population dynamics of *X. fastidiosa* insect vectors (Santoiemma et al. 2019) Implementing this analysis in the urban context - where the landscape use often is highly variable and spatially fragmented – will help identify ecosystems that retain higher numbers of distinct *X. fastidiosa* strains. Once this is complete, the urban forestry inventory data aggregated through the method of Love et al. (2022) could be used to identify areas where high *X. fastidiosa* diversity may be predicted based on both ecosystem type and olive abundance. In summary, remote sensing tools would help elucidate potential *X. fastidiosa* hotspots where olive production should be avoided.

Summary

The proposed project will be the first to systematically quantify the genetic diversity of olive-associated *X. fastidiosa* strains in Southern California. A greater number of olive isolates will be described with a higher degree of genetic fidelity than previous studies. The novel application of remote sensing technology will also help establish existing links between climate and genetic diversity/incidence of *X. fastidiosa* in olive trees. Taken together, these approaches will help generate biologically-informed risk assessments for California olive production.

Timetable for Project

Sample collection for Objective 1 will occur in the Summer and Fall of 2025, while sample processing and analyses will take place in subsequent Winter and Spring months. This is the same schedule we used in 2024; we have recently finished our sample collection trips and subsequently tested most samples for *X. fastidiosa*. The genotyping component of the work initiated recently. This schedule will provide us with two years to collect samples (2024 already done, and 2025), and the focus in 2025 will be under sampled areas or areas where more genetic diversity was detected in 2024. The landscape analysis components of the work require the availability of some of the data, so that computational pipelines can be developed. This work will likely start in Spring 2025, although we expect to focus on Objective 2 towards the end of the project when most/all the data have already been collected.

	Fall 2025	**Spring 2026**
Obj. 1. Genetic survey	**X**	**X**
Obj. 2. Landscape analyses	**X**	**X**
Data analyses, reports	**X**	**X**

Present Outlook and Estimated Success in Accomplishing Objective(s)

Our research group has three decades of experience working on the biology, ecology, and management of *X. fastidiosa* diseases in California and globally, including olive disease in Europe and Brazil. We have done extensive research on the diversity of *X. fastidiosa* using the tools and approaches proposed here, including performing MLST and whole-genome sequence analyses. We previously acknowledged possible difficulties in obtaining cultures from olive trees in California as this has been challenging to colleagues that have pursued similar research; we have found this difficult as well. We are reaching out to colleagues in the US and Europe that have had success obtaining isolates from olive trees, hoping to adapt and improve our protocols.

In 2023 we started working with arborists in San Diego Co. and we were able to sample from that region. Our initial estimates were to collect between 100-200 samples each year. In 2024, as part of our ongoing COC-funded project, we sampled 246 olive trees in southern California, focusing on San Diego County and Los Angeles County. Our lab group got access to the locations of urban olive trees (see Love et al. 2022) midway through our sampling season. This greatly expedited the pace of sampling; thus, we anticipate that we will be able to gather an equal (if not greater) number of samples next year. Approximately 1/3 of all sampled plants were PCR-positive for *X. fastidiosa*, and our group is now genotyping those samples. We attempted to culture live *X. fastidiosa* cells from about 1/3 of olive samples collected (n = 77) but were not successful. *X. fastidiosa* is notoriously challenging to isolate from olive trees. We are leveraging our international network of *X. fastidiosa* specialists to amend our protocols and improve our future success in isolating *X. fastidiosa* cells from olive. Our group is confident that we will be able to expand the scope and success of this research in 2025.

Rodrigo Almeida's brief CV can be found here:
https://nature.berkeley.edu/almeidalab/wp-content/uploads/2023/03/Almeida_2_page_CV.pdf
The publication record of Rodrigo Almeida's group is listed here:
https://nature.berkeley.edu/almeidalab/publications/

Budget Support Summary by Objective(s):

This project proposal, and related work, has not been submitted to any other funding source.

Personnel (~$7,000/year)
The research will be performed by a graduate student that is funded by a National Science Foundation (NSF) fellowship. These are prestigious fellowships that cover stipend, tuition, and fees for the student during the academic year. We request funding to support this student for one month (100% appointment) during each Summer of this project. The student will work on the project throughout the year, not only the one month in the Summer. Student salary and benefits follow required University of California rates.

Laboratory supplies and sequencing costs ($15,000/year)
We hope to generate data for 150-200 samples per year: more if technically feasible. The cost of DNA extractions and other materials and reagents is estimated to be $5/sample. Costs for culturing *X. fastidiosa* are estimated to be $2,000/year. The cost of genotyping one sample using MLST is $62 (7 genes, both strands - 14 reads); the cost of genotyping one sample using WGS is ~$140 (Illumina, 100-200x depth of coverage). Ideally we would only use WGS for each sample, but shipped samples may no longer have live cells when we receive them, or culturing we may be very challenging (based on the experience of others). Our estimate of $15,000/year for reagents/sequencing assumes a balance of 150-200 samples genotyped using MLST and/or WGS, in addition to other lab costs. Only after one season will we be able to provide a more accurate estimate of the breakdown of samples typed using MLST and/or WGS.

Travel costs ($4,500/year)
In addition to processing samples mailed to Berkeley, we will also perform three trips annually to collect samples. These trips will allow us to close geographical and landscape gaps in sampling. We estimate travel costs for one week as: flights: $200 per trip (x3) = $600; motel: ~$800 per week (x3) = $2,400; rental car: $500 per week (x3) = $1,500.

GAEL costs
The General, Automobile, and Employment Liability (GAEL) charge was instituted in 1998 to fund the campus's share of expenses associated with claims and lawsuits defended by the University; the GAEL charge is $1.75 per $100 of payroll. This applies to all funds, including gifts and grants, with the exception of direct federal contracts, grants, and flow-throughs. (https://riskservices.berkeley.edu/insurance-programs/liability)

Indirect costs
The program allows for indirect costs not exceeding 11% of the budget, as described in the RFP.

Total Budget Request:

	2025-2026
Personnel	$7,373
Lab reagents/sequencing costs	$15,000
Travel expenses	$4,500
GAEL costs	$148
Total direct cost	$27,021
Indirect cost (11%)	$3,340
Total cost	**$30,361**

Literature Cited:

Almeida, R. P. P., & Purcell, A. H. (2003). Transmission of Xylella fastidiosa to Grapevines by Homalodisca coagulata (Hemiptera: Cicadellidae). *Journal of Economic Entomology*, *96*(2), 264–271. https://doi.org/10.1093/jee/96.2.264

Almeida, R. P. P., Nascimento, F. E., Chau, J., Prado, S. S., Tsai, C.-W., Lopes, S. A., & Lopes, J. R. S. (2008). Genetic Structure and Biology of Xylella fastidiosa Strains Causing Disease in Citrus and Coffee in Brazil. *Applied and Environmental Microbiology*, *74*(12), 3690–3701. https://doi.org/10.1128/AEM.02388-07

Beal, D. J., Cooper, M., Daugherty, M. P., Purcell, A. H., & Almeida, R. P. P. (2021). Seasonal Abundance and Infectivity of Philaenus spumarius (Hemiptera: Aphrophoridae), a Vector of Xylella fastidiosa in California Vineyards. *Environmental Entomology*, *50*(2), 467–476. https://doi.org/10.1093/ee/nvaa178

Blua, M. J., & Morgan, D. J. W. (2003). Dispersion of Homalodisca coagulata (Hemiptera: Cicadellidae), a Vector of Xylella fastidiosa, into Vineyards in Southern California. *Journal of Economic Entomology*, *96*(5), 1369–1374. https://doi.org/10.1093/jee/96.5.1369

Coletta-Filho, H. D., Francisco, C. S., Lopes, J. R. S., De Oliveira, A. F., & Da Silva, L. F. D. O. (2016). First report of olive leaf scorch in Brazil, associated with Xylella fastidiosa subsp. Pauca. *Phytopathologia Mediterranea*, *55*(1), 130–135. https://doi.org/10.14601/Phytopathol_Mediterr-17259

Costa, H. S., Raetz, E., Pinckard, T. R., Gispert, C., Hernandez-Martinez, R., Dumenyo, C. K., & Cooksey, D. A. (2004). Plant Hosts of Xylella fastidiosa In and Near Southern California Vineyards. *Plant Disease*, *88*(11), 1255–1261. https://doi.org/10.1094/PDIS.2004.88.11.1255

Donegan, M. A., Coletta-Filho, H. D., & Almeida, R. P. P. (2023). Parallel host shifts in a bacterial plant pathogen suggest independent genetic solutions. *Molecular Plant Pathology*, *24*(6), 527–535. https://doi.org/10.1111/mpp.13316

EPPO PM 7/24(4) *Xylella fastidiosa*. (2019). *EPPO Bulletin*, *49*(2), 175–227. https://doi.org/10.1111/epp.12575

Haelterman, R. M., Tolocka, P. A., Roca, M. E., Guzmán, F. A., Fernández, F. D., & Otero, M. L. (2015). First Presumptive Diagnosis of Xylella Fastidiosa Causing Olive Scorch in Argentina. *Journal of Plant Pathology*, *97*(2), 393–393.

Kahn, A. K., & Almeida, R. P. P. (2022). Phylogenetics of Historical Host Switches in a Bacterial Plant Pathogen. *Applied and Environmental Microbiology*, *88*(7), e02356-21. https://doi.org/10.1128/aem.02356-21

Harper, S. J., Ward, L. I., & Clover, G. R. G. (2010). Development of LAMP and Real-Time PCR Methods for the Rapid Detection of Xylella fastidiosa for Quarantine and Field

Applications. *Phytopathology®*, *100*(12), 1282–1288. https://doi.org/10.1094/PHYTO-06-10-0168

Hornero, A., Hernández-Clemente, R., Beck, P. S. A., Navas-Cortés, J. A., & Zarco-Tejada, P. J. (2018). Using Sentinel-2 Imagery to Track Changes Produced by Xylella Fastidiosa in Olive Trees. *IGARSS 2018 - 2018 IEEE International Geoscience and Remote Sensing Symposium*, 9060–9062. https://doi.org/10.1109/IGARSS.2018.8517697

Hill, B.L. & Purcell, A.H. (1995). Acquisition and retention of *Xylella fastidiosa* by an efficient vector, *Graphocephala atropunctata*. *Phytopathology*. *85*(2), 209-212.

Krugner, R., Sisterson, M. S., Chen, J., Stenger, D. C., & Johnson, M. W. (2014). Evaluation of Olive as a Host of Xylella fastidiosa and Associated Sharpshooter Vectors. *Plant Disease*, *98*(9), 1186–1193. https://doi.org/10.1094/PDIS-01-14-0014-RE

Landa, B. B., Velasco-Amo, M. P., Marco-Noales, E., Olmo, D., López, M. M., Navarro, I., Monterde, A., Barbé, S., Montes-Borrego, M., Román-Écija, M., Saponari, M., & Giampetruzzi, A. (2018). Draft Genome Sequence of Xylella fastidiosa subsp. Fastidiosa Strain IVIA5235, Isolated from Prunus avium in Mallorca Island, Spain. *Microbiology Resource Announcements*, *7*(14), 10.1128/mra.01222-18. https://doi.org/10.1128/mra.01222-18

Loconsole, G., Saponari, M., Boscia, D., D'Attoma, G., Morelli, M., Martelli, G. P., & Almeida, R. P. P. (2016). Intercepted isolates of Xylella fastidiosa in Europe reveal novel genetic diversity. *European Journal of Plant Pathology*, *146*(1), 85–94. https://doi.org/10.1007/s10658-016-0894-x
Maggiore, G., Semeraro, T., Aretano, R., De Bellis, L. & Luvisi, A.(2019). GIS Analysis of Land-Use Change in Threatened Landscapes by Xylella fastidiosa. *Sustainability*, *11*(1), 253. https://doi.org/10.3390/su11010253

Nunney, L., Azad, H., & Stouthamer, R. (2019). An Experimental Test of the Host-Plant Range of Nonrecombinant Strains of North American Xylella fastidiosa subsp. Multiplex. *Phytopathology®*, *109*(2), 294–300. https://doi.org/10.1094/PHYTO-07-18-0252-FI

O'Leary, M. L., Burbank, L. P., Krugner, R., & Stenger, D. C. (2020). Complete Genome Sequence Data of Three Xylella fastidiosa subsp. Multiplex Strains Isolated from Olive Trees in California, U.S.A. *Phytopathology®*, *110*(11), 1759–1762. https://doi.org/10.1094/PHYTO-05-20-0167-A

Purcell, A. H., Saunders, S. R., Hendson, M., Grebus, M. E., & Henry, M. J. (1999). Causal Role of Xylella fastidiosa in Oleander Leaf Scorch Disease. *Phytopathology*, *89*(1), 53–58. https://doi.org/10.1094/PHYTO.1999.89.1.53

Sicard, A., Saponari, M., Vanhove, M., Castillo, A. I., Giampetruzzi, A., Loconsole, G., Saldarelli, P., Boscia, D., Neema, C., & Almeida, R. P. P. (2021). Introduction and adaptation of an emerging pathogen to olive trees in Italy. *Microbial Genomics*, *7*(12), 000735. https://doi.org/10.1099/mgen.0.000735

Tolocka, P.A. , Guzmán, F.A. , Paccioretti, M.D. , Roca, M.E. , Otero, M.L. & Haelterman, R.M. (2021) Identificación de la secuencia tipo ST78 de *Xylella fastidosa* en olivo de Argentina [poster]. *5 th Congreso Argentino de Fitopatologia*. Available at: https://repositorio.inta.gob.ar/xmlui/handle/20.500.12123/10519 [Accessed 28th February 2023].

Vanhove, M., Sicard, A., Ezennia, J., Leviten, N., & Almeida, R. P. P. (2020). Population structure and adaptation of a bacterial pathogen in California grapevines. *Environmental Microbiology*, *22*(7), 2625–2638. https://doi.org/10.1111/1462-2920.14965

Harris, Jordan L., Patrick L. Di Bello, Monica Lear, and Yilmaz Balci. "Bacterial Leaf Scorch in the District of Columbia: Distribution, Host Range, and Presence of *Xylella Fastidiosa* Among Urban Trees." *Plant Disease* 98, no. 12 (December 2014): 1611–18. https://doi.org/10.1094/PDIS-02-14-0158-SR.

Olmo, Diego, this link will open in a new tab Link to external site, Alicia Nieto, David Borràs, Marina Montesinos, Francesc Adrover, Aura Pascual, et al. "Landscape Epidemiology of Xylella Fastidiosa in the Balearic Islands." *Agronomy* 11, no. 3 (2021): 473. https://doi.org/10.3390/agronomy11030473.

Love, Natalie L.R., Viet Nguyen, Camille Pawlak, Andrew Pineda, Jeff L. Reimer, Jennifer M. Yost, G. Andrew Fricker, et al. "Diversity and Structure in California's Urban Forest: What over Six Million Data Points Tell Us about One of the World's Largest Urban Forests." *Urban Forestry & Urban Greening* 74 (August 2022): 127679. https://doi.org/10.1016/j.ufug.2022.127679.

Santoiemma, Giacomo, Giovanni Tamburini, Francesco Sanna, Nicola Mori, and Lorenzo Marini. "Landscape Composition Predicts the Distribution of Philaenus Spumarius, Vector of Xylella Fastidiosa, in Olive Groves." *Journal of Pest Science* 92, no. 3 (June 2019): 1101–9. https://doi.org/10.1007/s10340-019-01095-8.

Workgroup/Department: Olive / Plant Sciences, UC Davis

Project Year: April 1, 2025 – March 31, 2026. Anticipated Duration of Project: 1 year
(UC Davis Sponsored Programs Proposal # xxxxx- submitted)

Project Title:
Evaluation of effects of Accede® (ACC) at two different application rates on enhancing horticultural maturity and abscission zone development and commercial trunk shaking efficiency in table olives

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Objective(s) of Proposed Research:

1. Confirm 1500ppm ACC application efficacy on trunk shaker harvest efficiency as evidence for IR-4 application
2. Compare efficacy of 750 ppm ACC application to 1500 ppm ACC application on trunk harvesting efficiency
3. Perform ACC residue analysis on fruits sprayed by 750 ppm and 1500 ppm ACC application to estimate the lower limit of field application concentration and supplement the 2025 IR-4 application for residue testing in the 2026 season
4. Provide an economic analysis comparing the two concentrations of ACC application, considering all economic impacts in the trial including potential loss of yield if only mechanical harvesting, cost of Accede®, and cost of gleaning if a hand-harvest component is added

Justification and Importance of Proposed Research:

This proposal responds to the COC call of “Mechanical harvesting on existing and new high-density orchards” and “Development of Loosening Agents”.

Table olive production in California has historically relied on hand labor for harvest operations. The ever-rising minimum wage in California coupled with the scarcity of skilled labor, make this a top priority for optimization in the industry. A 2023 cost study of table olive production lists hand-harvest as the largest annual operating cost for growers at $3,250/acre, eclipsing the second largest annual expense of pruning at $868 per acre (Murdock and Goodrich 2023). Mechanical harvesting could potentially save growers thousands of dollars per acre, making table olives a more attractive and sustainable crop in California.

Through the past year’s work, we have established a recommendation of mechanical harvesting protocol for high efficiency

1. Skirting pruning during May - June
2. During pre-harvest (early September or starting at GDD 1000°C), Monitor fruit removal force, until it drops below 0.3 kg.
3. Appy 1500 ppm ACC afterwards, and harvest after 7-10 days post application.

This year’s results showed that skirting low branches in spring does not affect the total yields of the trees. This year’s results confirmed that ACC application after fruit removal force dropping below 0.3 kg can increase the mechanical harvesting efficiency by ~15%, similar to last year’s results.

Cellular analyses confirmed that ACC promotes the formation of fruit abscission zone (FAZ). In FAZ cell layer of ACC treated fruits, alkalization of cytosol and decrease of plasmodesmata callose were observed as in ripe fruits. This underlines natural cellular mechanisms of FAZ formation that can be accelerated with ACC application.

We have initiated the IR-4 application process. Next year we will obtain the third year’s data to fulfill the IR-4 application requirements.

Next year, we would like to test a lower concentration of ACC (Accede™) application, and we will perform residue test to estimate the minimum effective concentration of field application.

IR-4 progress of Accede™

Importantly, in 2024 the IR-4 program agreed to provide funding, in conjunction with the California Department of Food and Agriculture, to support continued research into the efficacy and crop safety of Accede™ when used as a loosening agent in table olive. Valent expressed support of continuing research toward the goal of registration. Representatives from Valent also expressed the importance in evaluating the efficacy of Accede™ at a lower rate to reduce the cost and improve accessibility for growers. The IR-4 program will contribute $10,000 for the 2025 season’s efficacy and crop safety research. If 2025 data continue to show good efficacy of Accede™, as expected based on our two years’ data tends, residue testing will be deemed necessary. Receiving support from the IR-4 program, in addition to having support from the registrant, Valent, and the table olive industry have clearly defined the path to registration for this product. Hence results of this project on evaluating efficacy of Accede™ for a third year and testing chemical residuals on the fruit in 2026 by IR-4 laboratories will be essential for the product registration.

Results of 2024 season

Fruit removal force reaches ~0.3 kg in late September

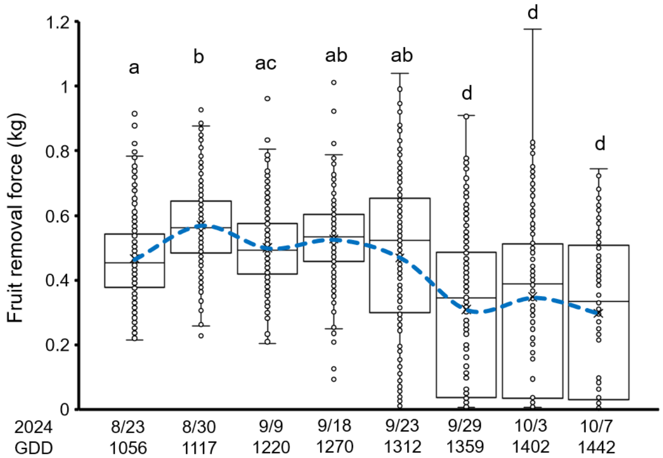


Figure 1. Fruit removal force during pre-harvest stages of olive fruit development. Each biological replicate is one fruit. Blue dashed line indicates the change of average during the sampling period. Different letter indicates significant difference between groups (n = 120 or 240, one-way ANOVA, Scheffe's Test, $p < 0.05$). Growing degree day (GGD), is shown on the x axis and is calculated using 15°C as base value and May 10th, 2024 as half bloom day, and mechanical harvest was performed on 10/7/24.

We continued to monitor fruit removal force in relation to growing degree days (GDD) and calendar dates. A similar trend was observed throughout the past three years and a major drop of fruit removal force was also observed near the end of September this year, at around 1350 GDD (**Figure 1**). We followed the criteria of 0.3 kg fruit removal force and sprayed with ACC at 1500 pm, with a 100 gallons per acre (GPA) rate on September 30th, 2024. The mechanical harvest was performed on October 7th, 2024, 7 days after the spraying the orchard.

ACC application significantly reduces FRF

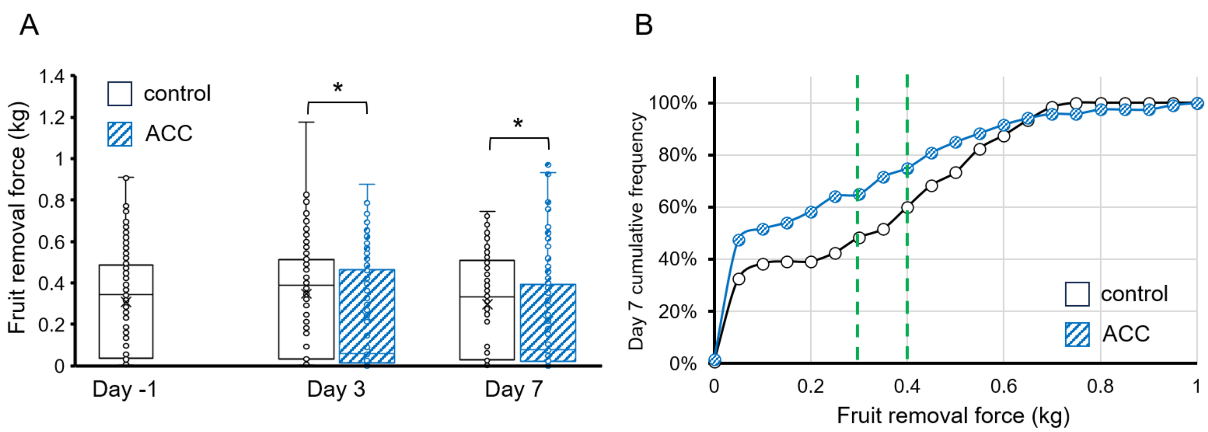


Figure 2. (A) Fruit removal force before and after ACC treatment. Each biological replicate is one fruit. Asterisk indicates significant difference between control and ACC treated group (n = 120, t-test, $p<0.05$). **(B) Cumulative frequency of fruit removal force of fruits sampled on the harvest day.** Green dashed lines suggest fruit removal force range corresponding to mechanical harvest removal rates.

ACC treatment (1500 ppm, 100 GPA) was performed on 9/30/24 (GDD 1369°C), and no significant FRF changes were observed during the 8 days in the control group (**Figure** 2) in the course of the ACC application period (9/30-10/7). However, in the ACC treated group, a significant lower FRF was observed on both the 3rd-day and 7th -day after treatment.

Cumulative frequency of FRF on 7th-day after treatment (**Figure 2B**) suggest that the fruits with lower than 0.4 kg FRF, represent 75% and 60% of the population in ACC treated and control group, which approximately matches the mechanical harvest efficiency of ACC treated and control group. Our data of 2024 (**Figure 2B**) suggested that this year the threshold for using the trunk shaker to remove a fruit from the tree is between 0.3 - 0.4 kg.

Spring skirting pruning does not affect yield

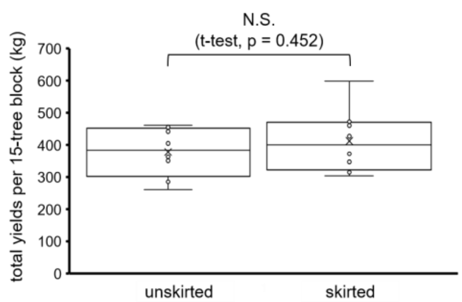
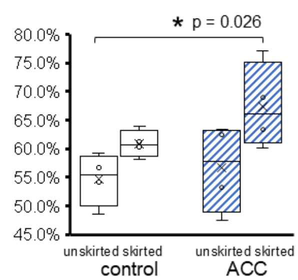


Figure 3. Total yields of two pruning treatment (unskirted vs skirted). Each biological replicate is a block of 15 trees (n = 8).

We found that low branches dangling on the receiver of trunk shaker cannot be efficiently harvested by the trunk shaker. Sometimes lower branches even directly block the receiver and get knocked off by the receiver. We performed a skirting pruning test (4 feet above the ground) this spring. The total yield of 15-trees, including the weight of mechanical harvest and hand gleaned fruits, are not significantly different between the unskirted and skirted groups (**Figure 3**). These results suggest that skirting improves mechanical harvesting efficiency without the penalty of yield. Skirting pruning should be the practice to follow, preparing the orchard in the spring for the upcoming fall mechanical harvesting.

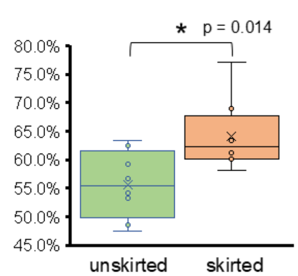
Both skirting pruning and ACC application increase mechanical harvest efficiency

A



	control	ACC	efficiency increase by ACC
unskirted	54.7%±2.3%	56.6%±3.8%	*3.5%*
skirted	60.9%±1.2%	67.4%±3.7%	*10.7%*
efficiency increase by skirting	*11.3%*	*19.1%*	*Combinational increase: 23.4%**

B



	combined
unskirted	55.6%±2.1%
skirted	64.2%±2.2%*
efficiency increase by skirting	*15.5%*

Figure 4. Efficiency comparison (**A**) of pruning and ACC treatments. Asterisk indicates significant higher efficiency (23.4%) observed in the group treated by both ACC spray and skirt pruning, compared to the no treatment control (n = 4, t-test, p = 0.026). (**B**) by combining spray treatment groups. Asterisk indicates significant higher efficiency is observed in skirted group (n = 8, t-test, p = 0.014). Each biological replicate is a block of 15-tree.

We tested effect of skirting and ACC spray on mechanical harvest this year. Both induced an increase in harvesting efficiency. Combinational treatment of ACC spray and skirting induced a **23.4% increase of harvesting efficiency** (**Figure 4A**), while improved efficiency was also observed by skirting alone (15%) (**Figure 4B**).

This year's result confirmed the efficacy of ACC implemented using current established orchard management practices. Further our 2024 results provide 2nd year supporting evidence for IR-4 application of ACC in the table olive mechanical harvesting.

Live staining confirms the efficacy of ACC to promote FAZ formation at the cellular level

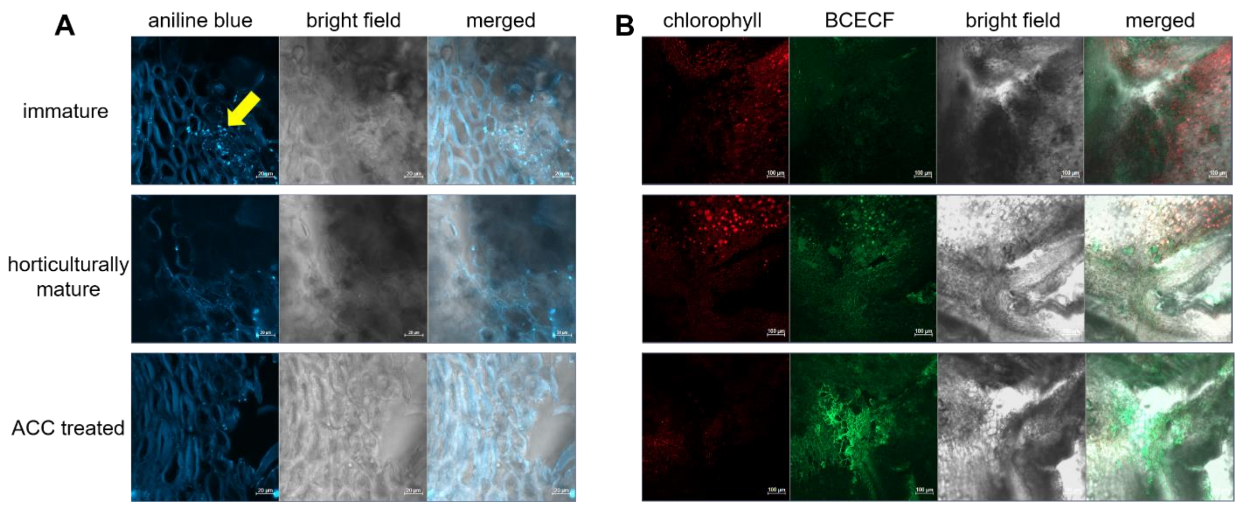


Figure 5. Live staining of callose (A) and cytosolic pH (B) at olive FAZ. (**A**) Fluorochrome aniline blue staining of callose at plasmodesmata. Yellow arrow points to blue fluorescent spots that indicate the presence plasmodesmatal callose at FAZ. Scale bar = 20 µm. (**B**) BCECF cytosolic staining of FAZ. Higher green signal indicates higher pH. Scale bar = 100 µm.

Less plasmodesmata callose staining signals were observed in horticulturally mature samples compared to immature samples at FAZ, suggesting that symplasmic trafficking is promoted during the maturation process. ACC treated samples also showed a higher rate of FAZs with low plasmodesmata callose (**Figure 5A**), suggesting ACC application advances the maturation process. We observed the same effect in ethephon treated samples in previous years' study.

Alkalization of abscission zone cytosolic pH was previously suggested as an indicator of fruit abscission zone formation. We now observed this phenomenon in a tree crop, olive. More cells with alkalized cytosol were observed in horticulturally mature and ACC treated samples (**Figure 5B**).

In summary, live staining of plasmodesmata callose and cytosol pH confirms the efficacy of ACC in promoting the formation of fruit abscission zone at cellular level.

Procedures to Accomplish our Objective(s):

Below are step by step our analyses for an in depth evaluation of ACC, Accede™ in improving harvesting efficiency and its mode of action in table olives.

1. **Orchard preparation**

The field application of the experiment will be conducted at the Leslie J. Nickels Trust Soils Laboratory, moderate- density 'Manzanillo' orchard in Arbuckle, Colusa County; 202 trees/acre in 12, 30-tree N-S rows, planted in 2001. The orchard will be mechanically skirted at 4' and topped at 12' in all rows and hedged on alternate rows 4' feet from the trunk to maintain a harvestable size. Trees will be hand pruned within the canopy to increase light interception in spring 2025. The need to chemically thin will be determined in spring 2025.

2. **Test the effect of ACC application on the trunk shaker harvest efficiency**

We will monitor the fruit removal force as we did in previous years. We will continue monitoring the growing degree day in the 2025 growing season, in order to build the growing degree day (GDD) model for the table olive mechanical harvesting and general olive industry. We will also compare other plant physiology based studies of olives to obtain a more accurate ceiling temperature for the olive GDD formula modification. Delaying the application of ACC until FRF reaching below 0.3 kg had a positive effect in the overall chemical application and reduction of fruit removal force. Additionally, we have been informed that Accede™ begins losing efficacy at 38°C. FRF data and predicted weather will determine when the Accede™ application will be made during the application period.

The original determination of concentration of Accede™ to be tested was determined by back calculating from effective concentrations of ethylene using Ethephon™ products. Efficacy studies in 2023 showed no reduction of harvest efficiency when 1500ppm Accede™ was applied at 100 gallons per acre (gpa) vs 400gpa. With no loss of efficacy at 1500ppm applied at 100gpa combined with the high cost of material at these rates, it is important to evaluate the product at lower concentrations. Not only does a lower rate provides potentially fewer registration hurdles due environmental sustainability, it also has the potential to be more financially accessible to small growers once registered.

Harvest timing will be 10 days after Accede® application depending upon the drop in FRF as described above and harvester availability.

At harvest, the mechanically harvested yield of the central 6 trees of the 10-tree plots will be weighed, and a subsample will be submitted for grading and determination of price per ton for each plot.

- 10 replications x (2 treatments + 1 controls) = 30 grading samples submitted to Musco Orland Receiving Station for quality grading.

These same 6 central trees will be hand gleaned after harvest and fruit weighed and a subsample graded. This will allow for evaluation of potential loss of value of mechanically harvested fruit due to damage and trash. Hand gleaned plots will generate an additional 30 samples for grading, resulting in a total of 60 samples to be graded for the entire trial.

Harvester efficiency of each treatment will be calculated as:

$$Efficiency = \frac{Mechanically\ harvested\ (lb)}{Manually\ harvested\ (lb) + \ Mechanically\ harvested\ (lb)} \times 100$$

All the additional trees in the plot will be mechanically harvested, and if possible, hand gleaned, to obtain a more representative effect of Accede® application on yield.

<table>
<tr><th colspan="14">Proposed 2025 Table Olive-Accede Efficacy Trial Plot Map</th></tr>
<tr><th>Tree#</th><th>Row 1</th><th>Row 2</th><th>Row 3</th><th>Row 4</th><th>Row 5</th><th>Row 6</th><th>Row 7</th><th>Row 8</th><th>Row 9</th><th>Row 10</th><th>Row 11</th><th>Row 12</th><th>Row 13</th></tr>
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Figure 6. Field plot map plan for 2025 table olive-ACC efficacy trial. Our group will be involved as indicated at the timetable section. Ten-tree plots will be sprayed with either 750 ppm ACC, 1500 ppm ACC, or the control spray + 0.025% non-ionic surfactant. Plots are arranged in a randomized complete block design, with each row being a block. Pull tests will be performed by 12 rows x 4 trees per row x 5 olives per tree = 240 olives per pull test.

3. Chemical analysis of ACC treated fruits

In our 2023 trial, we found that application of ACC at 1500 ppm at a rate of 100 GPA and 400 GPA did not show a difference in promoting the reduction of FRF and in enhancing mechanical harvesting efficiency, and both were significantly higher than the control. These results suggest that the plateau of ACC efficacy was reached by the ACC spraying application at 1500 ppm, 100 GPA.

In practice, the minimum effective concentration of a product is not always tested in field application setting. The past two years' results have confirmed the consistency of Accede's efficacy. Based on the consistency of the product in field application and for grower's benefit, we would like to check the efficacy of a lower concentration in field application. Accompanied with the economic analysis (see below), we also have a scientific base to test lower concentration of ACC application. The concentration of ACC at 1500 ppm (14.8 mM) significantly exceeds the highest K_m values (Michaelis constant, the concentration of substrate which permits the enzyme to achieve half V_{max}) of ACC oxidase reported in the literature, which are: 0.401 mM for one isoform of ACC oxidase in the climacteric fruit apple (Binnie & McManus, 2009) and 0.147 mM in the non-climacteric fruit winter squash (Mathooko et al., 1993). Therefore, we would like to test 750 mM (half of current tested concentration) of ACC during the next season.

We will perform residue test of sprayed fruits to estimate the lower limit of field application to exert full efficacy of ACC on olive mechanical harvest. Plants have the ability to adjust the pool of free ACC and therefore regulating ethylene biosynthesis. Besides converted to ethylene, the applied ACC can be stored as conjugated forms in vivo, which are not an active form, to serve as precursor of ethylene biosynthesis (de Poel & Van Der Straeten, 2014). Early works, found that ACC stays more as conjugated form of malonyl-1-aminocyclopropane carboxylic acid (MACC) at pre-climacteric stage, and more as free form as substrate of ACC oxidase at ripening stage in apple (Mansour et al., 1986). The amount of free ACC and conjugated ACC will be quantified by a established LC/MS method (Chauvaux et al., 1997), at UC Davis Food Safety and Measurement Facility. If applying 750 mM ACC at 100 GPA results in over 1 mM of free or conjugated ACC in olive fruit, this suggests that 750 mM is above the minimum effective concentration and should be retained for future use. Conversely, if less than 0.1 mM of free or conjugated ACC is found in the fruits sprayed by 1500 ppm ACC, it indicates that the olive fruit can fully convert the applied 1500 mM ACC at 100 GPA into ethylene. In this case, we should continue with 1500 ppm ACC to achieve full efficacy.

In parallel with the biochemical studies, we will confirm the FAZ development using imaging by lignin, plasmodesmata, and cytosolic pH staining to validate the results from previous years and provide a better characterization of the mode of action of the chemical.

4. **Economic analysis of proposed treatment combinations**

Dr. Domena Agyeman, UCCE Agricultural and Natural Resource Economics Advisor, joins the team this year to assist with the economic analysis of the use of Accede in table olive systems. The 2023 Sample Costs to Produce Table Olives, published by researchers at UCCE and UC Davis, will be used as a basis for all costs not affected by treatments imposed in the trial. Using the hourly labor rates listed in the cost study, current costs of Accede®, current rates for mechanical skirting and harvesting, and the crop value as provided in the grade sheets, the following costs and returns will be calculated for each treatment:

1. Cost of Accede® material and labor associated with application
2. Cost of contracted mechanical skirting
3. Cost of labor associated with manual pruning
4. Cost of contracted mechanical harvest based on time/acre
5. Cost of labor for hand-gleaning after mechanical harvest
6. Gross proceeds for crop harvested each treatment based on yield and grade sheet results

4. Statistical analysis of data

The field experimental design is a randomized complete block with each treatment combination repeated once within each block/replication.

An Analysis of Variance (ANOVA) with an LSD means separation test will be used to compare the 2 treatments and control on:

- yield
- harvesting efficiency
- quality and price per ton
- overall profitability

With the final objective of determining which concentration of Accede® provides adequate highest harvesting efficiency and provides the most economic benefit.

Present Outlook and Estimated Success in Accomplishing Objective(s):

The promising results of the 2023 and 2024 ACC field experiments as well as the progress made toward registration with IR-4 and registrant support encourage us to repeat the ACC treatment in 2025 with proposed modifications as discussed above. We want to confirm the efficacy of ACC in another year's experiment, determine the residual compound on the fruit, and evaluate the abscission zone responses as well as determine if lower concentrations of ACC is effective at improving harvesting efficiency. The mode of action and effect of ACC on fruit abscission zone induction will be determined and compared with studies using ethephon at the cellular level providing concluding evidence on their effect in enhancing abscission zone formation and thereby improving harvesting efficiency.

Timetable for Project:

Time	Planned work	Collaborators
April 2025	Monitoring bloom condition	Wheeler-Dykes, Ferguson, Drakakaki, Wang
May 2025	Will be determined by field condition: Thinning of fruits by NAA	Wheeler-Dykes & Ferguson, Wang
May 2025	Preparation of the orchard (Pruning and Hedging)	Wheeler-Dykes & Ferguson
August 2025	Monitoring pre-harvest fruit removal force	Wheeler-Dykes, Ferguson, Drakakaki, Wang

September & October 2025	- Monitoring fruit removal force until harvest - ACC spray - Trunk shaker harvest and hand glean - Send samples for scoring (adjusted value per ton)	Wheeler-Dykes, Ferguson, Drakakaki, Wang
October & November 2025	- Compare the effect of ACC and ethephon at the cellular level (Fucshin staining; BCECF live staining; JIM5 vs JIM7 staining; M1 vs M101 staining; callose staining - Complete economic analysis of Accede applications	Drakakaki, Wang, Wheeler-Dykes, Agyeman
December 2025	- qPCR validation of candidate gene expression in ACC treated samples and horticulturally mature samples in comparison to immature samples - Chemical analysis to quantify residue ACC in the fruits; estimate the minimum effective concentration of Accede™	Drakakaki, Wang
2025	IR-4 application	Wheeler-Dykes, Ferguson, Drakakaki, Wang

Total Budget Request:

Subaward to Drakakaki:

	% of Time On Project		2025 Budget
Personnel			
project scientist (Minmin Wang)	44.4%	$95506x44.4%	$42,366
Employee Benefits		$95506x44.4%x41.3%	$17,624
Supplies and Expenses			
Items and Cost	Laboratory supplies (tubes, dyes, antibodies against polysaccharides, fixation and embedding reagents, imaging supplies)		$5,300
	qPCR experiment reagent @$10 reaction x 4 biorep x 3 tech rep x 20 genes (RNA extraction reagent, cDNA synthesis reagent, qPCR kit)		$2,000
	Materials for biochemical detection of ACC in olive fruits (glass vials and inserts, filters)		$3,000
	Use of confocal microscope at UC Davis@ $42 hour for the duration of the project, maintenance of microtome and sectioning consumables		$3,500
	Use of UC Davis FSMF for Accede residue test (@$15 per injection x 146 injections)		$2,190

Equipment (itemize when cost >$1,000)			
Items/Cost/Justification			0
Travel			
Trips/Purpose/Costs	15 daily trips between UC Davis and Nickels Soil Laboratory ($101 each trip, $75 daily fee + mileage surcharge)		$1,515
Computer Time			
Total			$77,494
Indirect Costs		@rate 11%	$8,524.34
			$86,018.34

Subaward to Wheeler-Dykes:

	% of Time On Project	2025 Budget
Personnel		
SRA/Tech	**10%**	**$7,432**
Lab Assistant		
Other		
Employee Benefits	**10%**	**$1,147**
Supplies and Expenses		**$1,000**
Adjuvant, miscellaneous field supplies, office supplies, etc.		$1,000
Equipment (itemize when cost >$1,000)		
Items/Cost/ Justification		0
Travel		**$1,147**
12 trips from UCCE Glenn office to Nickels Soil Lab (Wheeler-Dykes and Mendoza)	1,440 miles	$964.80
2 trips from UCCE Butte to Nickels Soil Lab (Dr. Agyeman)	272 miles	$182.24
Computer Time		
Overhead (where appropriate)		
Subtotal		$10,726
Indirect Costs**		$1,179.86

Total to Wheeler-Dykes		**$11,905.86**

Subaward to Leslie J. Nickels Trust:

	% of Time On Project	Request Year One
Personnel		
SRA/Tech		
Lab Assistant		
Other		
Employee Benefits		
Supplies and Expenses		**$25,500**
Spring mechanical pruning		$3,500
Spring hand pruning		$3,000
Mechanical harvester contract		$6,000
Post mechanical harvest gleaning		$6,000
Crop destruct compensation		$7,000
Equipment (itemize when cost >$1,000)		
Travel		
Computer Time		
Overhead (where appropriate)		
IR-4 Funding		**($10,000)**
Subtotal		$15,500
Indirect Costs**		$1,705.00
Total to Nickels		**$17,205.00**

Total Budget Request including subawards: $115,129.2

Scope of Work

Dr. Minmin Wang:
Collect and analyze the data. This will include collecting temperature data, doing the pull tests, and assisting with all spray treatments, participate on pruning, mechanical harvesting, sample collection and delivery to the olive receiving facility, hand gleaning, and leaf drop and supervising data entry and management, analyzing data, producing graphs, writing the preliminary report, and producing PowerPoints. She will perform the cellular analysis, develop protocls and analyze residual ACC.

Dr. Georgia Drakakaki, Becky Wheeler-Dykes, and Dr. Louise Ferguson:
Direct and supervise the trial. This will include securing the experimental orchards, preparing the experimental orchard with pruning, thinning, coordinate spraying the treatments and all mechanical harvesting and post mechanical harvest gleaning, grade sample delivery, evaluating leaf drop and write interim and final reports, and make presentations. In addition, Dr Drakakaki will oversee the cellular, structural and physiological/biochemical analysis and the publication of the results.

Lizzeth Mendoza:
Assist PIs in field operations including mechanical hedging, topping, and skirting; application of Accede®, and harvest operations. Lead pull-force testing, sample submission, and data collection from processor. Gather, enter and analyze harvest data. Assist with writing the preliminary report.

Dr. Domena Agyeman:
Assist with the economic analyses comparing complete hand-harvest, mechanical harvest with no hand-gleaning, and mechanical harvest with hand-gleaning, with and without Accede applications at the two proposed rates.

Literature Cited:

Binnie, J. E., & McManus, M. T. (2009). Characterization of the 1-aminocyclopropane-1-carboxylic acid (ACC) oxidase multigene family of Malus domestica Borkh. *Phytochemistry*, *70*(3), 348–360. https://doi.org/10.1016/j.phytochem.2009.01.002

Chauvaux, N., Van Dongen, W., Esmans, E. L., & Van Onckelen, H. A. (1997). Quantitative analysis of 1-aminocyclopropane-1-carboxylic acid by liquid chromatography coupled to electrospray tandem mass spectrometry. *Journal of Chromatography A*, *775*(1–2), 143–150. https://doi.org/10.1016/S0021-9673(97)00307-5

de Poel, B. Van, & Van Der Straeten, D. (2014). 1-aminocyclopropane-1-carboxylic acid (ACC) in plants: More than just the precursor of ethylene! *Frontiers in Plant Science*, *5*(November), 1–11. https://doi.org/10.3389/fpls.2014.00640

Mansour, R., Latché, A., Vaillant, V., Pech, J. -C, & Reid, M. S. (1986). Metabolism of 1-aminocyclopropane-1-carboxylic acid in ripening apple fruits. *Physiologia Plantarum*, *66*(3), 495–502. https://doi.org/10.1111/j.1399-3054.1986.tb05957.x

Mathooko, F. M., Yasutaka, K., Inaba, A., & Nakamura, R. (1993). Partial characterization of 1-aminocyclopropane-1-carboxylate oxidase from excised mesocarp tissue of winter squash fruit. *Scientific Reports of the Faculty of Agriculture, Okayama University.*, *82*, 49–59.

CALIFORNIA OLIVE COMMITTEE

RESEARCH PROPOSAL

Project Year: 2025 **Project Period:** Year 3 of our 2023-2025 project

Project Title: Integrating Alternate Bearing Mitigation Strategies in a Commercial Table Olive Orchard

Project Investigators: Elizabeth Fichtner and Carol Lovatt

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CL-Professor of Plant Physiology, Emeritus & Professor in the Graduate Division, Botany and Plant Sciences-072, UC Riverside, CA 92521-0124; Phone: 951-660-6730; Fax: 951-827-4437; Email: carol.lovatt@ucr.edu

Collaborator:
Kent Daane, Professor and Cooperative Extension Specialist, Department of Environmental Science, Policy, and Management, UC-Berkeley, CA, and Kearney Agricultural Research and Education Center, 9240 S. Riverbend Ave., Parlier, CA 93648: Phone: 559-646-6522; Fax: 559-646-6593; E-mail: kdaane@ucanr.edu

KD-has offered to continue sharing his expertise and advice during year 2 of our research (1) to quantify black scale populations at important stages in their life cycle, honeydew produced by black scale, and sooty molds that grow on the honeydew, (2) to rate the impact on fruit quality, and (3) to assist the PIs in statistically analyzing the data interpreting the results.

Cooperators:
Ismael Gutierrez, I.G. Harvesting, 113 E. Hickory St., Lindsay, CA 93247; Phone: 559-805-8181; E-mail: Gutierrezismael559@gmail.com

Donald Cleek, Agricultural Supervisor, UC Lindcove Research & Extension Center, 22963 Carson Avenue, Exeter, CA 93221; Phone: 559-592-2408, Ext 1157; Email: dlcleek@ucanr.edu

IG will provide the new olive orchard, with black scale present, for the research project.

DC will apply the foliar treatments (approval for DC to spray off the Lindcove REC and on the cooperators property has been secured).

Year Initiated: 2023 Anticipated Duration of Project**:** 3 crop years 2025 request: **$29,156**

Objectives of Proposed Research:

The research proposed has four objectives and goals:

(1) to test the results of our prior COC-funded research in a second commercial 'Manzanillo' table olive orchard to confirm that foliar-applied NAA at full bloom (FB) or pruning (hedging and topping) 28 days after full bloom (DAFB) to one side of the tree and then the other side of the tree every other year (*biennially*) are the best crop reduction strategies for reducing the severity of alternate bearing (based on alternate bearing index, ABI) of total yield and yield of medium + larges (M+L) size fruit and increasing yields of M+L size fruit compared to the ON-/OFF-yields of alternate bearing (untreated) control trees, trees treated with NAA at FB or pruned 28 DAFB on one side of the tree and then the other side *annually*, trees pruned on two sides of the tree in winter *annually* (grower standard practice) or trees treated with urea at FB or 10 DAFB on one side of the tree and then the other side *annually* or *biennially*. Note: all pruning treatments include topping at the time of pruning. In addition, in a second orchard, our on-going research compares the efficacy of our strategy of pruning (hedging and topping) 28 DAFB to one side of the tree and then the other side every other year (*biennially*) with this pruning treatment applied to two sides of the trees in every other row every other year. The goal is to determine the management strategy that mitigates alternate bearing (AB) and maximizes yield of commercially valuable size fruit;

(2) to use the sum of the bloom estimates on two sides of an olive tree to decide when to use a crop reduction strategy in a 'Manzanillo' olive orchard to maintain high yields of commercially valuable size M+L fruit and thereby, test how well the relationships among estimated bloom, total yield and yield of M+L size fruit from our previous COC-funded research hold up in a second orchard and to make needed adjustments. The goal is to develop a decision support tool that growers will find easy, rapid and valuable to use annually across multiple acres of table olive trees to determine whether a crop reduction strategy is required to maintain yields of M+L size fruit;

(3) to quantify the effects of crop reduction strategies, which range from pruning both sides of the tree annually, pruning one side every other year, to no pruning (alternate bearing control) on the resurgence of black scale populations, honeydew, associated sooty mold, and fruit quality in a table olive orchard. The goal is to determine, to the degree climatic conditions during the 3-year experiment permit, whether the integration of specific alternate bearing mitigation strategies in a commercial table olive orchard has the potential to positively or negatively affect black scale pest management, e.g., pesticide use, and table olive fruit quality.

Justification and Importance of the Proposed research:

Alternate bearing (AB), production of a heavy, high yield "on crop" followed by a light, low yield "off crop", is a significant economic problem. In ON-crop years, trees produce numerous small size fruit with reduced commercial value. In OFF-crop years, trees produce large fruit, in some cases too large, but there are too few fruit to provide growers with a good income. In addition, our research also documented that fruit quality is reduced in OFF-crop years. The large fruit of OFF-crop trees turn black earlier in the season in some years, which can further exacerbate the problem of too few commercially valuable fruit. For olive, the ON-crop takes longer to mature, attain size, and accumulate oil. The delayed harvest further reduces floral intensity the following spring. Alternate bearing often occurs beyond the tree or orchard level, synchronizing across geographic regions, particularly when initiated by environmental conditions that reduce crop load (i.e., heat at bloom in ‘Manzanillo’ orchards). An industry-wide shortage of fruit in the OFF-crop year has a negative economic impact on every step in the production chain from farm to consumer, including orchard management, harvesting, packinghouse and processor operations, manufacture of value-added products, marketing, and consumer prices. Taken together, the negative effects of AB jeopardize the stability and sustainability of the table olive industry.

Climate is the major factor initiating AB. Adverse climate events, such as high or low temperatures, water-deficit stress or excessive winter rain causing soil hypoxia etc., that significantly reduce yield result in an OFF crop that is followed by an ON crop. Conversely, optimal climate conditions during flowering and fruit set, such that natural fruit thinning fails to occur result in an ON crop that is followed by an OFF crop. Climate events repeat in a random manner, creating a reoccurring need for a strategy to mitigate AB and the negative economic impact of AB on table olive growers and the industry.

Results of our prior COC-funded research demonstrated that the young developing fruit of the ON crop of ‘Manzanillo’ olive trees inhibit summer vegetative shoot growth and thereby reduce the number of nodes that bear floral (inflorescence) buds the following spring (Fichtner and Lovatt, 2018; Sibbett, 2000). The developing ON crop also inhibits the transcription of key genes required for inflorescence development and flower formation (Chao, 2014; Fichtner et al., 2021). The maturing fruit of the ON-crop significantly increase abscission of floral buds for next year’s bloom starting in September, explaining reports that later harvests further reduce return bloom (Chao, 2014; Fichtner and Lovatt, 2018; Fitchner et al., 2021). For ON-crop olive trees, the negative effects of fruit set on a shoot (localized effect) are stronger than the effects of the total number of fruit (crop load) on the tree (whole tree effect) (Fitchner et al., 2021). Thus, it is the nonbearing shoots on ON-crop trees, which are in the minority, that produce the inflorescences at spring bloom following the ON-crop year (Fichtner and Lovatt, 2018). Further, a cytokinin-based strategy was only effective in stimulating summer vegetative shoot growth and increasing return bloom on nonbearing shoots of ON-crop trees (Fichtner and Lovatt 2018; Fichtner et al., 2021). Thus, a crop reduction strategy that increases the number of nonbearing shoots is necessary to mitigate the negative effects of the ON-crop in an AB olive orchard. Results of our COC-funded research identified two effective crop reduction strategies. Foliar-applied NAA at FB and pruning (hedging and topping) 28 DAFB to one side of the tree and then the other side of the tree every other year, *biennially, not annually,* evened out total annual yield, but more importantly increased the annual yield of M+L size fruit, with yields near equal in the years after the year treatment was initiated (Fitchner and Lovatt, 2023). The results documented the need for a rest period of approximately one year between treating the second side of an olive tree with NAA at FB or pruning 28 DAFB (Fichtner and Lovatt, 2023; 2024). However, it is important to note that these results were obtained in a single field experiment. Additionally, the success of these two treatments was due in part to not carrying out the scheduled crop reduction strategy in the final year of the project due the low number of inflorescences at bloom based on the sum of the estimated bloom on the east and west sides of the trees in north-south running rows. Thus, it is not only important to know when to carry out crop reduction, but also to know when crop reduction should not be done in order to maintain high yields of M+L size fruit. Before this information is recommended for implementation by table olive growers, the results need to be validated in a second experiment conducted in a new commercial table olive orchard.

Crop reduction strategies reduce yield and are thus, economically viable in high yield ON-crop years only because they increase the yield of commercially valuable size fruit. Foliar application of NAA or pruning to low bloom/low yield trees can reduce yield of M+L size fruit below the profit margin. Pruning is critical to tree crop production to open the canopy for light penetration (no light, no flowers, not fruit), to increase canopy complexity, to create new fruiting shoots, and to balance the proportion of bearing vs. nonbearing shoots to maintain yield and fruit size on an annual basis, in addition to keeping rows open for orchard management, e.g., harvesting. Pruning both sides of the tree in winter has the disadvantage that nonbearing and bearing shoots from the previous year, which have a high and low potential to flower the following spring, respectively, are not as easy to distinguish. Given the random reoccurrence of climate conditions that result in OFF or ON blooms, decisions about when, how many sides of the tree, and how frequently to use a crop reduction strategy in an orchard are critical and are best made based on a visual inspection of bloom, with knowledge of the potential impact that each decision has on the yield of M+L size fruit, not just in the ON year but in subsequent years also. Additionally, it is important to know whether the strategy selected to mitigate AB will impact table olive orchard pest management. Choices for crop reduction starting in an ON-crop year include pruning both sides of the tree *annually*, one side of the tree every other year (*biennially*) or eliminating pruning in favor of using foliar-applied NAA or urea. Thus, the crop reduction strategy selected to mitigate alternate bearing regulates the degree to which the canopy is open or closed at different times of the year and would thereby affect black scale survival, associated honeydew production and sooty mold growth, fruit quality and pesticide use.

Kent Daane, our collaborator, explained the following to us. Black scale population densities and damage has long been associated with temperatures in the olive canopy. More open canopies and more frequent pruning result in higher temperatures and drier conditions – both of which result in greater scale mortality. This relationship is also clearly associated with summer temperatures, but traditional pruning strategies are timed to post-harvest winter periods, when the scale population has already established and reached the second to third instar stages. Proposed herein are pruning strategies timed closer to June when the scale first instars have hatched and are most vulnerable to hot, dry conditions. Increasing black scale mortality at this critical period could logically reduce damage resulting from associated honeydew production and growth of sooty mold and reduce insecticide use targeting black scale.

Our on-going research project is perfect for addressing the new COC priority "Investigation of urea as a thinning agent. What is the cost and optimal application rate? Where is it currently being used?". Our research includes all the appropriate comparisons to determine the efficacy of foliar-applied urea to increase the yield of CVS M+L fruit and mitigate AB: NAA and pruning as both annual and biennial applications and an untreated ON-crop control. In addition, with the low fruit set at our Woodlake site in 2023 and 2024, we decided in the summer of 2024 to select a new orchard and save part of our 2024 COC funds to do so. As a result, the urea treatments will be correctly integrated and replicated when we lay out the experiment in the new orchard and partially paid for with our 2024 funds.

The published literature reports that the use of urea at 2%, 4%, 6%, 8% and 10% at various times, including FB, and 5, 10, 15 and 20 DAFB, was effective for olive fruit thinning (crop reduction) of ‘Manzanillo’ olive and/or other table olive cultivars (Barratta et al., 1990; Hegazi et al., 2017; Osman, 2013). In two of three studies, urea was compared to 100 ppm and 150 ppm NAA (Hegazi et al., 2017; Osman, 2013). In both cases, the authors concluded that 4% urea applied 10 DAFB was an effective fruit thinning agent for ‘Manzanillo’ olive, but 150 ppm NAA was more effective for increasing fruit size (g/fruit). [In our research, we have consistently used 165 ppm NAA (label rate).] In the 2-year study of NAA and urea on ‘Manzanillo’ olive, the various urea concentrations produced different results in years 1 and 2, and NAA was more effective than urea in both years for increasing fruit size (g/fruit) (Hegazi et al., 2017). For ‘Dolce’ table olive, 2% and 4% urea increased fruit size to medium, whereas 100 ppm and 150 ppm NAA increased fruit size to large; total yield was significantly greater in year 1 with 150 ppm NAA but in year 2, total yields for 150 ppm NAA and 4% urea were equal (Osman, 2013). Urea (4%) and NAA (150 ppm) equally increased percent oil content of fruit on both a fresh and dry weight basis (Osman, 2013). Urea (6%) applied 20 DAFB increased fruit size (g/fruit) but resulted in low yields and phytotoxic damage to ‘Nocellara del Bolice’ olive (Barratta, 1990). Treatment effects on AB and fruit size as kg/tree were not reported in these publications. The published literature provides evidence that NAA is commercially used as a fruit thinning agent to increase olive fruit size throughout the global olive industry; no similar reports were found for urea. Based on the published results, a concentration of 4% urea was chosen for the research with application times at FB and 10 DAFB to one side of the tree and then the other *annually* and *biennially*. Urea will be applied to only one side of an olive tree, rather than the whole tree, because, like NAA, urea could result in over-thinning and leaf drop in response to high temperature due to ammonia toxicity. The cost of low-biuret urea ($40/50 lbs) applied at 4% in 100 gallons of water/acre is only $26.70/acre compared to $102.14 for NAA (Liqui-Stik $326.853/gallon) applied at the label rate (165 ppm) in 100 gallons of water/acre (Source for the commercial cost of products: Nutrien Ag Solutions, San Jacinto CA). Investigation of urea as a fruit thinning agent to increase fruit size and mitigate AB is warranted based on the published results and lower cost relative to NAA.

Procedures to Accomplish Objectives:
To meet objective 1, we will conduct a field experiment in a commercial ‘Manzanillo’ olive orchard going into an ON bloom/ON-crop year. The experiment will be a randomized complete block design with 14 individual tree replications per treatment and 10 treatments that specifically meet the three objectives of the proposed research. The treatments include:

1) Untreated ON-crop control (last pruned and topped in winter 2023), e.g., alternate bearing control
2) Pruning (hedging and topping) @ 28 DAFB to one side of the tree then the other side of the tree every other year - *biennially*
3) Foliar applied NAA @ FB to one side of the tree then the other side of the tree every other year - *biennially*

4) Foliar applied NAA @ FB to one side of the tree then the other side of the tree the following year - *annually*
5) Foliar applied urea @ FB to one side of the tree then the other side of the tree the following year - *annually*
6) Foliar applied urea @ FB to one side of the tree then the other side of the tree every other year - *biennially*
7) Foliar applied urea @ 10 DAFB to one side of the tree then the other side of the tree the following year - *annually*
8) Foliar applied urea @ 10 DAFB to one side of the tree then the other side of the tree every other year - *biennially*
9) Pruning (hedging and topping) @ 28 days after full bloom to one side of the tree then the other side on a flexible schedule, using a decision support tool based on estimated sum of the bloom on the two opposing sides of the tree – *flexible schedule*
10) Control – grower standard practice of pruning two sides of the tree and topping in winter - *annually*.

The NAA and urea treatments will be applied by our cooperator D. Cleek in the equivalent of 100 gallons of water/acre as described above. Liqui-Stik Concentrate® NAA (Loveland Products) will be applied according to the label directions (165 ppm), which is identical to and has the exact same label and application rate as AMVAC's Olive Stop®, which we used in our previous research. Low-biuret urea (46% N, 0.25% biuret) will be applied at 4%. Additionally, we will treat a subset of trees with 6% and 8% low-biuret urea at 10 DAFB to determine if these concentrations of urea cause phytotoxicity and/or damage the young developing fruit. If the results in year 1 demonstrate that 4% low-biuret urea does not cause thinning sufficient to increase the yield of M+L size fruit (kg/tree) relative to NAA or pruning and 6% (or 8%) low-biuret urea are demonstrated to be nonphytotoxic, in year 2 we will increase the urea concentration to 6% (or 8% if required) for the remaining 2 years of the research. Trees are pruned as scheduled and harvested in October of each year by our cooperator I.G. Harvesting.

To meet Objective 1 and address grower questions, we added two additional treatments in a 'Manzanillo' olive orchard in Exeter (Lindcove REC) to compare the efficacy of pruning one side of the tree 28 DAFB and then the other side *biennially* to pruning both sides of the trees in every other row every other year, e. g., both sides of trees in rows 1 and 3 pruned in years 1 and 3, with both sides of trees in rows 2 and 4 pruned in years 2 and 4. There were not enough trees to include this comparison at our Woodlake site. This research is on schedule. Moreover, this orchard was added to serve as a control for testing our estimated bloom model. We know our bloom model works with Lindcove REC orchard. If it doesn't work at Woodlake and continues to work at Lindcove, then we learn that modification is required to expand its use across orchards. If the model doesn't work at either site this year, then we learn that the model's efficacy is influenced by post-bloom climate.

In all cases, total yield and fruit size distribution (kg/tree) will be determined and used to calculate yield and fruit size distribution as number of fruit per tree. Starting with harvest in Year 2, alternate bearing index (ABI) will be calculated for total yield and yield of M+L size fruit. ABI = (year 1 yield – year 2 yield)/ (year 1 yield + year 2 yield), in which yield is in kilograms of fruit per tree and the difference in yield between years 1 and 2 is expressed as an absolute number. An ABI of zero means no alternate bearing, whereas an ABI of one is complete alternate bearing, i.e., crop one year, no crop the other year (Pearce and Dobersek-Urbanc, 1967). Analysis of variance (ANOVA) will be used to test for treatment effects on bloom estimates, yield and fruit quality parameters, and ABI using the General Linear Model procedure of SAS (version 9.3; SAS Institute, Cary, NC). When ANOVA testing indicates significant differences, post-hoc comparisons will be run utilizing Fisher's protected least significant difference (LSD) test. Pearson's product moment correlation coefficients will be calculated to identify significant relationships ($r > 0.5$, $P \leq 0.05$). Significant correlations will be subjected to regression analyses, using the least squares method for the generalized linear model. The experiment is designed to determine the management strategy that maximizes yield of commercially valuable size fruit both during and after mitigation of alternate bearing, the goal of objective 1.

To meet objective 2, we will estimate the bloom on two opposing sides of the tree on a scale from 0, no bloom, to 3, heavy bloom, and calculate the sum of the bloom. The estimated sum of the bloom, based on the relationships with total yield and yield of M+L size fruit observed in our previous COC-funded research, will be used to determine when and when not to prune the trees in treatment 9. We are currently testing a threshold value of 4.5 ± 0.25. Trees with an estimated bloom above this value would require crop reduction, trees with an estimated sum of the bloom below this value would not be treated. Data collected annually will be used to analyze the relationships among bloom estimates, total yield and yield of M+L size fruit by calculating Pearson's product moment correlation coefficients to identify significant relationships ($r > 0.5$, $P \leq 0.05$) to determine how well the new data fit our earlier results. Significant correlations will be subjected to regression analyses, using the least squares method for the generalized linear model, and more sophisticated analyses as warranted. Prior results suggest that the variability in the relationship between the range in total yields that result in high yields of M+L size fruit is narrow. If the yield data from this second experiment prove this to be the case, the sum of bloom estimates on two opposing sides of the tree should be able predict when and when not to impose a crop reduction strategy to better maintain yields of M+L size fruit from one year to the next across orchards of similar size trees pruned similarly; a new relationship will likely need to be established for trees in high-density plantings. In addition, the model will indicate poor crop years when the yield of commercially valuable size fruit is best maintained by eliminating crop reduction.

The second part of objective 2 is to work with our grower cooperator and the COC to determine how to collect the data and implement the results in a manner acceptable to a grower with extensive commercial olive acreage to meet the goal of objective 2, which is to develop a decision support tool that growers will find easy, rapid and valuable to use annually across multiple acres of table olive trees to maintain yields of M+L size fruit.

To meet objective 3, in addition to selecting a new orchard going into an ON-crop year, we will select an orchard with a history of black scale, which has not been treated in recent years, and with visual proof of black scale presence. Black scale population numbers were monitored in Years 1 and 2 and will be monitored in the spring and fall of each year in the new orchard. The presence of honeydew droplets on olive leaves in March and April, which correspond to a rapid increase in scale size, is often the earliest signal of increased scale density in the orchard. In April, the terminal ends [20 inches long (about 50 cm)] of four branches will be monitored on eight trees (replications) per treatment and honeydew and sooty mold accumulation will be rated on a 0-3 scale (0 = no honeydew, 1 = presence of honeydew, 2 = honeydew and sooty mold on < 30% of the branch, and 3 = honeydew and sooty mold on > 30% of the branch. In April and October, the scale density will be evaluated by counting the number of mature scales (third instar to adult) on the terminal ends of the four branches on eight trees (replications) per treatment. For categorical ratings of scale honeydew and sooty mold accumulation, treatment effects will be compared in a 2 by 2 contingency table with treatments separated using Pearson's Chi-square test. Scale densities will be compared using the General Linear Model function, with treatments separated using Tukey or Dunnett Pairwise comparison. We also calculated the net change in honeydew and sooty mold ratings and black scale numbers from April to October and year to year and will do this in the new research orchard.

The treatments in our experiment provide a range in canopy openness and closure and pruning times, which combined with our detailed analyses of black scale at two periods in the life cycle, plus honeydew and sooty mold, will enable us to determine whether the integration of specific AB mitigation strategies in a commercial table olive orchard have the potential to positively or negatively affect black scale pest management, e.g., pesticide use, and table olive fruit quality (goal 3).

An analysis of the cost of each treatment versus the increase in yield of CVS M+L fruit, with and without savings in black scale management, will be completed at the end of the research project.

Timetable for Project:

The research proposed addresses the economic problem of alternate bearing. The goal is to document that the putative best crop reduction strategies are ones applied every other year. The orchard we selected in Woodlake, despite having an ON bloom in 2023 and 2024, produced low yields in 2023 and 2024. Observing the poor set in summer of 2024, we decided to obtain a new 'Manzanillo' olive orchard to complete our research, rather than risk a third year with limited results. Thus, we did not spend all of the 2024 budget allocated to UCR, which has enabled us to reduce UCR's year 3 (2025) budget. Due to the cost of establishing the research, which now includes four treatments to test the efficacy of urea, in the new orchard, the ANR budget has increased. Two additional years of funding will be required to complete the research in the new orchard, including the addition of the new COC priority testing the efficacy of foliar-applied urea as a fruit-thinning agent to increase fruit size and mitigate AB. We plan to submit typical budgets for these two added years, approximately $30,000 to $32,000 per year, depending on ANR salary increases and cost increases at the Lindove REC. No problem was encountered with the research being conducted at the Lindcove REC, which compares pruning one side of the tree and then the other side *biennially* with pruning both sides of the trees in every other row every other year. This research is anticipated to be completed in June 2026 with a no cost extension through June 2026. All treatments will be applied in 2025 in the new orchard going into an ON bloom (with a known black scale problem) beginning in February with the winter pruning treatment (control – standard grower practice). NAA will be applied at FB, low-biuret urea will be applied at FB and 10 DAFB, and pruning treatments will be applied 28 DAFB. The pruning treatments at Lindcove will also be applied 28 DAFB. If the results in year 1 demonstrate that 4% low-biuret urea does not cause thinning sufficient to increase the yield of M+L size fruit (kg/tree) relative to NAA or pruning and 6% (or 8%) low-biuret urea are not phytotoxic, then in year 2 we will increase the urea concentration to 6% (or 8% if required) for the remaining 2 years of the research. The treatment application times will remain the same for each year that they are applied. Bloom estimates will be completed each year just prior to full bloom, statistically analyzed, and then used to evaluate whether a treatment should be applied or not, but the decision will be implemented only in treatment 9. Harvests will be in October each year with fruit samples collected one to two days before harvest for the analysis of fruit size and quality. All yield data will be statistically analyzed to determine the significance of treatment effects on all yield parameters (as described above). After harvest each year, the relationships among the sum of the bloom estimate, total yield and yield of M+L size fruit will be analyzed (as described above). A decision support tool will be evaluated across commercial table olive acreage for speed and accuracy, using the current or refined model, based on data obtained from the new orchard compared with current and prior results obtained at Lindcove. Each year in April and October, the presence of honeydew droplets and sooty mold, and the number of mature scales (third instar to adult) will be quantified on four branches on eight trees (replications) per treatment. All results, with the possible exception of the yield data and final black scale data to be collected in October, will be presented in the Interim Progress Report in October, with the final black scale results and further statistical analysis of the relationships among sum of the bloom estimates, total yield and yield of M+L size fruit included in the final report in June the following year (with approval of a no cost extension).

<u>Present Outlook and Estimated Success in Accomplishing the Objectives:</u>
<u>Objective 1.</u> The present outlook is that we will secure a new orchard going into an ON-bloom year that will produce an ON crop and has a history and visual presence of black scale to successfully complete objective 1 (and objective 3), which includes the COC's new priority, "Investigation of urea as a thinning agent. What is the cost and optimal application rate? Where is it currently being used?"

Research at Lindcove, which compares pruning one side of the tree and then the other side *biennially* with pruning both sides of the trees in every other row every other year is on schedule. In 2023, the ON-bloom (sum of the 2 sides of the trees > 5.1-6.1) but resulted in an OFF-crop and the sum of the bloom was not correlated with total yield or yield of M+L size fruit; whereas in 2024, the sum of the bloom was was less than 4.5 ± 0.25, resulted in a greater crop than in 2023, and was significantly correlated with both total yield (r = 0.72, P = < 0.0001) and yield of M+L size fruit (r = 0.58, P = < 0.0001), consistent with a post-bloom climate event in 2023. Total yield was predictive of the yield of M+L size fruit in both years (2023 r = 0.74, P = < 0.0001; 2024 r = 0.73, P = < 0.0001). There was a slight trend towards greater annual and 2-year cumulative total yields and yields of M+L size fruit for trees pruned on one side and then the other *biennially* compared to pruning both sides of the trees in every other row every other year. However, at the end of 2 years the yield differences were not statistically significant (Table 1). Over the 2 years of the experiment, there were no treatment effects on the severity of alternate bearing based on ABI. At harvest in 2024, there were no significant effects due to pruning strategy on the percentage of green fruit per tree; green fruit comprised 86% and 82% of the crop on trees pruned on one side and then the other *biennially* compared to pruning both sides of the trees in every other row every other year, respectively. Logically, it is less costly to prune only one side of the tree and then the other side *biennially*, with no pruning expense every other year, compared to pruning both sides of half the trees in an orchard annually. For the research at Lindcove, we will be able to report the best pruning strategy for mitigating AB and improving yield of M+L size fruit. In the new orchard, we will be able to report the best management strategy (among NAA, urea, and pruning treatments) for mitigating AB and increasing and sustaining the yield of CVS M+L fruit and to compare the cost of each treatment relative to annual and cumulative yields of CVS M+L fruit and the uniformity of these yields across years based on ABI. Two additional years of funding will be required to support the research initiated in 2025 in the new replacement orchard.

Objective 2. Our present outlook is that our current approach of using the sum of estimated bloom to decide whether or not to implement a crop reduction strategy will work effectively in the new orchard. In our previous research, trees with sum of the bloom estimates on opposing sides of the tree between 4.5 ± 2.5 resulted in higher yields of M+L size fruit than trees with lower or higher estimated sum of the bloom estimates (Fichtner and Lovatt, 2023). We anticipate comparable results and that the additional bloom and yield data will enable us to better define this range and improve our efforts to develop a reliable decision support tool. The more difficult aspects of objective 2 are (i) the development of a sampling method that can be used to rapidly evaluate the bloom status for a large portion of a block that is sufficient to make a decision about crop reduction for that area, and (ii) how large a portion of the orchard needs to be represented by an evaluation procedure and how much time a grower is willing to allocate to doing the evaluation to use the decision support tool. The addition of the second orchard at Lindcove provides a control for testing our estimated bloom model that ensures our success. We know our bloom model works in the Lindcove REC orchard. If it doesn't work in a new orchard but continues to work at Lindcove, then we learn that modification is required to expand its use across orchards. If the model doesn't work at either site in a specific year, then we learn that the model's efficacy is influenced by post-bloom climate.

Objective 3. Our ability to successfully meet objective 3 depends on climate conditions being conducive to black scale development early in the season and sufficiently hot to cause scale mortality on trees with open canopies in summer. We will select an orchard with a history of black scale that has not been treated for black scale in recent years. We think this objective addresses an aspect of importance when deciding which, if any, crop reduction strategy to implement in an alternate bearing orchard where black scale is a potential problem. This objective adds minimal additional cost to the proposed research. The treatments necessary to meet objective 1 provide a progression from open to closed canopies that are perfect for meeting objective 3. Moreover, our collaborator Kent Daane, Professor and Cooperative Extension Specialist, is an expert on black scale. Dr. Daane has agreed to continue to provide his expertise and advice (1) to quantify black scale populations at two stages in their life cycle based honeydew produced by black scale and sooty molds that grow on the honeydew in April and again in October along with quantifying the number of mature black scale, (2) to rate the impact on fruit quality, and (3) to assist us in analyzing the data and interpreting the results, i.e., economic impact. Thus, the only additional cost is the additional salary hours for the technical staff.

Budget summary by Objectives: Note that the Year 3 budget requested by UCR is less than for Year 2, because we did not expend our full budget in year 2 at Woodlake. Despite an ON bloom, the trees set too few fruit to warrant harvesting the experiment. We also did not collect honeydew, sooty mold or black scale data at harvest due to low values in April of Year 2. Residual funds from 2024 will be used to partially cover the costs of treatment applications, data entry, statistical analyses, and bloom model testing in 2025. The ANR budget increased due to the person-hours needed to layout the experiment in a new orchard (tagging trees by treatment and replication, and tagging shoots for honeydew, sooty mold and black scale evaluation).

Objective 1 accounts for the majority of the budget needs for each PI in each year of the project as follows:

Year 1 (2023)

119 trees were tagged and harvested in Woodlake; 64 trees at Lindcove

34 trees had foliar treatments applied at two separate application times in Woodlake

68 trees were pruned in Woodlake, 64 trees in Lindcove

4 shoots on 56 trees were evaluated for honeydew, sooty mold, and black scale in April and October in Woodlake

100% of salaries for technical assistants were used for objectives 1, 2 and 3 divided as 75%, 0% and 25%, respectively; 100% of G. Klein's contract hours were used for data entry, management, statistical analyses, model testing, and data presentation divided as 50% to meet Objective 1, 40% to meet Objective 2, and 10% to meet Objective 3.

Year 2

119 trees were NOT harvested in Woodlake; 64 trees were harvested at Lindcove

34 trees had foliar treatments applied at two separate application times at Woodlake

68 trees were pruned in Woodlake; 64 trees were pruned in Lindcove

4 shoots on 56 trees are evaluated for honeydew, sooty mold, and black scale in April, but NOT in October, at Woodlake

100% of salaries for technical assistants were used for objectives 1, 2 and 3 divided as 50%, 25% and 25%, respectively; 50% of G. Klein's contract hours were used for data entry, management, statistical analyses, model testing and data presentation divided as 50% to meet Objective 1, 40% to meet Objective 2, and 10% to meet Objective 3.

Year 3

140 trees will be tagged, treated and harvested in a new orchard plus 30 trees to evaluate foliar-applied urea at 6% and 8%; 64 trees will be pruned and harvested at Lindcove

84 trees (total) will have foliar treatments applied at two separate application times in a new

orchard
42 trees will be pruned in a new orchard and 64 trees will be pruned in Lindcove
4 shoots on 70 trees will be evaluated for honeydew, sooty mold, and black scale in April and October in the new orchard
100% of Salaries for technical assistants will be used for objectives 1, 2 and 3 divided as 50%, 25% and 25%, respectively; 100% of G. Klein's contract hours will be used for data entry, management, statistical analyses, model testing and data presentation divided as 50% to meet Objective 1, 40% to meet Objective 2, and 10% to meet Objective 3.

Literature Cited:

Barratta, B., Caruso, T., Crescimanno, P.L, Inglese, P. 1990. Using urea as thinning agent in olive: the influence of concentration and time of application. Acta Hort. 286:163-166.

Chao, Y.Y. 2014. Alternate Bearing in Olive (*Olea europaea* L.). MS Thesis. University of California, Riverside, CA.

Fichtner, E., Lovatt, C.J. 2018. Alternate bearing in olive. Acta Hortic. 1199:103-108. doi:10.17660/ActaHortic.2018.1199.17and PGRs

Fichtner, E., Lovatt, C.J. 2023. Alternate bearing in olive - *Mitigation with properly timed foliar-applied naphthaleneacetic acid or pruning.* Acta Hortic., In review.

Fichtner, E. and Lovatt, C. 2024. Yield benefits from a new strategy using naphthaleneacetic acid to manage olive crop load. Progressive Crop Consultant, Topics in Subtropics Newsletter; UCCE SJV Website. https://ucanr.edu/blogs/blogcore/postdetail.cfm?postnum=60638.

Fichtner, E.J., Y.Y. Chao, L. Ferguson, J.S. Verreynne, L. Tang and C.J. Lovatt. 2021. Repeating cycles of ON and OFF yields in alternate bearing olive, pistachio, and citrus trees — *Different mechanisms, common solutions*. Acta Hortic. 1315. DOI 10.17660.

Hegazi, E.S., Hegazi, A.A., El Kholly, D.M. 2017. Effect of some chemical thinning agents on fruit quality and oil content of Manzanillo and Eggizi Shami olive cultivars. J. Plant Production, 8(2):315-320.

Osman, I.M.S. 2013. Effect of different concentrations of foliar applications of urea, NAA and ethrel on fruit thinning of 'Dolce' olive cv. Egypt. J. Hort. 40(1):133-142.

Pearce, S.C. and S. Dobersek-Urbanc. 1967. The measurements of irregularity in growth and cropping. J. Hort. Sci. 42(3):295–305.

Sibbett, S. 2000. Alternate bearing in olive trees. California Olive Oil News. 3(12),1

Table 1. Effect of pruning (hedging and topping to 14 feet) 28 days after full bloom (DAFB) to one side of the tree and then the other side *biennially versus* pruning 28 (DAFB) on both sides of the trees in every other row every other year on annual and 2-year cumulative total yield and yield of commercially valuable medium plus large (M+L) size fruit as kg/tree averaged across treated and untreated rows as specified below for 2023 and 2024. Trees were harvested in October each year.

Treatments	2023 total yield (M+L size fruit) (kg/tree)	2024 total yield (M+L size fruit) (kg/tree)	2023-2024 cumulative total yield (M+L size fruit) (kg/tree)
Pruned 28 DAFB on west side of the tree in 2023; pruned on the east side in 2025 (no trees were pruned in 2024	56 a (7 a)	101 a (44 a)	158 a (52 a)
Pruned 28 DAFB on both sides of the tree in years 2023 and 2025 and the other row pruned 28 DAFB on both sides of the tree in 2024	46 a (4 a)	91 a (36 a)	142 a (40 a)
P-value	0.2916 (0.2861)	0.3521 (0.1847)	0.6335 (0.1295)

[z] In 2023, trees were pruned and topped on June 20, 2023, and harvested in October. In 2024 trees were pruned and topped on June 3, 2024, and harvested in October.

[y] Mean values within a vertical column followed by different letters are significantly different at the specified *P* level by Fisher's Protected LSD test.

University of California, Riverside Budget
Project Period: **01/01/25-12/31/25**

ITEM	2025 Budget
PERSONNEL:	
Personnel Subtotal:	**0**
FRINGE BENEFITS:	
	0
	0
SUPPLY/SERVICES, OTHER	
Field work: Individual contractors: (1) Recharge to Lindcove REC – spraying off-site; I.G. Harvesting (billed through Lindcove REC - pruning treatments and harvesting data trees = $4,500; (3) Independent contractor-Grant Klein-statistician-data entry, statistical analyses, model design and testing, tables and figures $3,000.	7,500
Services Subtotal:	**7,500**
TRAVEL:	
Field Work: Vehicle Use: four roundtrips UCR to Woodlake and Lindcove (512 mi x 4 = 2,048 mi x $0.677/mi) = $1,387	1,387
Lodging; $187 x 3 nights = $561	561
Meals: $79/day x 3 days = $237	237
Travel Subtotal:	**2,185**
CONTRACTUAL	
Subaward to Elizabeth Fichtner	18,406
Contractual Subtotal:	**18,406**
UCR TOTAL DIRECT COSTS w/o exclusions (figure out IDC):	**9,685**
UCR INDIRECT COSTS @ 11% TDC:	**1,065**
UCR TOTAL DIRECT COSTS - to PI Carol Lovatt:	**10,750**
UCR + ANR Grant TOTAL:	**29,156**
UCANR	**18,406**
UCR Budget	**10,750**
UCANR + UCR	**29,156**

University of California, ANR Budget
Project Period: **01/01/25-12/31/25**

ITEM	2025 Budget
PERSONNEL:	
Research assistant $61,300/year @ 16.66% time = $10,213	10,213
Personnel Subtotal:	**10,213**
FRINGE BENEFITS: 41% x $10,213 = $4,187	4,187
Fringe Benefits Subtotal:	**4,187**
SUPPLY/SERVICES, OTHER	1,776
Field work: Lindcove REC: Field assistants for tagging trees, collecting fruit samples, analyses of fruit size and quality, and harvest (48 person-hours x $37/person-hour) = $1,776	
Services Subtotal:	**1,776**
TRAVEL:	
COC work: Vehicle Use: Ten roundtrips to orchards in Woodlake and Lindcove (600 mi round trip @ $0.677/mi = $406)	406
Other Direct Costs (GAEL)	
Travel Subtotal:	**406**
CONTRACTUAL	
Contractual Subtotal:	**0**
ANR Total Direct Cost	**16,582**
Exclusion amount (Subs)	**0**
ANR TOTAL DIRECT COSTS w/o exclusions (figure out IDC):	**16,582**
ANR INDIRECT COSTS @ 11% TDC:	**1,824**
ANR to PI Elizabeth Fichtner Grant TOTAL:	**18,406**

University of California, ANR Budget
Project Period: **01/01/23-12/31/25**

ITEM	2023-25 Budget
PERSONNEL:	
Yr 1 Salary *(Junior Specialist at 9% FTE)*	4,495
Yr 2 Salary *(Junior Specialist at 9% FTE)*	7,731
Yr 3 Salary *(Junior Specialist at 9% FTE)*	10,213
Personnel Subtotal:	**22,439**
FRINGE BENEFITS:	
Yr 1 Benefits (*38.9%*)	1,748
Yr 2 Benefits (*38.9%*)	0
Yr 3 Benefits (*38.9%*)	4,187
Fringe Benefits Subtotal:	**5,935**
SUPPLY/SERVICES, OTHER	
Yr 1 Field work	1,440
Yr 2 Field work	1,440
Yr 3 Field work	1,776
Services Subtotal:	**4,656**
TRAVEL:	
Yr 1 Travel	3,203
Yr 2 Travel	656
Yr 3 Travel	406
Travel Subtotal:	**4,265**
CONTRACTUAL	
Contractual Subtotal:	**0**
ANR Total Direct Cost	**37,295**
Exclusion amount (Subs)	**0**
ANR TOTAL DIRECT COSTS w/o exclusions (figure out IDC):	**37,295**
ANR INDIRECT COSTS @ 11% TDC:	**4,102**
ANR to PI Elizabeth Fichtner Grant TOTAL:	**41,398**

Approved by:

PRIMARY PI SIGNATURE PAGE: UNIVERSITY OF CALIFORNIA

______________________________	10/24/2024
Originator's Signature	Date
______________________________	____________
Department Chair/County Director	Date
______________________________	10/30/2024
Liaison Officer	Date

SUBCONTRACT SIGNATURE PAGE: UNIVERSITY OF CALIFORNIA

______________________________ 10/28/24
Originator's Signature Date

Karmjot Randhawa 10/28/24
Department Chair/County Director Date

______________________________ _10/28/2_____
Liaison Officer Date

California Olive Committee

Research Proposal

Project Year: 2025 **Project Period: 2 years**

Title: Designing a web app for predicting risk of olive fruit fly—a tool for California olive growers and pest control advisors

Project Investigators

1. Elizabeth Fichtner, PhD. UC ANR Farm Advisor, Tulare/Kings Counties
2. Santosh Bhandari, MS. UC ANR Associate Specialist, UCCE Tulare County

Project Collaborators

1. Becky Wheeler-Dykes, PhD. UC ANR Farm Advisor, Glenn, Tehama, and Colusa Counties
2. Sudan Gyawaly, PhD. UC ANR IPM Advisor, Area IPM Advisor, Butte, Sutter, Glenn, Colusa, Tehama Counties.
3. Cindy Kron, PhD. UC ANR IPM Area Advisor, North Coast.
4. Kent Daane, PhD. Professor of Cooperative Extension, UC Berkeley

Project Cooperators

1. Jim Stewart, Ag IPM Consultants, Inc.
2. Bert Quezeda, Ag IPM Consultants, Inc.
3. Ernie Simpson

Summary

Olive fruit fly (*Bactrocera oleae*) (OLFF) is the most important insect pest of olive worldwide. The California Olive Committee has funded long term pest management programs in the Sacramento Valley and southern San Joaquin Valley, generating a large dataset available for analysis of the impact of weather variables on pest activity. This data can be analyzed and used to develop a model to assist growers with assessing pest risk based on anticipated weather parameters, thus allowing for assessment of risk prior to the physical detection of the insect in orchards. The goal of this proposal is to evaluate historic OLFF trapping data to develop an algorithm and web-based app using Python programming language that can predict future populations of OLFF based on the input of weather variables. This model would be posted on the UC ANR Fruit and Nut Research and Information Center and the California Olive Committee (COC) websites for use by olive growers, ranch managers, and pest control advisors, thus allowing for enhanced timing of insecticide applications based on predicted risk of pest populations. As a proof of concept, a preliminary model was constructed using regional weather data obtained from California Irrigation Management Information System stations as correlative variables with historic OLFF populations generated from an industry-sponsored trapping program. This preliminary model functions at approximately 77% accuracy but requires orchard-level collection of weather data in concert with OLFF trapping to improve accuracy for industry adoption. Having developed the skill to develop a predictive model, we now request funding from the COC to install dataloggers at 10 OLFF trapping sites, managed by our industry partners (Ag IPM Consultants) and Ernie Simpson, to create a fine-

tuned predictive model based on orchard-level weather patterns and corresponding OLFF populations. This project integrates entomology and computer programming to support pest management decisions made by growers, orchard managers and pest control advisors.

Objectives of Proposed Research:

1. Evaluate historic trends in OLFF populations by analysis of trap-catch data from COC-sponsored OLFF monitoring programs.
2. Collect in-orchard weather data concurrent with OLFF trap counts for model development.
3. Develop an algorithm and web-based app using Python programming language that can predict future populations of OLFF based on the input of weather variables.

Justification and Importance of Proposed Research

California's 34,000 acres of olives represent 99% of American olive production with a total value of $63 million (CDFA, 2023). Olive fruit fly (OLFF) (*Bactrocera oleae*) has become the most important insect pest of olive since it's introduction to Los Angeles in 1998 (Rice et al 2003). It is now established in all commercial olive growing regions of California. OLFF adversely affects olive oil quality; however, orchards grown for oil have higher thresholds of tolerance for the pest than table olive orchards. Because OLFF oviposits in the fruit resulting in larval infestation, the black ripe table olive industry has zero tolerance for the pest.

In response to the introduction of the OLFF to California, since 2001 the California Olive Committee (COC) has supported an ongoing monitoring program to inform growers of OLFF seasonal activity in two olive-growing regions of the state. The purpose of the program is to facilitate improved insecticide application timings by informing growers of OLFF population trends. Weekly trap counts of male and female OLFF are recorded and the data are shared with growers and pest control advisors (PCAs) by email. In the southern San Joaquin Valley (SSJV), the trapping program utilizes yellow sticky traps baited with pheromone from Alphascents (Marylhurst, OR, USA). In the Sacramento Valley, the trapping program utilizes wet traps. Because the trapping programs in the north and south part of the state use different trapping strategies, statistical comparisons can't be made between regions. The SSJV trapping program has utilized identical trapping methodology at 10 sites over 16 years (2007-2022), thus generating a large historical dataset. After communicating with the Glenn County Ag Commissioner's office and the California Olive Committee, we have been able to obtain and compile the Sacramento Valley OLFF trap data from 2012-2022.

The development of a predictive model leverages the 20+ years of historic OLFF trapping data funded by the COC to allow growers to predict risk of OLFF prior to the first detection of the pest in traps. The use of a predictive model may reduce the frequency of insecticide applications, thus reducing production costs and improving the longevity of insecticide efficacy by reducing selection pressure on pest populations. Pest management options have become increasingly limited as insecticides have lost registration (ie. Supracide, an organophosphate) due to associated environmental and human-health risks. The three main current control strategies for OLFF all have shortcomings that either affect product efficacy or adversely influence non-target species. GF-120 (Spinosad) is an organically-labeled product that serves as an industry standard, but resistance has already been reported in the OLFF populations in Europe and California (Kakani et al. 2010). Danitol 2.4 EC, a pyrethroid, is a conventional chemistry that may impact non-target insects, including beneficials, and has documented toxicity to aquatic systems, particularly fish. Kaolin clay is used to deter OLFF oviposition; however, it clogs the spiracles of non-target insects, including beneficial insects (Pasqual et al. 2010).

In 2024, we conducted a preliminary analysis of SSJV OLFF population trends in relationship to local weather variables, weekly average air temperature, humidity, and windspeed corresponding to historic trapping intervals were secured from the online California Irrigation Management Information System (CIMIS) database. Data from two weather stations near trap locations were utilized to perform a simple linear regression model to evaluate the influence of weather parameters on OLFF populations.

To make a preliminary model, we used Random Forest, a commonly used supervised machine-learning approach, for training on the most recent weather and OLFF population data collected in the SSJV from 2019 to 2023. Random Forest is a predictive model in Python used to determine the likelihood of occurrence of an event based on historical data. The model, when trained, detects specific patterns from the past and responds to new entries based on the developed trends. Training of data requires that the model works on specific classification algorithms that can vary in their ability to use supervised or unsupervised learning approaches. Supervised learning uses the labelled data sets for training algorithms to predict outcomes. Unsupervised learning does not require human intervention. Trained with the labelled data sets, Random Forest classifier was used to predict the presence or absence of OLFF, and regression classifier was used to predict the number of male and female flies. Eighty percent of the total data volume was used for training and 20% was used for testing model efficiency. The precision and accuracy percentages were recorded and cross-validation was made. We conducted an analysis of randomly selected two years of data (2011 and 2012) using the model and determined that the model had an accuracy of 77% for predicting female fly populations in the SSJV in 2013. The design of this preliminary model provided us an opportunity to develop the skills needed to create an improved, deployable prediction model based on in-orchard weather data collected concurrent with OLFF trap data.

Procedures to Accomplish Objectives:

Objective 1: Historic olive fruit fly trapping data has been obtained from COC and Ag IPM Consulting Inc records and compiled into Excel to allow for statistical analyses. In the southern San Joaquin Valley, historical trap records have been compiled from 2001 to 2022. From 2001 through 2006 trap sites spanned from Bakersfield to Madera; however, since 2007 the trapping sites were restricted to Tulare County. Additionally winter sampling (December-March) was discontinued in 2007. Historic OLFF data from the Sacramento Valley was obtained from the COC, with data available from 2012-2022. For the ease of uniformity, preliminary descriptive and inferential statistics were performed on 16 years (2007-2022) of male and female population data. These analyses allow for identification of sites with historically high OLFF populations, thus identifying the ideal sites for future in-orchard microclimate data compilation in concert with OLFF trapping (ie. objective 2). We additionally propose to evaluate ratios of male and female OLFF in traps over time as well as the location by year interactions of trap catch data. The analyses will be useful in understanding the population distribution of the olive fruit fly across different years and locations and for future pest-management considerations. It also provides a historical record of olive fruit fly population dynamics that will allow future consideration of the impact of climate trends on pest populations.

Objective 2: Development of concurrent in-orchard trap catch and weather data. To transition our preliminary model (based on regional CIMIS data) to a deployable model for industry use, in March-October 2025 we propose to establish HOBO dataloggers at 10 sites that house the OLFF-trapping program, with five dataloggers installed at Sacramento Valley trap sites and five at SSJV trap sites. These dataloggers will be installed at sites with historically high OLFF populations, based on the analyses in objective 1. These dataloggers will allow us to gather in-orchard estimates of daily high and low temperatures, and relative humidity. These weather parameters will be utilized to improve our prediction model. OLFF populations are sensitive to climate trends as evidenced by the reduction in adult flies trapped in July and August and resurgence of populations in September when temperatures are more moderate (Yokoyama et al. 2008). OLFF are inactive at temperatures below 62°F (Avidov 1954) and normal flight and oviposition activity occur at temperatures between 73°F and 84°F (Johnson et al. 2011). At temperatures above 95°F they become motionless (Johnson et al. 2011); consequently, temperatures above 95°F are associated with reduced trap counts and mortality is associated with temperatures above 100°F (Wang et al. 2009a and b).

Objective 3: Improved Model Development and Deployment. The weather data captured by the HOBO dataloggers combined with the OLFF trap data will be utilized to make a predictive model. We will use Random Forest, a predictive model in Python to determine the likelihood of occurrence of an event based on historical data. The model, when trained, detects specific patterns from the past and responds to new entries based on the developed trends. Training of data requires that the model works on specific classification algorithms that can vary in their ability to use supervised or unsupervised learning approaches. Supervised learning uses the labelled data sets for training algorithms to predict outcomes. Unsupervised learning does not require human intervention. Trained with the labelled data sets, Random Forest classifier was used to predict the presence or absence of OLFF, and regression classifier was used to predict the number of male and female flies.

Project Timeline

Objective 1: Compilation and analysis of historic OLFF data will be completed in winter-spring 2025. These analyses will be used for site selection for objective 2. Analysis of historic data will be included in the interim report.

Objective 2: Install HOBO dataloggers at 10 sites in March 2025. Dataloggers will be monitored approximately monthly throughout the trapping season. At the conclusion of the trapping season (November 2025), trap catch data and correlative weather variables will be compiled for analysis into the winter of 2025/2026.

Objective 3: Model development will be completed in winter 2025/2026, pending a request for a no cost extension through June 30, 2026. A preliminary model should be available for testing during the 2026 trapping season.

Year 2: We will be requesting similar funds in 2026 to gather a second season of trap catch and in-orchard weather data for model improvement and testing.

Present Outlook and Estimated Success in Accomplishing Objectives:

Our team has already assembled and analyzed historic OLFF data from the SSJV from 2007-2022; consequently, we need to add the 2023 and 2024 OLFF trapping data to the study. We have also compiled the historic data from the Sacramento Valley from 2012-2022 and will begin analyzing this data in winter 2025/2025. We have also developed a preliminary predictive model using weather data collected from CIMIS stations. This preliminary model has 77% accuracy when tested on historical data. Consequently, we are confident that in-orchard microclimate data corresponding to OLFF population data will result in model improvement. Last, we have determined how to use Google Cloud to deploy the model.

Anticipated Deliverables, Outcomes, and Impacts. By gathering in-orchard weather data concurrent with OLFF trapping data, we propose this integrative project, utilizing computer programming skills and entomology, to make a web app for stakeholder use. The app will be posted on the UC ANR Fruit and Nut Research and Information Center and COC websites. The prediction model is designed to reduce the workload of PCAs, reduce pesticide use (including chemistries toxic to non-target organisms, including humans), and thus have a broader positive impact for the public. Incorporation of predictive model use into the management strategy for OLFF may reduce the number of traps needed for insect monitoring and/or the frequency of trap monitoring required for effective pest management. Employment of the predictive model may allow growers and PCAs to make an informed decision of timing of insecticide application.

Budget Summary by Objectives:

Objective 1: We request 2 weeks of salary for S. Bhandari, Assistant Specialist, for compilation, analysis, and summarization of historic OLFF.

Objective 2: We request funding for travel to/from field sites for HOBO installation and maintenance during the trapping system ($487 Fichtner; $500 Gyawaly; $500 Wheeler-Dykes), as well as funds for supplies ($500 Gyawaly; $500 Wheeler-Dykes).

Objective 3: We request 7 weeks of salary for S. Bhandari for model development and testing, specifically Python programming.

References

Avidov, Z. 1954. Further investigations on the ecology of the olive fruit fly (*Dacus oleae*, Gmel.) in Israel. Ktavim Q. J. Agric Res Stn. Beit Dagan Rehovot Isr. 4: 39-50.

California Department of Food and Agriculture. 2023. California Agricultural Production Statistics. www.cdfa.ca.gov/Statistics

Johnson M., Wang, X., Nadel, H., Opp, S., Lynn-Patterson, K., Stewart-Leslie, J., Daane, K. 2011. High temperature affects olive fruit fly populations in California's Central Valley. California Agriculture, 65(1), 29-33.

Kakani, E.G., Zygouridis, N.E., Tsoumani, K.T., Seraphides, N., Zalom, F.G. and Mathiopoulos, K.D. (2010), Spinosad resistance development in wild olive fruit fly *Bactrocera oleae* (Diptera: Tephritidae) populations in California. Pest. Manag. Sci., 66: 447-453.

Pasqual, S., Cobos, G., Series, E., Gonzales-Nunes, M. 2010. Effects of processed kaolin on pests and non-target arthropods in a Spanish olive grove. J Pest Sci 83: 121-133.

Rice, R., Phillips, P., Stewart-Leslie, J., and Sibbett, G. Olive fruit fly populations measured in Central and Southern California. California Agriculture, 57(4), 122-127.

Wang, X.-G., Johnson, M.W., Daane, K.M., Nadel, H. 2009. High summer temperatures affect the survival and reproduction of olive fruit fly (Diptera: Tephritidae). Environmental Entomology 38: 1496-1504.

Wang, X.-G, Johnson, M.W., Daane, K.M., Opp, S. 2009. Combined effects of heat stress and food supply on flight performance of olive fruit fly (Diptera: Tephritidae). Annals of the Entomological Society of America 102: 727-734.

Team Member Roles and Scopes of Work

Elizabeth J. Fichtner, PhD: As a UC ANR farm advisor, Fichtner has developed relationships with the COC, local olive growers, and Ag IPM Consultants. She is responsible for gathering the historic data, liaising with the local OLFF trapping program and organizing the data into a useable form.

Santosh Bhandari, MS. Assistant Specialist, UC ANR: Bhandari has his MS in Entomology and is a skilled Python programmer and biostatistician. Bhandari has developed the preliminary model based on historic OLFF trap data and CIMIS data.

James Stewart and Bert Quezada, Ag IPM Consultants, Inc.: Stewart and Quezada have run the COC-sponsored OLFF trapping program in the SSJV since its inception. Stewart and Quezada have collaborated with Bhandari and Fichtner on the development of this project since spring 2024.

Ernie Simpson: Manger of Sacramento Valley Olive Fruit Fly trapping program. Will share location of 5 trapping sites with historically high olive fruit fly counts to enable our team to install dataloggers.

Becky Wheeler-Dykes, PhD: As a UCCE Farm Advisor assigned to olives in the Sacramento Valley, Wheeler-Dykes will liaise with the Sacramento Valley olive fruit fly trapping program to install dataloggers in her region assist with the development of future extension efforts to make the product available to growers, PCAs, and ranch managers.

Sudan Gyawaly, PhD: As an area IPM Advisor in the Sacramento Valley, Dr. Gyawaly will also liaise with the Sacramento Valley olive fruit fly trapping program to install dataloggers in his region and assist with the development of future extension efforts to make the product available to growers, PCAs, and ranch managers.

Kent Daane, PhD: As a Professor at UC Berkeley, Daane has a research background working with OLFF with extensive experience elucidating the ecology and biocontrol of OLFF in California.

Cindy Kron, PhD: As a UC IPM Advisor working on olive fruit fly management in oil olive orchards, Dr. Kron will provide academic support in the future development of this project and will serve as a hands-on collaborator if funds are secured for similar research in oil olive systems.

Budget

01/01/2025-12/31/2025

Fichtner-UCCE Tulare County		
Item	**2025 Budget**	**2026 Budget**
Personnel		
Salary and Fringe Benefits Two months salary plus benefits for S. Bhandari, Associate Specialist, UC ANR	14,405	14,405
Fringe Benefits: Included above		
Personnel Subtotal	**14,405**	**14,405**
Supplies/Services/Other Batteries for dataloggers, tools and supplies for mounting	1000	1000
Supplies/Services/Other Subtotal	**1000**	**1000**
Travel		
Travel from UCCE Tulare County office to field trapping sites in Tulare County. Estimate of 720 miles @ $0.677/mile	487	487
Other Direct Costs (GAEL)		
Travel Subtotal	**487**	**487**
Contractual		
Subaward to S. Gyawaly	1,000	1,000
Subaward to B. Wheeler-Dykes	1,000	1,100
Contractual Subtotal	**2,000**	**2,000**
ANR Total Direct Cost	**17,892**	**17,892**
Exclusion amount (Subs)		
ANR TOTAL DIRECT COSTS w/o exclusions (figure out IDC)		
ANR INDIRECT COSTS @ 11% TDC:	**1,968**	**1,968**
ANR to PI Elizabeth Fichtner Grant TOTAL:	**19,860**	**19,860**
Grand Total (Primary Plus Subawards)	**19,860**	**19,860**

Budget Justification

We are requesting funds for two months salary and benefits for S. Bhandari, MS, who serves as both an entomologist and Python programmer for this project. We also ask for $487 to support travel to our local trapping sites and to meet with local collaborators (J. Stewart and B. Quezada).

Primary PI Signature Page: Fichtner, UC ANR

______________________________ 10/22/24

Originator **Date**

Karmjot Randhawa 10/30/24

Department Chair/County Director **Date**

Suz Burton 10/29/2024

Sr. Contracts and Grants Analyst

Contracts and Grants Officer **Date**

Gyawaly-UCCE Butte, Sutter, Glenn, Colusa, Tehama Counties		
Item	**2025 Budget**	**2026 Budget**
Personnel		
Personnel Subtotal		
Supplies/Services/Other Batteries for dataloggers, tools and supplies for mounting	500	500
Supplies/Services/Other Subtotal	**500**	**500**
Travel		
Travel from UCCE offices to field sites. Estimate of 740 miles @ $0.677/mile	500	500
Other Direct Costs (GAEL)		
Travel Subtotal	**500**	**500**
Contractual		
Contractual Subtotal		
ANR Total Direct Cost	**1,000**	**1,000**
Exclusion amount (Subs)		
ANR TOTAL DIRECT COSTS w/o exclusions (figure out IDC)		
ANR INDIRECT COSTS @ 11% TDC:	**110**	**110**
ANR to Gyawaly Grant TOTAL:	**1,100**	**1,100**

Subcontract Signature Page: Gyawaly, UC ANR

______________________________ __10/22/2024____________

Originator **Date**

Preet Ahluwalia

______________________________ ____10/29/2024_________

Department Chair/County Director **Date**

Wheeler-Dykes-UCCE Glenn, Butte, and Tehama Counties		
Item	**2025 Budget**	**2026 Budget**
Personnel		
Personnel Subtotal		
Supplies/Services/Other Batteries for dataloggers, tools and supplies for mounting	500	500
Supplies/Services/Other Subtotal	**500**	**500**
Travel		
Travel from UCCE offices to field sites. Estimate of 740 miles @ $0.677/mile	500	500
Other Direct Costs (GAEL)		
Travel Subtotal	**500**	**500**
Contractual		
Contractual Subtotal		
ANR Total Direct Cost	**1,000**	**1,000**
Exclusion amount (Subs)		
ANR TOTAL DIRECT COSTS w/o exclusions (figure out IDC)		
ANR INDIRECT COSTS @ 11% TDC:	**110**	**110**
ANR to Wheeler-Dykes Grant TOTAL:	**1,100**	**1,100**

Subcontract Signature Page: Wheeler-Dykes, UC ANR

Becky Wheeler-Dykes ________________ 10/29/24

Originator • **Date**

Preet Ahluwalia

________________ 10.29/2024 ___

Department Chair/County Director **Date**

Project Title: Development of screening tools to determine *Xylella fastidiosa* tolerance in olives

Principal Investigator(s) (PI): Franklin Lewis, Pat J Brown,

Plant Sciences, UC Davis
One Shields Ave, Davis CA 95616
(530) 752-5604

Cooperator(s):

UC Davis Contained Research Facility

Training, Biosafety Level 3 research facility

Cristian Silvestri, University of Tuscia

Olive micropropagation consultation and collaboration, may be able to help import certain previously introduced varieties they have in culture

Lindsey Burbank, USDA Parlier

Pathology consultation and will provide the cultures of the De Donno strain when we have the appropriate permitting and training completed

Abhaya Dandekar, Ramona Abbattista, Chuck Leslie, UC Davis

Consultation regarding project design, maintenance of materials, protocol development, and sampling

Objective(s) of Proposed Research:

-Establish in vitro cultures of putatively resistant and susceptible varieties of olives

-Complete training and permitting for work with *Xylella fastidiosa* subsp. pauca strain De Donno at the UC Davis Contained Research Facility

-Obtain and maintain cultures of *Xylella fastidiosa* subsp. pauca strain De Donno from the USDA Crop Diseases, Pests and Genetics Research station in Parlier

-Develop target tissues for future molecular or transgenic approaches

<u>Justification and Importance of Proposed Research</u>:

Xylella fastidiosa (Xf) subsp. *pauca* De Donno (ST53) has been associated with Olive Quick Decline Syndrome (OQDS) (Serio et al 2024) and is not yet known to exist in California. Symptomatic pauca strains have been detected in Europe (Morelli et al 2021) and South America (Coletta-Filho, 2016. Armange et al, 2021). Previous work in California (Burbank et al, 2024) has shown that mechanical inoculation of potted plants is possible and disease symptoms take 18 months post inoculation to appear and demonstrated Glassy-wing sharpshooter (GWSS) to be a suitable vector for the spread of the pathogen, and demonstrated major cultivars of the California olive industry are susceptible to the pathogen. The USDA in Parlier also found DNA extraction and re-isolation of pathogens from olive field material challenging, presumably due to the same characteristics which make olives particularly suitable to arid environments such as a thick waxy cuticle and large quantities of trichomes. The unique physiology of in vitro derived materials (Hazarika, 2006) would be expected to have fewer of these barriers for PCR detection of successfully colonized plant material, and fewer contaminants while extracting xylem sap.

Our collaborators in the Dandekar lab at UC Davis have built expertise in studying the metabolic mechanisms of infection as well as identifying host molecular markers for pathogen resistance and susceptibility for various woody crop diseases including *Xanthamonas arboricola* pv. *juglandis* (Sagawa et al 2022), *Phytophthora pini* (Zaini et al 2021), *Xylella fastidiosa* subsp. *fastidiosa* (Dourado et al 2023), as well as *Xylella fastidiosa* subsp. *pauca* (de Souza et al 2020). The proteomic studies done in this lab group by Ramona Abbattista (2023, preprint) on several olive cultivars showing different degree of susceptibility to Xf, may help build biomarkers for susceptibility and tolerance and would be aided by tissue culture systems to accelerate germplasm and cultivar screening. The ongoing work by the Dandekar lab with Pierce's Disease causing *Xf* will help expedite the development of *in vitro* inoculation and screening protocol development.

In vitro screening tools have been utilized for other pathogens such as *Erwinia amylovora* (Viseur et al 1986), *Phytophthora pini* (Zaini et al 2021), *Fusarium oxysporum* (Parris et al 2024) and especially challenging to evaluate field pathogens such as *Armillaria mellea* (Baumgartner et al 2013, Adelberg et al 2021), and on curative controls of phytoplasma (Tanno et al 2018). Based on the growing number of *in vitro* assays for rapid pathogen tolerance screening, we expect that protocols to screen olive germplasm and cultivars for tolerance to *Xf pauca* should be possible.

An added benefit to this work being performed is that we will be able to share the proficiencies and expertise developed. Optimizing protocols for maintaining cultivars important to California would be shared with Foundation Plant Services. This should facilitate the development of protocols in the future to create clean, virus-free, plant materials of currently inaccessible varieties of table olives such as Manzanilla Cacereña which are currently unable to be released from quarantine due to the presence of detectable viral particles.

We will be seeking co-funding from the Olive oil council when proposal submissions open, from the IAB, and will work with the USDA in Davis to open a Crop Germplasm Committee for Mediterranean tree fruits which would include olives

Proposed Procedures to Accomplish Objective(s):

Establish clean in vitro cultures of olives from disinfested field and/or greenhouse material.

Work with the UC Davis Contained Research Facility to observe and to acquire training and expertise in handling plant pathogens prior to being granted access to the facility.

Register with the proper Federal and State agencies and updated Biological Use Authorizations to work with the pathogen at the Contained Research Facility

Our collaborators in the lab of Cristian Silvestri, at the University of Tuscia Viterbo, IT, have developed protocols for creating repetitively embryogenic cultures of olive from various tissue types, a pathway that may be suitable for future non-transgenic molecular tools or transgenic approaches should they be desired by the industry to address *Xf* or other problems.

Timetable for Project:

January-December: Obtain greenhouse, field, and *in vitro* materials from private and public partners

January: Begin the permitting and training process

April-June: Introduce field material to culture

May-August: Collect immature fruit for generating repetitively embryogenic cultures

September: Coordinate delivery of De Donno strain of *Xf* to the contained research facility

Ongoing: Coordinate with the USDA ARS Germplasm Program regarding the foundation of a Crop Germplasm Committee that would include the olive collection, generating a potential future source of evaluation funding. Dissecting molecular mechanism of susceptibility and resistance to Xf in olive in non-infected and infected tissue from olive tissue culture by omics analyses.

Present Outlook and Estimated Success in Accomplishing Objective(s):

Because the De Donno strain is already located within a contained research facility within the state, permitting is expected to be straightforward..

The Silvestri lab at Tuscia University, the most experienced olive propagation lab in the world, has offered to collaborate with us and consult on the establishment and maintenance of the necessary cultivars. Establishing olives from field conditions will require initial removal of significant exogenous and endogenous contamination in order to culture and propagate the in vitro shoots needed to begin pathogen challenges.

Developing target tissues for future molecular work will be based on previously established work by our colleagues at Tuscia University. We would be mirroring some of their planned work, and they have offered to provide necessary plant materials should we need them.

Establishment of olive tissue culture in vitro will pave the way for developing fast in vitro screenings including not only biotic stress but also abiotic stress such as water stress. Additionally, molecular analyses of olive tissue culture will provide more consistent and reproducible results therefore enabling the discovery of more reliable markers for disease resistance as well as general stress resilience which could also target the improvement of drupe quality.

Budget Support Summary by Objective(s):

There is no cost from the Federal or State governments for the permitting processes.

The first year costs will include training at the Contained Research Facility will require approximately 30 hours of training as well as 10 supervised work periods which we expect to cost $1,500. The ongoing monthly fees to have working space at the CRF has been quoted approximately $225 per month with annual increases to those fees approximately 5% per year.

Collecting, establishing, and maintaining materials in vitro will be the largest portion of the costs with the labor costs supporting Associate Specialist Franklin Lewis, domestic travel expenses between locations, and consumables during micropropagation

We will also be seeking funding to support this work through the California Olive Oil Council, California Fruit Tree, Nut Tree, and Grapevine Improvement Advisory Board (IAB), and funds from the USDA when a Crop Germplasm Committee is established that supports olive evaluation research.

Total Budget Request:

	% of Time On Project	Request Year One	Request Year Two	Projected Year 3
Personnel	5%	$3852		
SRA/Tech				
Lab Assistant				
Other				
Employee Benefits		$1591		
Supplies and Expenses		$4000		
Items and Cost				
Tissue Culture Supplies		$2500		
Contained Research Training		$1500		
Equipment (itemize when cost >$1,000)				
Items/Cost/Justification*				
Travel		$1000		
Trips/Purpose/Costs				
Computer Time				
Overhead (where appropriate)				
Indirect Costs**				
Total		$10,443		

<u>Literature Cited</u>:

Adelberg, J., Naylor-Adelberg, J., Miller, S., Gasic, K., Schnabel, G., Bryson, P., ... & Reighard, G. (2021). In vitro co-culture system for Prunus spp. and Armillaria mellea in phenolic foam rooting matric. *In Vitro Cellular & Developmental Biology-Plant*, *57*, 387-397.

Armange E.M., Souza A.A., Coletta-Filho H.D.. "Articficial inoculation of Xylella fastidiosa subsp. pauca strains in olive plants; an overview of greenhouse experiments." 3rd European Conference on Xylella fastidiosa and XF-ACTORS final meeting, 29 April 2021, 00.00 - 30 April 2021. Online.

Baumgartner, K., Fujiyoshi, P., Browne, G. T., Leslie, C., & Kluepfel, D. A. (2013). Evaluating paradox walnut rootstocks for resistance to armillaria root disease. *HortScience*, *48*(1), 68-72.

Coletta-Filho, H. D., Francisco, C. S., Lopes, J. R. S., De Oliveira, A. F., & de Oliveira Da Silva, L. F. (2016). First report of olive leaf scorch in Brazil, associated with Xylella fastidiosa subsp. pauca. *Phytopathologia mediterranea*, *55*(1).

de Souza, J. B., Almeida-Souza, H. O., Zaini, P. A., Alves, M. N., de Souza, A. G., Pierry, P. M., ... & Nascimento, R. (2020). Xylella fastidiosa subsp. pauca strains Fb7 and 9a5c from citrus display differential behavior, secretome, and plant virulence. International Journal of Molecular Sciences, 21(18), 6769.

Dourado, M. N., Pierry, P. M., Feitosa-Junior, O. R., Uceda-Campos, G., Barbosa, D., Zaini, P. A., ... & Araújo, W. L. (2023). Transcriptome and Secretome Analyses of Endophyte Methylobacterium mesophilicum and Pathogen Xylella fastidiosa Interacting Show Nutrient Competition. Microorganisms, 11(11), 2755.

Hazarika, B. N. "Morpho-physiological disorders in in vitro culture of plants." Scientia horticulturae 108.2 (2006): 105-120.

Morelli, Massimiliano, et al. "Xylella fastidiosa in olive: A review of control attempts and current management." Microorganisms 9.8 (2021): 1771.

Parris, S. M., Jeffers, S. N., Olvey, J. M., Olvey, J. M., Adelberg, J. W., Wen, L., ... & Saski, C. A. (2022). An in vitro co-culture system for rapid differential response to Fusarium oxysporum f. sp. vasinfectum race 4 in three cotton cultivars. *Plant Disease*, *106*(3), 990-995.

Sagawa, C. H., Assis, R. D. A., Zaini, P. A., Saxe, H., Wilmarth, P. A., Salemi, M., ... & Dandekar, A. M. (2022). De novo arginine synthesis is required for full virulence of Xanthomonas arboricola pv. juglandis during walnut bacterial blight disease. Phytopathology®, 112(7), 1500-1512.

Serio, Francesca, et al. "A Decade after the Outbreak of Xylella fastidiosa subsp. pauca in Apulia (Southern Italy): Methodical Literature Analysis of Research Strategies." Plants 13.11 (2024): 1433.

Tanno, K., Maejima, K., Miyazaki, A., Koinuma, H., Iwabuchi, N., Kitazawa, Y., ... & Namba, S. (2018). Comprehensive screening of antimicrobials to control phytoplasma diseases using an in vitro plant–phytoplasma co-culture system. *Microbiology*, *164*(8), 1048-1058.

Viseur, J., & Figueroa, T. M. (1986, June). In vitro co-culture as a tool for the evaluation of fire blight resistance in pears and apples. In *IV International Workshop on Fire Blight 217* (pp. 273-282).

Zaini, P. A., Lee, S. H., Leslie, C. A., Walawage, S. L., Jiang, C. Z., Browne, G. T., ... & Kasuga, T. (2021). A rapid in vitro phenotypic assay of walnut shoots for prescreening resistance to Phytophthora pini. Plant Health Progress, 22(3), 235-239.

CALIFORNIA OLIVE COMMITTEE – OLIVE MAPPING

PREPARED FOR: Elise Oliver/California Olive Committee

PREPARED BY: Joel Kimmelshue/Land IQ
Joel Crowther/Land IQ
Casey Gudel/Land IQ

DATE: October 28, 2024

INTRODUCTION

This proposal and scope of work was developed at the request of the California Olive Committee (COC) for the purpose of developing an approach for assessing field-by-field olive acreage, age, and the differentiation between hedgerow and traditional olive plantings, a line of evidence for determining if the olives are harvested for table or olive oil.

LAND IQ BACKGROUND

Land IQ is a private technology, research, and science-based consulting firm that specializes in integrating agronomic sciences with spatial sciences to address large-scale land management and landscape analysis challenges. Specifically, we integrate agronomic land-based sciences, remote sensing, GIS, and advanced data management to better understand agricultural management and production systems at the field-scale. We also build specific, web-based interactive data management tools and solutions for the purposes of viewing results and dissemination of data. This expertise aligns well with the needs of the COC, as do some of the base data that we have already developed for the entire state of California, including current and historic olive mapping.

EXPERIENCE

CROP CLASSIFICATION

Land IQ has developed a remotely sensed California statewide land use mapping dataset for the California Department of Water Resources (DWR) covering the following crop years and systems:

- 2014 Main Season Summer Crop
- 2016 Main Season Summer Crop
- 2017/2018 Hydrologic Year (includes multi-cropping)
- 2018/2019 Hydrologic Year (includes multi-cropping)
- 2019/2020 Hydrologic Year (includes multi-cropping)
- 2020/2021 Hydrologic Year (includes multi-cropping)
- 2021/2022 Hydrologic Year (includes multi-cropping)
- 2022/2023 Hydrologic Year (includes multi-cropping)

This land use dataset is the standard by which the state of California provides comprehensive land use for multiple entities for the purpose of managing various land and water resources.

Spatial land use information, including agricultural and other land uses, are essential for severalregional and statewide regulatory, planning and resource management purposes, marketing efforts, crop forecasting, water management, and land use change. Accurate and timely land use information is the foundation of these analyses and is vital to objective and accurate decision-making and modeling processes.

Increased availability of digital satellite imagery, aerial photography, and new analytical tools make remotely sensed land use surveys possible at a field scale (e.g. as fine as 1.0 acre and less). These technologies allow accurate, large-scale crop and land use identification to be performed at time increments as desired and make possible comprehensive statewide crop-by-crop land use information available for the first time. The most recent effort classified over 10.7 million acres of agriculture into 58 crop categories that align with the DWR standard land use legend with a classification accuracy exceeding 97% statewide (Figure 1). Standard classification of fields is approximately 2.0 acre or greater, however there are instances where smaller fields that are in close proximity to larger fields have also been identified.

Agricultural land use is continually changing. It is important to understand the impacts of crop type and distribution, crop change, acreage, permanent crop age, irrigation method, and associated production practices. These data can be used to inform decisions on water resources management, marketing activities, ownership, location, greatly refine hydrologic models, evaluate groundwater recharge suitability, and better assess the role of agriculture in the management and sustainability of surface and groundwater resources.

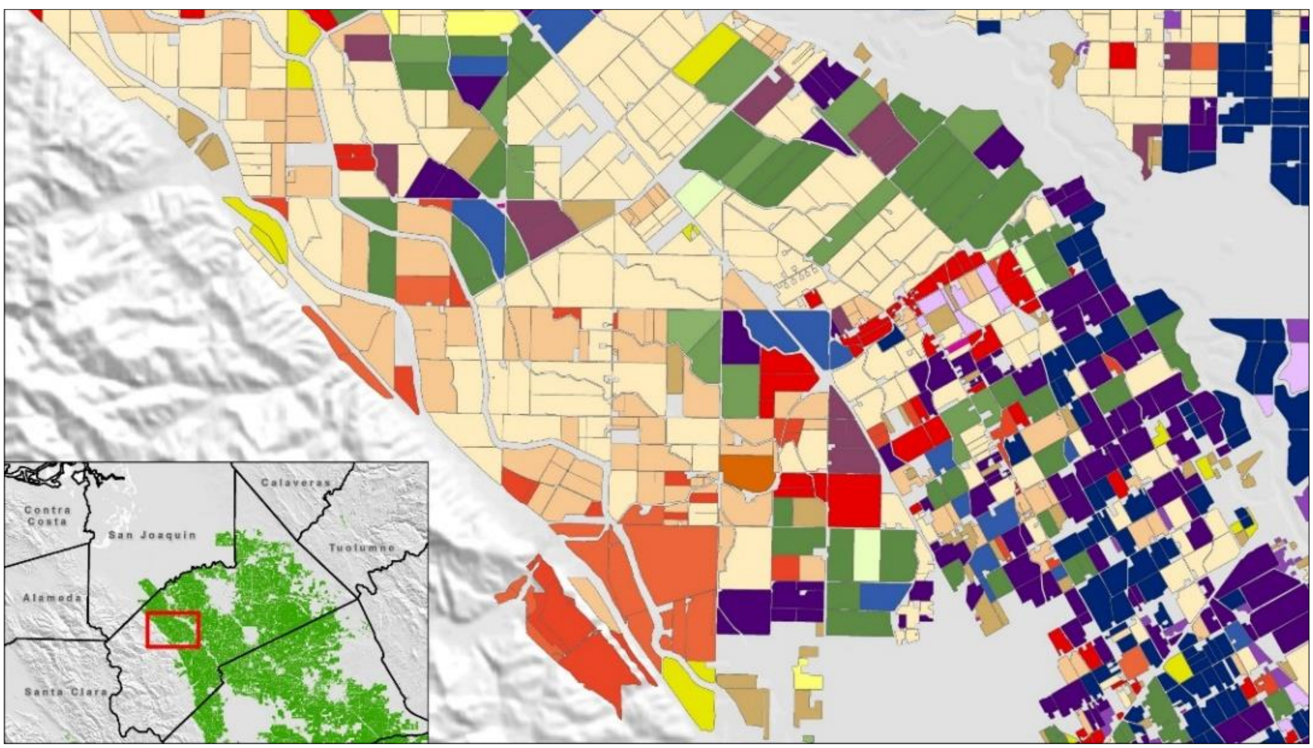

Figure 1. Field Scale Crop Mapping

AGE CLASSIFICATION

Once olives are mapped, further analysis can be conducted to determine the age of the crop utilizing a backwards looking time series analysis on available imagery as far back as 1984. Each crop is analyzed for spectral and textural signature changes from a crop to bare ground. At that point, this is considered the planting year (Figure 2). Land IQ has also completed age mapping of permanent crops in California to the actual planting year (+/- 1-2 years at >90% accuracy) for the statewide crop mapping effort for DWR in the 2018 - 2023 crop years.

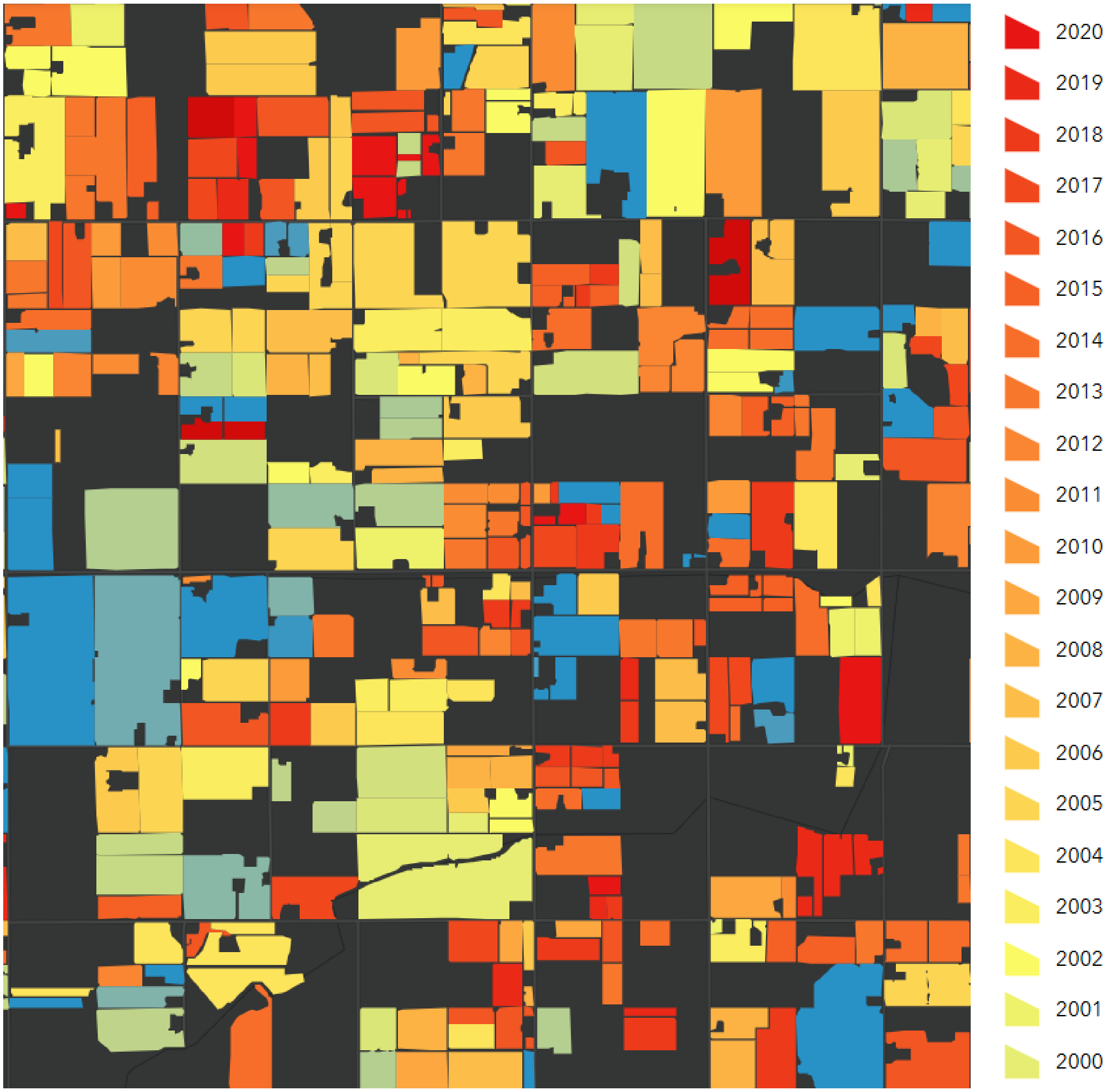

Figure 2. Example of Land IQ Age Analysis in Almonds

WEB MAP TOOLS

Growers and commodity groups need to understand the impacts of land use, crop location, crop change, acreage, tree age, proximity to other crops and urban areas, and best management practices on environmental attributes and impacts such as water quality, air quality, disease, and/or pest vectors. Conversely, environmental factors, such as climate change and sensitive habitats, increasingly influence how much and where these crops are grown. For these purposes, as well as many others, as these spatial mapping layers continue to be developed, it is important for the information to be accessible by designated end users to aid in effective decision-making and other applications. Land IQ develops web mapping applications for these purposes as shown in Figure 3.

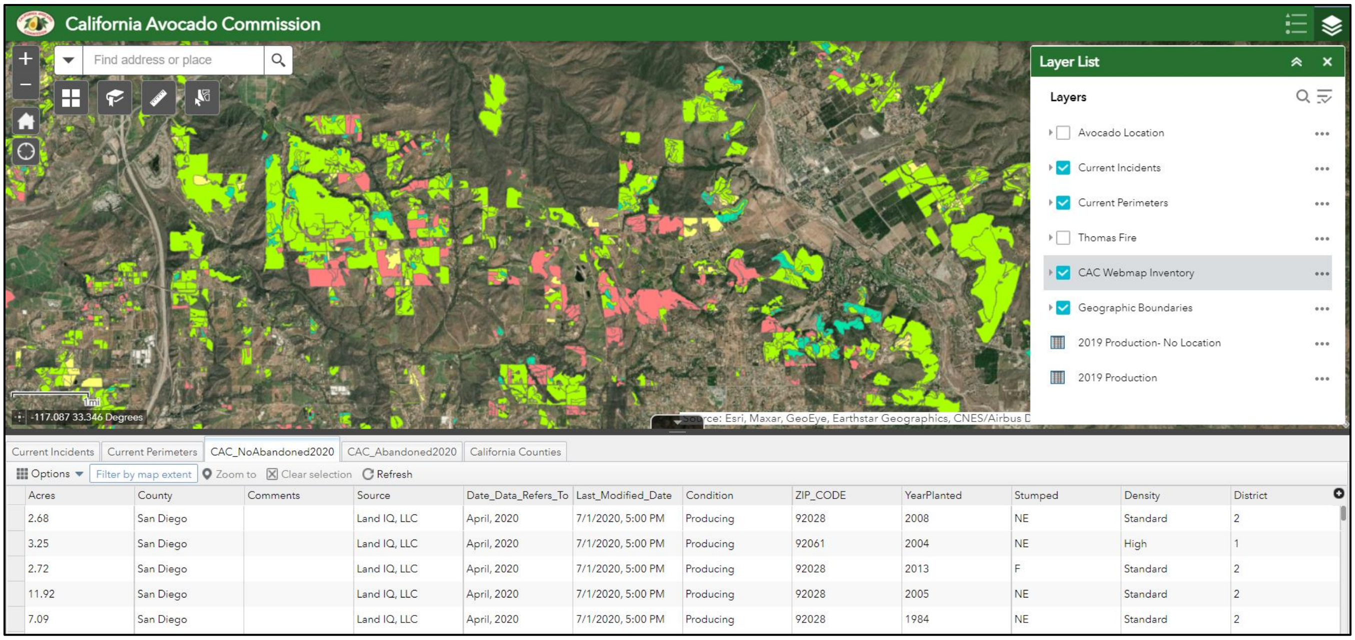

Figure 3. Web Mapping Application and Example Data from the California Avocado Commission

STAFFING RESOURCES

Staff expected to work on this project from Land IQ have been involved in various aspects of statewide crop mapping for the last 15 years, and in agricultural remote sensing, geospatial analysis and large-scale data management for the last 27 years. Other appropriately qualified staff may also participate to facilitate timely completion of any tasks approved by COC as a part of this proposed scope of work.

- Principal Agricultural Scientist – Joel Kimmelshue, PhD
- Project Manager – Casey Gudel, MS
- Principal Agricultural Scientist – Mica Heilmann, BS
- Remote Sensing Analyst – Diya Chowdhury, MS
- Project Agricultural Scientist – Joel Crowther, MS
- Geospatial Developer and Programmer – Tianyi Sun, MS
- GIS Analyst and Data Management Specialist – Justin Sitton, BS
- Support Staff – Various as needed

TASKS

Two tasks are included in this scope of work, as follows:

- Task 1 – Olive Mapping
- Task 2 – Web Mapping Application

TASK 1: OLIVE MAPPING

Scope of Work: The Land IQ mapping conducted for the DWR currently includes the crop category of Olives (Figure 4), which includes both table olives and olives for oil. Land IQ proposes utilizing this mapping, in addition to our team's agronomic knowledge, ground truth data, Department of Pesticide Regulation's Pesticide Use

Reports, image analysis, county crop reports, USDA-NASS Cropland Data Layer and remote sensing techniques to further refine the classification by indicating planting system (hedgerow or traditional), an indicator of those grown for olive oil or table olives. Understanding some plantings deviate from the standard of hedgerow-planted olives being used for oil production, Land IQ will consult with industry representatives to confirm those used for table olives. The dataset will be updated to reflect standing olive acreage at the end of the 2024 water year (September 30, 2024).

Once the mapping has been completed, further analysis can be conducted to determine the age of the crop using a backwards looking time series analysis on available imagery as far back as 1984. Each crop is analyzed for the spectral and temporal changes from a crop to bare ground. At this point, this is considered the planting year to within +/- 1 year.

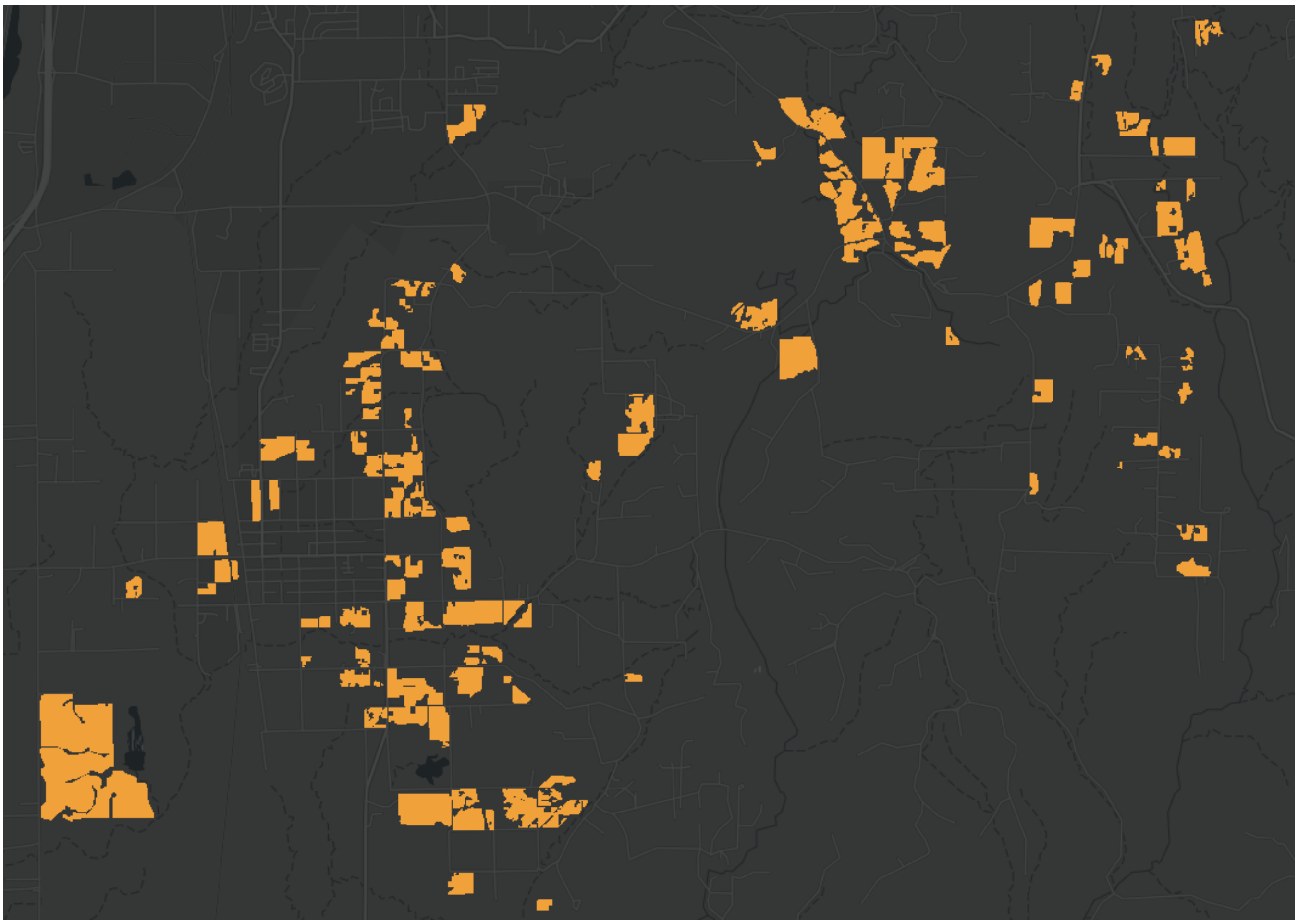

Figure 4. Land IQ Olive Mapping

Deliverables: The deliverables for this task will include a tabular dataset. This will include planted acreage by year planted and county as well as a summary of acreage planted by planting system. Results will also be loaded into the web mapping application described in Task 2.

Assumptions: The Land IQ accuracy for classifying the crop category of olives in the DWR dataset is greater than 98%. A small number of groves will not be correctly classified. These errors are rare but do exist. As a result, it is assumed the COC will notify Land IQ when errors exist so Land IQ can collaboratively update and improve the accuracy of the dataset for COC. It is also assumed that Land IQ will work collaboratively with the COC to identify the main olive oil and table olive growing regions to help focus the analysis.

Schedule: This task can be completed in 2-3 months following a notice to proceed.

TASK 2: WEB MAPPING APPLICATION

Scope of Work: This task enhances the grower focused web mapping application by including an administrative portal for a central, efficient means of communicating the results of spatial mapping and analysis efforts. Access to the administrative portal will be password protected for intended users only.

Authorized users will be able to view groves and their attributes including:

- Grove location and irrigated area boundary (Task 1)
- Grove age (Task 1)
- Grove planting system – hedgerow or traditional (Task 1)
- Grove use – table or olive oil (Task 1)

Other publicly available layers will be added including:

- County
- Water District boundaries
- Groundwater Sustainability Agency boundaries

Features included in this web mapping application include:

- Selection tool and data download
- Ability to select a grove and change the grove use between table and oil

Deliverables: The deliverable for this task is a flexible, informed, editable and updatable web mapping application.

Assumptions:

- The resultant web mapping application can be modified and further customized moving forward to include other data or advanced functions as desired to continue to serve the needs of COC. This additional work is not considered as part of this scope of work or cost estimate.
- The web mapping tool and associated data will be licensed for use by the COC only. If COC prefers and Land IQ agrees, additional licensed users can be established.
- Additional web tool capabilities analytical queries and spatial data can be added at additional cost.

Schedule: This task can be completed within 2 weeks of completing the mapping.

COST ESTIMATE & PAYMENT TERMS

The following table provides a detailed breakdown of cost by task.

Detailed Cost Estimate for Project

Task	Description	Cost
Task 1	Olive Mapping	$22,400
Task 2	Web Mapping Application	$8,300

Payment shall be made by COC to Land IQ at the following milestones:

1. 50% of the cost upon execution of the contract documents including a licensing agreement by both parties
2. Remaining 50% of costs will be invoiced upon acceptable completion of the mapping and web mapping application according to the terms of this scope of work and contract documents
3. Any additional efforts requested will be invoiced as agreed upon in a scope amendment

All payments shall be made within 30 days of receipt of invoices from Land IQ.

ANNUAL MAINTENANCE AND LICENSING

MAINTENANCE

Annual maintenance costs for maintaining and hosting the Web Map Information System is $4,200 for the second year. Maintenance is included in the cost for development during the first year. Maintenance costs may increase slightly over time, but will not increase by more than 3% annually. Annual maintenance will include:

- Web site hosting and data fees, etc. Maintenance fees will be due after the first year of web map operation and annually thereafter, as long as the client desires to maintain the web map.
- Web mapping software license fees and data hosting hardware costs.
- Three users with editing ability and unlimited users with viewing ability.
- Uploading of new base image maps, street layers, updated irrigation district boundaries, and updated regulatory (e.g., Irrigated Lands Regulatory Program, Sustainable Groundwater Management Act, etc.) boundaries, political boundaries, watershed boundaries, etc. that are agreed to as initial additional layers in the original web map itself.
- General data management and routine QA/QC and clean-up of the underlying base layer data.

LICENSING AND CONFIDENTIALITY

Mapping products will be licensed for use by COC only. A license agreement will be executed by both parties at initial contracting.

Land IQ will maintain complete confidentiality of all products and data created for, supplied to, or provided from COC.

Proposal: New prospects for the control of black scale in California olive groves

November 2024; Period of Performance: 2025-2029

Research Team

USDA-ARS European Biological Control Laboratory, Montpellier France: N. Manoukis (Coordinator); G. Desurmont, J. Kashefi and R.F.H. Sforza (investigators)

USDA-ARS San Joaquin Valley Agricultural Sciences Center: R. Krugner (investigator)

University of California: K. Daane and S. Triapitsyn (investigators)

Perrotis College, American Farm School, Thessaloniki, Greece: Ch. Vasilikiotis, A. Gertsis (investigators)

Background

From 1970-2000, black scale (*Saissetia oleae*) was the major insect pest of California olives. Prior to that it was controlled by insecticide treatments targeting the olive scale, *Parlatoria oleae*, which was the more serious pest at that time. When the biological control of olive scale became fully effective during the late 1960s, annual treatments for it were discontinued, and black scale emerged as the predominant insect pest. Black scale is thought to be native to sub-Saharan Africa. This "soft scale" (Family Coccidae) is currently found in most Mediterranean and semi-tropical regions of the world. Hosts include almond, apple, apricot, citrus, coyote brush, fig, fuchsia, grape, grapefruit, oleander, peppertree, plum, prune, and rose. With such a wide host range it can be found throughout California and will readily infest olive and citrus orchards where it can cause serious economic losses.

Black scale typically has one generation per year; however, because development is driven by temperature and host quality, there can be two generations when conditions permit. For example, in California's interior valleys, scale development is slower during winter and summer because temperatures approach the scales tolerable levels, resulting in one generation per year. In fact, prolonged periods of high temperatures will cause significant mortality of the early development stages. Therefore, cultural practices that lower canopy temperatures in summer (e.g., closed pruning, flood irrigation) reduce scale mortality and can alter scale development to result in a second or partial second generation. On the coast, where temperatures are milder, there is often two generations per year. The natural reduction of black scale, even when natural enemies are present, depends on a 70-90% kill – or desiccation – of the early development stages during the spring and summer. Therefore, canopy structures that affect the microclimate will influence black scale mortality and development. When not pruned for many years, the canopy becomes dense or "closed," and the spring and summer heat is moderated; in this protected environment the scale can better survive hot summers and develop outbreak populations in mild summers. Regular pruning opens the canopy and exposes the scale to higher temperatures and dryer conditions. This is the best cultural control available today.

Many natural enemies have been imported and released in California to control black scale starting in the late 19th century. About 15 parasitoid species have become established. The most common parasitic wasps include *Metaphycus helvolus*, *Metaphycus anneckei*, *Metaphycus hageni*, *Coccophagus ochraceus*, *Coccophagus lycimnia*, *Coccophagus scutellaris*, and *Scutellista caerulea*. Insect predators are also present, such as green lacewings (*Chrysoperla* spp.), and lady beetles (e.g., *Hippodamia convergens* and *Hyperaspis* sp.). These predators feed primarily on young black scale. Unfortunately, these parasitoids and predators have had limited success in controlling black scale below economic injury levels. One problem is that the parasitoids cannot reproduce without suitable black scale host stages. Because the scale often has only one generation per year, there are often periods when the proper host stage, or size, is not available and the parasitoid numbers drop to very low levels.

The recent resurgence of the black scale in California illustrates that natural control of this pest by its natural enemies is not optimal under current olive production practices. Combined with increased interest in non-chemical control and organic olive production, the situation requires new approaches to manage black scale in the state, especially those based around safe, effective and cost-efficient biological and cultural methods.

Objectives

1) **Test and implement cultural practices that maintain a substantial population of agents before the pest population grows**. Add and maintain additional plant species to provide sustenance during off-season for parasitoid species including *Metaphycus*.
2) **Develop an augmentorium strategy adapted to the black scale and its parasitoids**. The augmentorium will be designed to enhance the impact of populations of *Metaphycus* and other parasitoids in olive groves.
3) **Using sterilized ladybug nymphs by Neem bioinsecticide as massive release to control black scale**. By sterilizing ladybugs with a bio-insecticide like Neem, nymphs will not develop into adults and become potentially problematic (opposed to the Asian ladybug)
4) **Collection, taxonomy and molecular analysis of the scale and its parasites** from California and Europe for a better understanding of scope of the problem and its solutions

Approach and methods

<u>Objective 1:</u> Test and implement cultural practices that maintain a substantial population of agents before the pest population grows

In **Year 1**, ground covers with different sown plant species mixtures (containing brassicas, legumes, umbellifers and grasses) plus a control field, will be established in three olive groves in northern Greece. **For Years 2 - 4**, the effect of available food source to adult parasitoids in field will be monitored and how this improves the attraction of new scale parasitoid populations to the field, their establishment and survival in the orchard compared to traditional olive groves. This use of cover crops beside a possible positive effect on the biological control of Black Olive Scale, could have also a positive influence on the biological control of other olive pests and on soil health and water preservation (One health).

<u>Objective 2</u>: Develop an augmentorium strategy adapted to the black scale and its parasitoids

The augmentorium is an environmentally-friendly pest management tool that aims at reducing pest numbers while enhancing the impact of parasitoids naturally occurring in the field, without requiring the introduction and release of new natural enemies (Klungness et al., 2005; Jang et al., 2007). An augmentorium is a tent-like structure where infested plant parts can be deposited, in this case olive twigs/branches infested with black scales. The augmentorium is designed so that insect pests (adult scales and crawlers) are retained but parasitoids emerging from the scales can escape freely. This approach has been used with success to control tephritid flies in several tropical agroecosystems. The low cost and low maintenance associated with this strategy, coupled with its easiness to use and overall effectiveness have made it a popular management strategy among growers.

Due to the natural occurrence of several species of *Metaphycus* parasitoids attacking black scales in olive groves in California, the augmentorium strategy has high potential. To be successfully implemented, this strategy requires growers to prune out some scale-infested twigs from olive trees during spring, when scales are not yet releasing crawlers, and place them in the augmentorium. No further maintenance or intervention is required. The augmentorium serves as a refuge and breeding ground for parasitoids and lets them then disperse in the field.

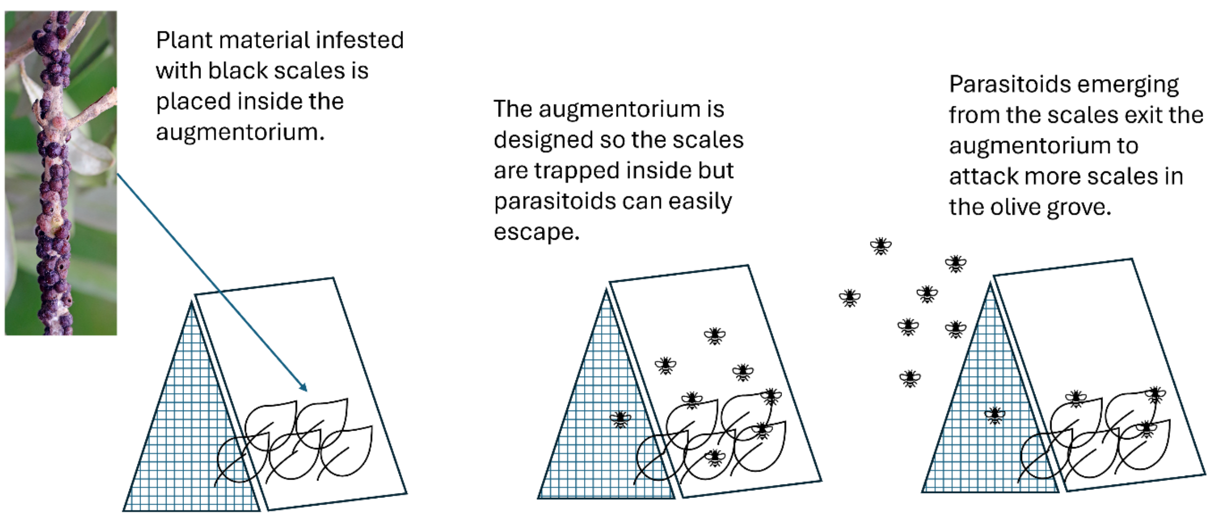

Figure 1. Principle of the augmentorium. The tent-like structure needs to be adapted to the black scale system, i.e. to have the right type of mesh and design to retain adult scales and crawlers but let parasitoids move freely.

In **Year 1**, Development of a prototype of augmentorium adapted to the black scale and its parasitoids. Tests will be performed under laboratory conditions at EBCL to determine the best mesh type and tent design to guarantee optimal release of parasitoids while retaining most of the collected scales. During **Years 2-3**: Tests under semi-field conditions to evaluate the efficacy of the augmentorium prototype designed during year 1. Large field cages with and without augmentoria will be used and the percentage parasitism of black scales will be measured over the course of several weeks or months to calculate the realized impact of the implementation of an augmentorium on the regulation of scale populations. Finally, **Year 4**: Transfer of the augmentorium technology to US-based collaborators for field tests in olive groves.

Objective 3: Using sterilized ladybug nymphs by Neem bioinsecticide as massive release to control black scale

On **Year 1**, a major task will be to set up an in-door black scale (BS) colony and select one or several common Californian ladybug species and rear it/them at the EBCL quarantine facility (indoor in quarantine or outdoor if present in France or commercially available in the EU); we then plan to test predation pressure in laboratory conditions. On Y**ear 2**, if in routine colonies were successfully obtained, optimize metamorphosis disruption of ladybug by neem treatments (different doses will be tested) and measure the impact of predation by sterilized nymphs on various black scale densities in Petri dishes. Azadirachtin has been identified as neem's principal active compound and comes from neem (A*zadirachta indica*), a tree that grows in tropical regions such as India. Azadirachtin affects insect physiology by mimicking a natural hormone. At the larval stage, azadirachtin can inhibit molting, preventing nymphs from developing into pupae. Azadirachtin is known to act against the insect juvenile hormone and 20 hydroxyecdysone. The tasks of **Year 3 & 4** will be to test in semi open field conditions (with caged infested olive trees of the same age) the sterilized ladybug nymph releases and follow up on predation compared to controls. If optimal results are obtained, then a technology transfer to CA will be the next step, by which experimental set-ups in Cal olive organic orchards could be proposed. The final task of this objective (**Year 5**) will be to deliver an ad hoc protocol to develop mass reared sterilized ladybugs to serve as predators in organic olive orchards and produce grey and scientific literature.

Objective 4: Collection, taxonomy, and molecular analysis of the scale and its parasitoids.

Collections of 'black scale' will be made throughout California on different host plants. Collected material will be stored in alcohol and sent to the California Department of Food and Agriculture (CDFA) for morphological identification. Concurrently, collections of black scale will be made in France, Greece and Italy and compared with Californian populations. About 6 organic olive orchards in California will be identified with assistance from UC Extension personnel from Tulare and Fresno Counties and selected for monitoring. Scale insects will be collected from each orchard and taken to the USDA facility in Parlier for rearing of any parasitoid species. Emerged parasitoids will be placed in alcohol and sent to Dr. Serguei Triapitsyn for identification. This procedure will be conducted for three years. In **Years 1 to 3**, analysis of population dynamics of black scales and parasitoids in California will be conducted to provide ecological insights to host-natural enemy interactions. In **Year 2**, populations of black scale will be collected in Europe and sent to the CDFA for morphological identification. Specimens collected in Europe will be compared with those collected from California. If necessary, in **Year 3**, molecular characterization methods will be developed and implemented for identification of black scale populations and associated parasitoids.

Timeline

Year	2025				2026				2027				2028				2029			
Quarter	**1**	**2**	**3**	**4**	**1**	**2**	**3**	**4**	**1**	**2**	**3**	**4**	**1**	**2**	**3**	**4**	**1**	**2**	**3**	**4**
Objective 1																				
Preparation of fields	X	X																		
Monitoring scale and parasitoid populations				X	X	X		X	X	X		X	X	X	X					
Analysis and peer reviewed paper														X	X	X				
Objective 2																				
Augmentorium prototype development		X	X	X	X															
Field tests, EU & US						X	X	X	X	X	X	X								
Demonstrations for growers, US														X	X					
Peer reviewed paper																	X	X	X	
Objective 3																				
Set up colonies black scale and ladybugs			X	X	X	X														
Optimize Neem doses					X	X	X	X												
Open field tests									X	X	X		X	X	X					
Peer reviewed paper																		X	X	
Objective 4																				
Monitoring of scale and parasitoid populations (CA)		X	X	X	X	X	X	X	X	X	X	X								
Collections in Europe						X	X													
Analysis scale/natural enemy collections							X	X			X	X	X	X						
Peer reviewed paper																	X	X		

Deliverables

1. Methods for improved survival and performance of natural enemies already present in California.
2. Augmentorium method tested and defined for black scale to enhance the action of natural enemies, particularly parasitoids.
3. A mass rearing protocol for labybugs and optimal doses of sterilization by Neem for ladybug nymphs.
4. Optimal release protocol for ladybugs with regards to black scale population densities.
5. Information on best matching species and strains of parasitoids that attack black scale in California.
6. Optimal release strategies for parasitoids with regards to black scale population dynamics.
7. Information on any species or strains of parasitoid that may be effective new agents against black scale.
8. Annual reports to COC on progress.
9. Peer-reviewed publications.

Budget

Table 1: Cost breakdown by objective, spending category, and year (in US Dollars).

Objective/Category	Year 1	Year 2	Year 3	Year 4	Year 5	Total
Objective 1	**$12,000**	**$18,250**	**$18,250**	**$16,150**	**$0**	**$64,650**
Supplies	$0	$2,000	$2,000	$2,920	$0	$6,920
Services	$12,000	$16,250	$16,250	$13,230	$0	$57,730
Travel for experiments	$0	$0	$0	$0	$0	$0
Scale collection trips	$0	$0	$0	$0	$0	$0
Objective 2	**$3,500**	**$29,500**	**$29,000**	**$5,000**	**$5,000**	**$72,000**
Supplies	$3,500	$9,500	$4,000	$0	$5,000	$22,000
Services	$0	$0	$0	$0	$0	$0
Travel	$0	$0	$5,000	$5,000	$0	$10,000
Personnel	$0	$20,000	$20,000	$0	$0	$40,000
Objective 3	**$22,000**	**$22,000**	**$9,000**	**$9,000**	**$5,000**	**$67,000**
Supplies	$1,500	$1,500	$2,500	$2,500	$0	$8,000
Services	$500	$500	$500	$500	$0	$2,000
Travel	$2,000	$2,000	$2,000	$2,000	$5,000	$13,000
Personnel	$18,000	$18,000	$4,000	$4,000	$0	$44,000
Objective 4	**$35,500**	**$35,500**	**$35,500**	**$0**	**$0**	**$106,500**
Supplies	$0	$0	$0	$0	$0	$0
Services	$6,500	$6,500	$6,500	$0	$0	$19,500
Travel	$4,000	$4,000	$4,000	$0	$0	$12,000
Personnel	$25,000	$25,000	$25,000	$0	$0	$75,000
Annual Total	**$73,000**	**$105,250**	**$91,750**	**$30,150**	**$10,000**	**$310,150**

Resources

The European Biological Control Laboratory (EBCL) was established in 1991 near Montpellier. This new laboratory facility resulted from a fusion of the former European Parasite Laboratory, established in Paris in 1919, and a Biological Control of Weeds Laboratory, established in Rome in 1958. The new EBCL facility, inaugurated in October 1999, is the leading overseas laboratory of ARS (Agricultural Research Service) of USDA (US Department of Agriculture) and the first of its research facilities constructed and owned outside the USA. EBCL also maintains a research unit in Thessaloniki, Greece. The overall goal of research at EBCL is to develop biological control technologies that can be used to suppress invading weeds and insect pests. Several olive pests were recently studied at EBCL, e.g. olive fruit fly (*Bactrocera oleae*), olive psyllid (*Euphyllira olivina*), as well as pests in the scale family, like the vine mealybug (*Planococcus ficus*). For these projects, host specific parasitoids were collected in Europe and Africa and brought into the EBCL quarantine where colonies were maintained. Multiple shipments were scheduled, and parasitoids were released in Californian olive groves and vineyards. Collaborations with California stakeholders (CDFA) and academic entities (UC Berkeley) were maintained over the years.

The Crop Diseases, Pests, and Genetics (CDPG) Research Unit is housed within the USDA-ARS San Joaquin Valley Agricultural Sciences Center (SJVASC) in Parlier, California. CDPG occupies approximately 150 m^2 of office, 600 m^2 of laboratory, and 450 m^2 of greenhouse space, and has available a 260-m^2 containment facility for regulated arthropods and pathogens. CDPGRU laboratories are fully equipped for entomology.

At the University of California at Riverside, the Entomology Research Museum has the largest collection of the voucher specimens of black scale parasitoids in the world. These include materials from past surveys in California (e.g., Lampson & Morse 1992) during the past 100 years or so, and specimens imported from Africa and released in California as classical biocontrol agents. Also present are specimens reared in various states and private insectaries. These specimens will serve as an important resource for taxonomic comparison and identification of the newly collected species. At UC Berkeley, researchers have investigated black scale and its parasitoids for many decades (Daane and Caltagirone 1989, 1990 and 1999; Daane et al. 1991 and 2000; Barzman and Daane 2001).

Perrotis College, a division of the American Farm School (AFS), established in 1996, is a distinctive non-profit educational institution in northern Greece with focus on agriculture, food science, the environment and entrepreneurship. Perrotis College participates in many EU funded research projects and its faculty and students carry out various scientific experiments in the green houses, well equipped laboratories and experimental fields available at AFS. It also has close collaboration with many farmers throughout Greece. Experienced personnel can maintain and take care of any needs the collaborator could have during the research phase.

References

Barzman, M. S., and Daane, K. M. 2001. Host-handling behaviours in parasitoids of the black scale: a case for ant-mediated evolution. Journal of Animal Ecology 70(2): 237-247.

Daane, K. M., and Caltagirone, L. E. 1989. Biological control in olive orchards: cultural practices affect control of black scale. California Agriculture 43(1): 9-11.

Daane, K. M., and Caltagirone, L. E. 1990. Black scale distribution in the olive canopy: development of a sampling plan. Acta Horticulturae 286: 347-350.

Daane, K. M., Barzman, M. S., Kennett, C. E., and Caltagirone, L. E. 1991. Establishment of Prococcophagus probus Annecke and Mynhardt and Coccophagus rusti Compere (Hymenoptera: Aphelinidae): Parasitoids of black scale in California. Pan-Pacific Entomologist 67: 99-106.

Daane, K. M., and Caltagirone, L. E. 1999. A new species of Metaphycus (Hymenoptera: Encyrtidae) parasitic on black scale, Saissetia oleae (Olivier) (Homoptera: Coccidae). Pan-Pacific Entomologist 75(1): 13-17.

Daane, K. M., Barzman, M. S., Caltagirone, L. E., and Hagen, K. S. 2000. Metaphycus anneckei and M. hageni: two discrete species parasitic on black scale, Saissetia oleae. Biocontrol 45(3): 269-284.

Desurmont, G. A., Tannières, M., Roche, M., Blanchet, A., & Manoukis, N. C. (2022). Identifying an optimal screen mesh to enable augmentorium-based enhanced biological control of the olive fruit fly Bactrocera oleae (Diptera: Tephritidae) and the Mediterranean fruit fly Ceratitis capitata (diptera: tephritidae). *Journal of Insect Science*, *22*(3), 11.

Dhra, G., Ahmad, M., Kumar, J., and Patanjali, P. K. (2018). Mode of action of azadirachtin: a natural insecticide. Int. Res. J. 7, 41–46.

Kilani-Morakchi, Samira et al. (2021) Azadirachtin-Based Insecticide: Overview, Risk Assessments, and Future Directions. Frontiers in Agronomy.

Jang, E. B., L. M. Klungness, and G. T. Mcquate. 2007. Extension of the use of Augmentoria for Sanitation in a cropping system susceptible to the alien terphritid fruit flies (Diptera: terphritidae) in Hawaii. Journal of Applied Sciences and Environmental Management 11.

Klungness, L., E. B. Jang, R. F. Mau, R. I. Vargas, J. S. Sugano, and E. Fujitani. 2005. New sanitation techniques for controlling tephritid fruit flies (Diptera: Tephritidae) in Hawaii. Journal of Applied Sciences and Environmental Management 9: 5-14.

Lampson LJ, Morse JG. 1992. A survey of black scale, *Saissetia oleae* (Hom.: Coccidae) parasitoids (Hym.: Chalcidoidea) in southern California. Entomophaga 37: 373-390.

GAYLORD A. DESURMONT, Ph. D.

Date of birth: 25/02/1981
Nationality: French
European Biological Control Laboratory (EBCL), USDA-ARS
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Phone: +33 4 99 62 30 11; cell: +33 6 09 38 01 31
E-mail: g.desurmont@gmail.com ; gdesurmont@ars-ebcl.org
Languages: French (native speaker), English (fluent).

CURRENT POSITION

Research Entomologist / Quarantine officer, European Biological Control Laboratory (EBCL), USDA-ARS, Montferrier-sur-lez (France)

EDUCATION

June 1998: Sciences Baccalaureate (Major: Biology) (Roubaix, France)
1998-2000: 2 year advanced math and science program (Lycée Albert Châtelet, Douai, France). Intensive preparatory course for M.S. entrance exams.
2000-2003: Master of Sciences (M.S.) in Agronomy and agricultural practices, option ecology and environment, Agricultural school of Dijon (ENESAD: Etablissement National d'Enseignement Supérieur Agronomique de Dijon).
2003-2005: M.S. in Entomology, Cornell University, Ithaca, NY. Thesis title: "Evaluation of *Podisus maculiventris* (Say) as a biological control agent against viburnum leaf beetle [*Pyrrhalta viburni* (Paykull)]". Main Advisor: John Sanderson.
2006-2009: Ph.D. in Entomology (awarded August 17th 2009), Cornell University, Ithaca, NY. Thesis title: "Oviposition of viburnum leaf beetle [*Pyrrhalta viburni* (Paykull)]: from ecology to biological control of an invasive pest." Main advisor: Ann Hajek.

EMPLOYMENT

Fall 2003-Fall 2005: Graduate Research / Teaching assistant, Entomology Department, Cornell University (NY, USA). Supervisor: John Sanderson
Spring 2006-Spring 2009: Graduate Research / Teaching assistant, Entomology Department, Cornell University (NY, USA). Supervisor: Ann Hajek.
Summer 2009: Graduate Research assistant, Ecology and Evolutionary Department, Cornell University (NY, USA). Supervisor: Anurag Agrawal.
Fall 2009-2010: Post doctoral Researcher, Ecology and Evolutionary Department, Cornell University (NY, USA). P.I: Anurag Agrawal

Spring 2011-Spring 2015: Postdoctoral Researcher (80%), Fundamental and Applied Research in Chemical Ecology (FARCE) laboratory, Institute of Biology, Neuchâtel University (Switzerland). P.I: Prof. Ted Turlings.
Spring 2014-Spring 2015: Postdoctoral Researcher (20%), Institute of Systematic Botany, University of Zürich (Switzerland). P.I: Prof. Florian Schiestl.
Spring 2015-Winter 2015: Postdoctoral Researcher (80%), Evolutionary Entomology laboratory (E-Vol), Institute of Biology, University of Neuchâtel (Switzerland). P.I: Dr. Betty Benrey.
Spring 2016-Present: Research Entomologist / quarantine officer, European Biological Control Laboratory (EBCL), USDA-ARS, Montferrier-sur-lez (France)

HONORS & AWARDS

Gyrisco award for outstanding graduate research in applied entomology (annual award, Cornell University) 2008.
First prize student competition (poster presentation), Entomological Society of America Eastern branch meeting 2009.
John Henry Comstock award for outstanding Ph.D. in Entomology (annual award, Entomological Society of America) 2010.

PUBLICATIONS

51. Kosciolek C., G.A. Desurmont, T. Thomann, A. Zamprogna, and V. Caron. "Toward a push–pull strategy against invasive snails using chemical and visual stimuli." Scientific Reports 14, no. 1 (2024): 11511.

50. **Desurmont, G.A.,** and A. Blanchet 2023. Florivory is an alternative but suboptimal diet for an invasive leaf-feeding beetle. **Ecological Entomology**, 48 (4), 508-511.

49. Grof-Tisza, P., S. Morelon, **G.A. Desurmont**, and B. Benrey 2022. Population-Specific Plant-To-Plant Signaling in Wild Lima Bean. **Plants**, 11(18), 2320.

48. Cuny, M.A.C, D. la Forgia, **G.A. Desurmont**, C. Bustos-Segura, G. Glauser, and B. Benrey 2022. Top-down cascading effects of seed-feeding beetles and their parasitoids on plants and leaf herbivores. **Functional ecology**, *in press*.

47. **Desurmont, G.A**., M. Tannières, M. Roche, A. Blanchet, and N.C. Manoukis 2022. Identifying an optimal screen mesh to enable augmentorium-based enhanced biological control of the olive fruit fly *Bactrocera oleae* and the medfly *Ceratitis capitata.* **Journal of Insect Science** 22.3: 11. https://doi.org/10.1093/jisesa/ieac027

46. Hanache, P., T. Thomann, V. Caron, **G.A. Desurmont** 2021. Can aestivation preferences be used to develop novel management tools against invasive Mediterranean snails? **Insects**, 12.12: 1118.

45. Mann, L., D. Laplanche, T.C.J. Turlings, **G.A. Desurmont** 2021. A comparative study of plant volatiles induced by insect and gastropod herbivory. **Scientific reports**, 11(1), 1-10.

44. Wang X., N. Ramualde, **G.A. Desurmont**, L. Smith, D.E. Gundersen-Rindal, M.J. Grodowitz 2021. Reproductive traits of the egg parasitoid *Aprostocetus fukutai*, a promising biological control agent for invasive citrus longhorned beetle *Anoplophora chinensis*. **BioControl**, *in press*. https://doi.org/10.1007/s10526-021-10118-2

43. Lin T., K. Vrieling, D. Laplanche, P.G.L Klinkhamer, Y. Lou, L. Bekooy, T. Degen, C. Bustos-Segura, T.C.J. Turlings, and **G.A. Desurmont** 2021. Evolutionary changes in an invasive plant support the defensive role of plant volatiles. **Current Biology**, *in press*. https://doi.org/10.1016/j.cub.2021.05.055

42. **Desurmont G.A.**, E. Kerdellant, and N. Lambin 2021. Between a rock and an egg-crushing place: selection pressure from natural enemies and plant defenses on eggs of the viburnum leaf beetle in its native range. **Ecological Entomology**, 46(2) 482-486. https://doi.org/10.1111/een.12936

41. **Desurmont, G.A**., M. van Arx, T.C.J. Turlings, and F.P. Schiestl 2020. Floral odors can interfere with the foraging behavior of parasitoids searching for hosts. **Frontiers in Ecology and Evolution**, 8, 148. https://doi.org/10.3389/fevo.2020.00148

40. Escobar, Y., F. Guermache, M. Bon, E. Kerdellant, L. Petoux, and **G.A. Desurmont** 2020. Biology, ecology, and impact of *Cryptonevra nigritarsis* Duda, a potential biological control agent against the giant reed *Arundo donax*. **Biological control**, 147, 104287. https://doi.org/10.1016/j.biocontrol.2020.104287

39. Trunz, V., M. Luchetti, D. Bénon, A. Dorchin, **G.A. Desurmont**, C. Kast, G. Glauser, and C.J. Praz 2020. To bee or not to bee – the “raison d’être” of toxic secondary compounds in the pollen of Boraginaceae. **Functional Ecology**, 34 (7), 1345-1357. https://doi.org/10.1111/1365-2435.13581

38. **Desurmont, G.A.**, S.P.G. Morelon, and B. Benrey 2020. First insights into the chemical ecology of an invasive pest: Olfactory preferences of the viburnum leaf beetle *Pyrrhalta viburni* (Coleoptera: Chrysomelidae). **Environmental Entomology**, 49 (2), 364-369. https://doi.org/10.1093/ee/nvaa007

37. **Desurmont, G.A**., M. Bon, E. Kerdellant, F. Guermache, T. Pfingstl, and M. Tixier 2020. An integrative approach combining molecular analyses and manipulative laboratory experiments to evaluate predation of insect eggs by a mite. **Ecosphere**, 1 (3), e03065. https://doi.org/10.1002/ecs2.3065

36. Gols, R., **G.A. Desurmont**, and J.A. Harvey 2019. Variation in performance and resistance to parasitism of Plutella xylostella populations. **Insects**, 10.9, 293.

35. Cuny, M.A.C., D. La Forgia, **G.A. Desurmont**, G. Glauser, and B. Benrey 2019. Role of cyanogenic glycosides in the seeds of wild Lima bean, Phaseolus lunatus: Defense, nutrition or both? **Planta**, 250 (4), 1281–1292.

34. **Desurmont, G.A**., E. Kerdellant, T. Pfingstl, and M. Tixier 2019. Mites associated with egg masses of the viburnum leaf beetle *Pyrrhalta viburni* (Paykull). **Acarologia** 59 (1), 57-72.

33. Jaffuel, G, V. Půža, A. Hug, R. G. Meuli, J. Nermuť, T. C. J. Turlings, **G. A. Desurmont**, and R. Campos-Herrerae 2019. Molecular detection and quantification of slug parasitic nematodes from the soil and their hosts. **Journal of invertebrate pathology**, 160, 18-25.

32. Kellenberger, R. T., **G. A. Desurmont**, P. M. Schlüter, and F. P. Schiestl 2018. Trans-generational inheritance of herbivory-induced phenotypic changes in Brassica rapa. **Scientific reports**, 8 (1), 3536.

31. **Desurmont, G. A**., A. Guiguet, and T. C. J. Turlings 2018. Invasive insect herbivores as disrupters of chemically-mediated tritrophic interactions: effects of herbivore density and parasitoid learning. **Biological Invasions**, 20 (1), 195-206.

30. **Desurmont, G. A**., and F. P. Schiestl 2017. The distracting power of flowers: floral volatiles can interfere with the host-searching behavior of parasitoids. **Proceedings of the 11th CIRAA**, Montpellier, France.

29. Carrasco, D.*, **G. A. Desurmont***, D. Laplanche, M. Proffit, R. Gols, M. Larsson, H. Luka, T. C. J. Turlings, and P. Anderson 2017. The multitrophic consequences of concurrent insect invasions: a range-expanding herbivore and its associated parasitoid affect native tritrophic interactions. **Global Change Biology**, 24(2), 631-643. **shared first authorship*

28. **Desurmont, G. A**., A. Köhler, D. Laplanche, D. Maag, H. Xu, J. Baumann, C. Demairé, D. Devenoges, M. Glavan, L. Mann, and T. C. J. Turlings 2017. The spitting image of plant defenses: effects of plant secondary chemistry on the efficiency of caterpillar regurgitant as an anti-predator defense. **Ecology and Evolution**, 7 (16), 6304-6313.

27. Danner Holger, **G.A. Desurmont**, S.M. Cristescu, N.M. van Dam 2017. Herbivore-induced plant volatiles accurately predict history of co-existence, diet breadth, and feeding mode of herbivores. **New Phytologist**, 220(3), 726-738.

26. Xu, H., **G. Desurmont**, T. Degen, G. Zhou, D. Laplanche, L. Henryk, and T.C.J Turlings 2017. Combined use of herbivore-induced plant volatiles and sex pheromones for mate location in braconid parasitoids: HIPVs strongly attract virgin parasitoids. Plant, cell, and the environment. **Plant, Cell & Environment**, 40 (3), 330-339.

25. **Desurmont, G. A**., H. Xu, and T.C.J. Turlings 2016. Powdery mildew suppresses herbivore-induced plant volatiles and interferes with parasitoid attraction in *Brassica rapa.* **Plant, Cell & Environment**, 39 (9), 1920-1927.

24. **Desurmont, G.A**., M.A. Zemanova, and T.C.J. Turlings 2016. The Gastropoda menace*: Arion* slugs on *Brassica* plants affect caterpillar survival through predation and interference with parasitoid attraction. **Journal of Chemical Ecology**, 42(3), 183-192.

23. Dominik Klauser, **G. A. Desurmont**, G. Glauser, A. Vallat, P. Flury, T. Boller, T.C.J. Turlings, and S. Bartels 2015. The Arabidopsis Pep-PEPR system is induced by herbivore feeding and contributes to JA-mediated plant defence against herbivory. **Journal of experimental botany,** 66 (17): 5327-5336.

22. **Desurmont, G. A**., D. Laplanche, F. P. Schiestl, and T. C. J. Turlings 2015. Floral volatiles interfere with plant attraction of parasitoids: ontogeny-dependent infochemical dynamics in Brassica rapa. **BMC ecology** 15 (1).

21. Chabaane, Y., D. Laplanche, T. C. J. Turlings, and **G. A. Desurmont** 2015. Impact of exotic insect herbivores on native tritrophic interactions: a case study of the African cotton leafworm, *Spodoptera littoralis*. **Journal of Ecology**, 103 (1), 109-117.

20**. Desurmont, G.A.**, and P.A. Weston 2014. Switched after birth: Performance of the viburnum leaf beetle [*Pyrrhalta viburni* (Paykull)] after transfer to a suboptimal host plant. **Insects**, 5 (4): 805-817.

19. **Desurmont, G. A,** A. E. Hajek, and A. A. Agrawal 2014. Seasonal decline in plant defense is associated with relaxed offensive oviposition behavior in the viburnum leaf beetle *Pyrrhalta viburni* . **Ecological Entomology**, 39 (5), 589-594.

18. **Desurmont, G. A**., and I. S. Pearse 2014. Alien plants versus alien herbivores: does it matter who is non-native in a novel trophic interaction? **Current Opinion in Insect Science**, 2: 20-25.

17. **Desurmont, G. A.,** J. Harvey, N. M. van Dam, S. Cristescu, F. P. Schiestl, S. Cozzolino, P. Anderson, M. C. Larsson, P. Kindlmann, H. Danner, and T. C. J. Turlings 2014. Alien interference: Disruption of infochemical networks by invasive insect herbivores. **Plant, Cell & the Environment**, 37(8): 1854-1865 .

16. Florian P. Schiestl, H. Kirk, L. Bigler, S. Cozzolino, **G. A. Desurmont** 2014**.** Herbivory and floral signalling: phenotypic plasticity and trade-offs between reproduction and indirect defence. **New Phytologist**, 203 (1): 257-266.

15. **Desurmont, G.A.**, and A. A. Agrawal 2014. Do plant defenses predict damage by an exotic herbivore? A comparative study of the viburnum leaf beetle. **Ecological applications**, 24 (4): 759-769.

14. **Desurmont, G.A.**, P.A. Weston, and A.A. Agrawal 2013. Reduction of oviposition time cost and larval group feeding: two potential benefits of aggregative oviposition for the viburnum leaf beetle. **Ecological Entomology**, 39 (1): 125-132.

13. **Desurmont, G.A.**, F. Hérard, and A. A. Agrawal 2012. Oviposition strategy as a means of local adaptation to plant defense in native and invasive populations of the viburnum leaf beetle. **Proceedings of the Royal Society B**, 279 (1730): 952-958.

12. **Desurmont, G.A**., M. J. Donoghue, W. L. Clement, and A. A. Agrawal 2011. Evolutionary history predicts plant defense against an invasive pest. **PNAS**, 108 (17): 7070-7074.

11. **Desurmont, G.A.**, and P.A. Weston 2011. Aggregative oviposition of a phytophagous beetle overcomes plant defenses. **Ecological Entomology,** 36 (3): 335-343.

10. **Desurmont, G.A.**, and P.A. Weston 2010. Stimuli associated with viburnum leaf beetle aggregative oviposition behavior, **Entomologia Experimentalis et Applicata**, 135 (3): 245-251.

9. **Desurmont, G. A.**, C. M. Fritzen, and P. A. Weston 2009. Oviposition by *Pyrrhalta viburni* (Paykull) on dead plant material: Successful reproductive strategy or maladaptive behavior? *In*: P. Jolivet, J. Santiago-Blay, and M. Schmitt (eds), **Research on Chrysomelidae vol. 2.** Brill academic publishers, Leiden, The Netherlands.

8. **Desurmont, G**.**A.** 2009. Oviposition of viburnum leaf beetle [*Pyrrhalta viburni* (Paykull)]: from ecology to biological control of an emerging landscape pest. **Ph. D. Thesis**, Cornell University.

7. **Desurmont, G. A**., and P. A. Weston. 2008. Predation by *Podisus maculiventris* (Say) (Hemiptera: Pentatomidae) on viburnum leaf beetle, *Pyrrhalta viburni* (Paykull) (Coleoptera: Chrysomelidae) under laboratory and field conditions. **Environmental Entomology** 37(5): 192-202.

6. Weston, P. A., and **G. A. Desurmont**. 2008. Pupation by viburnum leaf beetle (Coleoptera: Chrysomelidae): behavioral description and impact of environmental variables and entomopathogenic nematodes. **Environmental Entomology** 37 (4): 845-849.

5. Weston, P. A., **G. A. Desurmont**, and M. D. Diaz. 2008. Ovipositional ecology of viburnum leaf beetle *Pyrrhalta viburni* (Coleoptera: Chrysomelidae). **Environmental Entomology** 37 (2): 520-524.

4. **Desurmont, G. A.**, and P. A. Weston. 2008. Biological and environmental factors influencing predation of *Podisus maculiventris* (Say) on viburnum leaf beetle [*Pyrrhalta viburni* (Paykull)]. **Canadian Entomologist** 140: 1241-1251.

3. Weston, P. A., **G. Desurmont**, and E. R. Hoebeke. 2007. Viburnum leaf beetle: Biology, invasion history in North America, and management options. **American Entomologist** 53: 96-101.

2. **Desurmont, G.** 2006. Evaluation of *Podisus maculiventris* (Say) as a biological control agent against viburnum leaf beetle [*Pyrrhalta viburni* (Paykull)]. **M.S. Thesis**, Cornell University.

1. Weston, P. A., and **G. Desurmont**. 2002. Suitability of various species of Viburnum as hosts for *Pyrrhalta viburni*, an introduced leaf beetle. **Journal of Environmental Horticulture** 20(4): 224-227.

PRESENTATIONS & SEMINARS (First author and presenter)

03/20/05. ESA Eastern branch meeting, student competition (oral presentation). Natural occurrence and predatory efficiency of several predators against viburnum leaf beetle. Location: Harrisburg, Pennsylvania.

12/13/05. M.S. defense. Evaluation of *Podisus maculiventris* (Say) as a biological control agent against viburnum leaf beetle [*Pyrrhalta viburni* (Paykull)]. Location: Ithaca, New York.

12/16/05. ESA National meeting, student competition (oral presentation): Potential of *Podisus maculiventris* (Hemiptera: Pentatomidae) for biological control of viburnum leaf beetle, *Pyrrhalta viburni* (Coleoptera: Chrysomelidae). Location: Fort Lauderdale, Florida.

03/12/06. ESA Eastern branch meeting, student competition (oral presentation): Factors influencing predation of *Podisus maculiventris* (Hemiptera: Pentatomidae) on viburnum leaf beetle, *Pyrrhalta viburni* (Coleoptera: Chrysomelidae), a major landscape pest in Eastern US. Location: Charlottesville, Virginia.

12/11/06. ESA national meeting, student competition (oral presentation): Switched after birth: Performance of viburnum leaf beetle (*Pyrrhalta viburni* [Paykull]) larvae after transfer to suboptimal hosts. Location: Indianapolis, Indiana.

18/03/07. ESA Eastern branch meeting. student competition (oral presentation): Evidence of an aggregative oviposition behavior for viburnum leaf beetle [*Pyrrhalta viburni* (Paykull)] and costs and benefits associated. Location: Harrisburg, Pennsylvania.

12/10/07. ESA National meeting. student competition (oral presentation): Benefits of aggregative oviposition behavior for viburnum leaf beetle [*Pyrrhalta viburni* (Paykull)]. Location: San Diego, California.

03/09/08. ESA Eastern branch meeting, student competition (oral presentation). Stimuli associated with aggregative oviposition of viburnum leaf beetle [*Pyrrhalta viburni* (Paykull)]. Location: Syracuse, New York.

07/11/08. International Congress of Entomology (ICE). Invited speaker at the symposium "Advances in implementing biological control for perennial ornamental crops grown outdoors". Potential of *Podisus maculiventris* (Hemiptera: Pentatomidae) for biological control of viburnum leaf beetle, *Pyrrhalta viburni* (Coleoptera: Chrysomelidae), an emerging landscape pest in North America. Location: Durban, South Africa.

07/09/08. International Congress of Entomology (ICE). Invited speaker at the symposium "Biology of the Chrysomelidae (Coleoptera)". Oral presentation. Aggregative oviposition of viburnum leaf beetle [*Pyrrhalta viburni* (Paykull)]: Mechanisms and benefits of an unusual social behavior. Location: Durban, South Africa.

08/13/08. 12th International Behavioral Ecology Congress (ISBE). Poster presentation. Aggregative oviposition of viburnum leaf beetle [*Pyrrhalta viburni* (Paykull)] and a realized benefit of overcoming plant defenses. Location: Ithaca, NY.

02/13/09. Jugatae Seminar series (Entomology department). Invited speaker. Viburnum leaf beetle [*Pyrrhalta viburni* (Paykull)]: from ecology to biological control of an emerging landscape pest. Location: Ithaca, NY.

02/18/09. Plant Insect Group (Ecology and Evolutionary Biology Department). Invited speaker. Viburnum leaf beetle [*Pyrrhalta viburni* (Paykull)]: from ecology to biological control of an emerging landscape pest. Location: Ithaca, NY.

03/20/09. ESA Eastern branch meeting, student competition (poster presentation). Aggregative oviposition behavior of viburnum leaf beetle and plant defenses of North American and European viburnums. Location: Harrisburg, Pennsylvania.

04/21/09. Graduate Student Seminar Series (Entomology Department). Invited speaker. Cooperation, competition, conflict: complexity of a subsocial behavior. Location: Ithaca, New York.

07/24/09. Ph.D defense. Oviposition of viburnum leaf beetle [Pyrrhalta viburni (Paykull)]: from ecology to biological control of an emerging landscape pest. Location: Ithaca, New York.

10/16/09. Cooperation: Self-interest and Mutual interest (One-day conference). Poster presentation. Viburnum leaf beetle aggregative oviposition: cooperation and competition in avoidance of plant defenses. Location: Ithaca, New York.

10/24/09. Cornell University-Penn State University Chemical Ecology meeting. Poster presentation. Viburnum leaf beetle aggregative oviposition: cooperation and competition in avoidance of plant defenses. Location: Ithaca, New York.

01/21/10. Cornell Ecology and Evolutionary Biology 2-day symposium. Egg-crushing through time and space: evolution of plant defenses against a major invasive pest. Location: Ithaca, New York.

02/24/10. Gordon Conference on plant-herbivore interaction. Poster presentation. Can the evolutionary history of plant defenses explain the success of an invasive species? A phylogenetic study of *Viburnum* & a specialist leaf beetle. Location: Galveston, Texas. *Presenter*: Anurag Agrawal.

03/09/10. ESA Eastern branch meeting. Symposium "IDEP (Insect detection, Evaluation and Prediction)". Invited speaker. Invasion of viburnum leaf beetle: Understanding susceptibility to infestation of native and exotic viburnums.

03/09/10. ESA Eastern branch meeting. Student symposium invited speaker (Comstock award recipient). Tale of a Ph.D: viburnum leaf beetle biological control and ovipositional ecology.

03/30/10. Ohio State University Entomology seminar. Invited speaker. Egg-crushing and subsociality: understanding the ecological context of viburnum leaf beetle invasion in North America.

03/31/10. Ohio State University, 1-day outreach seminar for Master Gardeners. Viburnum leaf beetle: biology, ecology, and management of an emerging invasive pest.

12/14/10. ESA National meeting (San Diego, CA). Symposium “ESA showcase of excellence: fostering our innovative and emerging entomologists”. Invited speaker. Understanding the invasion success of viburnum leaf beetle, an emerging landscape pest in the Northeastern U.S.

03/14/11. ESA North Central meeting (Minneapolis, MN). Symposium “Invaders from the East”, invited speaker. Invasion of viburnum leaf beetle, *Pyrrhalta viburni*, in North America: current status and research directions.

08/15/11. Symposium of Insect-Plant Interactions (SIP) meeting (Wageningen, the Netherlands). Poster presentation. A behavioral adaptation to plant defenses facilitates the invasion of viburnum leaf beetle [*Pyrrhalta viburni* (Paykull)] in North America.

01/30/12. Gordon Research Conference on Plant Volatiles: Ecology, Biosynthesis, Regulation and Animal Perception of Floral and Vegetative Volatiles (Ventura, CA). Poster presentation. Alien interference: disruption of volatile-mediated interactions between plants and parasitoids by invasive insect herbivores.

07/02/12. 2nd International Society for Horticultural Science (ISHS) Symposium on Woody Ornamentals of the Temperate Zone (Ghent, Belgium). Poster presentation. Viburnum leaf beetle: ecology and management of an invasive landscape pest.

08/10/12. Ecological Society of America (ESA) National meeting (Portland, OR). Symposium “Community Context of Species’ Range Expansions: Novel Community Associations In Response to Biological Invasions and Climate Change”, invited speaker. Conquering a defense-free space: history and ecological context of viburnum leaf beetle invasion in North America.

11/03/12. 1st Viburnum world summit, Yale University (New Haven, CT), invited speaker. Viburnum leaf beetle: impact of a specialist herbivore on the evolution of defenses in the *Viburnum* genus.

01/22/2013. National Centre of Competence in Research (NCCR) final meeting, Neuchâtel University (Switzerland). Poster presentation. Impact of non-host herbivores on a plant-parasitoid association: a test of the robustness of infochemicals.

02/08/2013: Biology 13 conference, Basel University (Switzerland). Poster presentation. Impact of non-host herbivores on a plant-parasitoid association: a test of the robustness of infochemicals.

02/26/2013: Gordon Research Conference on plant-herbivores interactions (Ventura, CA). Poster presentation. Alien interference: effects of exotic herbivores on volatile-mediated interactions between plants and parasitoids.

04/03/2013: EuroVol mid-project meeting (Florence, Italy), invited speaker. The dazzling effect of exotic perfumes: impact of exotic herbivores on the foraging behavior of native parasitoids.

01/21/2014: Gordon Research Conference on Plant Volatiles (Ventura, CA). Poster presentation. Impact of volatiles induced by exotic herbivores on the attraction of natural enemies.

08/22/2014: Symposium on Insect-Plant Interactions (SIP meeting) (Neuchâtel, Switzerland). 1) Poster presentation. The interplay between pollinator and parasitoid attraction: impact of Brassica rapa floral volatiles on the foraging behavior of parasitoids. 2) Invited speaker. Alien interference : disruption of infochemical networks by invasive insect herbivores.

11/16/2014: Entomological Society of America (ESA) National meeting (Portland, OR). Impact of exotic herbivores on native tritrophic interactions: A case study of the African cotton leafworm, Spodoptera littoralis.

12/15/2014: Joint 2014 Annual Meeting British Ecological Society and Société Française d'Ecologie (Lille, FR). Impact of exotic insect herbivores on chemically mediated plant-parasitoid interactions.

01/21/2015: Socité Entomologique Neuchâteloise (SEN) Seminar series (Museum of Natural History, Neuchâtel, Switzerland). Invited speaker. Impact des insectes invasifs sur l'écologie chimique des relations plantes-insectes.

07/07/2015: International Society of Chemical Ecology (ISCE) meeting (Stokholm, Sweden). Invited speaker: the true cost of exotic perfumes. Impact of exotic herbivores on infochemical networks.

25/06/2016: Ecole Thématique Ecologie Chimique (ETEC) (Roscoff, France). Poster presentation. Conséquences de la domestication de *Brassica rapa* pour les interactions plante-insectes.

25/09/2016: International Congress of Entomology (Olrando, Florida). Oral presentation, Invited speaker. Alien interference: understanding the impact of invasive herbivores on infochemical networks and plant-parasitoid interactions.

2017: Neuchâtel talk, CABI Talk, STUDIUM talk, ESA poster, GDR talk, Monpt chemical ecology talk

2018: ESA eastern branch meeting, ISBCW 2018, ESA national meeting

2019: Perugia meeting

2021: Entomo 21

2023: Chemical Ecology CEFE, BREBCA Portugal

2024: QuadriCosta Rica

RESEARCH SUPPORT

Summer 2004: Petro Canada Company (Pesticide evaluation against viburnum leaf beetle): **$2,500**.
Summer 2005: Cleary Chemical Company (Pesticide evaluation against viburnum leaf beetle): **$2,500**.
2005-2006 academic year: U.S Forest Service (Hemlock wooly adelgid management): **$ 35,000**.
Summer 2006: New York State IPM program (Viburnum leaf beetle management): **$3,000**.
Summer 2007: New York State Biodiversity Research Institute (Viburnum leaf beetle ecological impact): **$1,500,** IR-4 Northeast support (Viburnum leaf beetle management): **$1,000**.
Summer 2008: U.S Forest Service (Gypsy moth pathogens project): **$2,500**.
2011-2014: Inva-Vol project, EuroVol and EUROCORES program, European Science Foundation grant: **$225 000.**
2014-2015: Inva-Vol project, EuroVol and EUROCORES program, European Science Foundation grant 1-year extension: **$75 000**.
2016-2019: National Swiss Foundation grant (Exploring the chemical Ecology of Insect-Slug-Plants interactions): **$500 000**.
2022-2024: U.S. Department of Agriculture's (USDA) Agricultural Marketing Service through grant 21SCBPCA1002-00 (Evaluation of the parasitoid Diadegma semiclausum as a biological control agents against Diamondback moth in the USA): **$97 100**

TEACHING EXPERIENCE

Fall 2004: Teaching assistant (TA) for the course ENTOM 443, Cornell University: Entomology and Pathology of Trees and Shrubs. Teacher: Paul Weston.
Spring 2005: TA for the course ENTOM 277, Cornell University: Natural Enemies Managing Pests: An Introduction to Biological Control. Teachers: Ann Hajek and Jan Nyrop.
Fall 2005: TA for the course ENTOM 452, Cornell University: Insect Ecology. Teacher: Jennifer Thaler.
Spring 2007: TA for the course ENTOM 201, Cornell University: Alien Empire. Teacher: John Losey.
Fall 2007: TA for the course ENTOM 444, Cornell University: Integrated Pest Management. Teachers: John Losey and Antonio DiTommaso.
Spring 2008: TA for the course ENTOM 277, Cornell University: Natural Enemies and Invasive species. Teacher: Jan Nyrop.
Fall 2008: TA for the course ENTOM 4630, Cornell University: Invertebrate pathology. Teacher: Ann Hajek.
Fall 2011-2013, spring 2015: Teacher of the M.S. Biology course "Integrated Pest management/Fighting insect pests", University of Neuchâtel.
Spring 2012-2015: Teacher for the course "Apprentissage par problème (APP): vers une agriculture durable" (learning through experimenting: toward a sustainable agriculture), University of Neuchâtel.

OUTREACH EXPERIENCE

Insectapalooza (Cornell Entomology Department open house 1-day event) 2004-2009: volunteer, co-organizer of Cockroach Races (2004-2009)/Integrated pest management demos (2006-2007).

College of Agriculture and Life Sciences centennial celebration 2005: volunteer.
Cornell Turf & Ornamentals Field Day 2005: volunteer.
Intenational Behavioral Ecology Congress (ISBE) 2008: volunteer and session coordinator.
"Expanding your horizons in science and mathematics" conference (Cornell University) 2009-2010: volunteer.
Self interest and mutual interest one-day conference (Cornell University) 2009: volunteer and press rapporteur.
Cornell University-Penn State University chemical ecology meeting 2009: volunteer and event coordinator.
One-day outreach seminar 2010, Ohio State University: volunteer/viburnum leaf beetle expert.

COMMITTEE AND PROFESSIONAL SERVICES

Provided reviews for the following journals: Ecology letters, Ecology, New Phytologist, Molecular Ecology, Functional Ecology, Animal Behaviour, Oecologia, Oikos, Biological Invasions, Frontiers in Chemical Ecology, Journal of Chemical Ecology, Biology letters, Ecological Entomology, Environmental Entomology, Entomologia Experimentalis et Applicata, Diversity, Journal of Pest Science, Basic and Applied Ecology, Journal of arthropod-plant interactions, Plant Biology, Insects.
Member of the ESA (Entomological Society of America): 2004-2011; 2014; 2017
Member of the Insectary and Greenhouse committee (Entomology Department, Cornell University): 2005-2007.
Entomological Society of America (ESA) student representative (Entomology Department, Cornell University): 2006-2008.
Jugatae President (Entomology Department graduate student organization, Cornell University): 2007.

RECOMMENDERS

More information and reference letters can be obtained from:

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Cornell University
Corson Hall, Ithaca, NY – USA 14853
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Javid Kashefi, Support Scientist

Education

Aristotle university of Thessaloniki, School of Agricultural, BS. in Plant propagation, 1985-1990

Aristotle university of Thessaloniki, School of Agricultural, MS. entomology, 1990-1992

Work Experience

1992 - present Support scientist, USDA-ARS European Biological Control Laboratory, Thessaloniki, Greece and Montpellier, France

Worked in plant pathology in collaboration with USDA ARS laboratory in Forte Ditrich, MD and department of plant pathology at school of agriculture, university of Thessaloniki.
2012, After a three weeks intensive mosquito identification course at university of Florida, now I am a certified mosquito taxonomist.
2024, After a two weeks intensive course in tick taxonomy at the US national tick collection center, now I am experienced in tick identification.
2013 to 2019, Voluntary work at Natural history museum London helping with curation of their tick collection.
Extensive traveling experience in Europe, north Africa, middle east, southeast Asia and the US for collection and planning and execution of experiments for biological control of weeds and arthropod pests and vectors.
Some of my collaborators are: State university of Montana, UC Davis, USDA ARS laboratories in Albany, CA, USDA ARS laboratory in Edinburg, TX, National Institute of Veterinary Research and Plant Protection Research Institute, Hanoi, Vietnam. I have a very close collaboration with Agricultural university of Plovdiv, Bulgaria, Joint Genomic Center in Sofia, Bulgaria, Agricultural university of Tirana, Albania, Albanian ministry of Agriculture, USAID in Albania, Agricultural university of Athens, School of agriculture, Aristotle university of Thessaloniki, Greek forestry service, Foundation for Research and Technology, Crete, Greece, and Benaki Phytopathological Institute, Athens Greece.

Some Publications

Bon MC., Kashefi J., Soforza R., How sweet is the extrafloral nectar secreted by the invasive alien Tree of Heaven, Ailanthus altissima Mill.?; September 2023International Journal of Plant Research 11(1):1135

Chaskopoulou A., Miaoulis M., Kashefi J., Ground ultra low volume (ULV) space spray applications for the control of wild sand fly populations (Psychodidae: Phlebotominae) in Europe. Acta Tropica, Volume 182, 2018, pp. 12-13

Goolsby J., Racelis A., Moran P.J., Kirk A., Kashefi J., Smith L., Bon M., Lacewell R., Vacek A., Martinez-Jimenez M., Perez De Leon A.A., Integration of mechanical topping methods to accelerate biological control of Arundo donax. Integration of mechanical topping methods to accelerate biological control of Arundo donax. International Symposium on Biological Control of Weeds. p. 290 2019

Marshall M., Goolsby J., Vacek A.T., Mastoras A., Kashefi J., Chaskopoulou A., Smith L., Badillo I., Reilly F.J., Perez De Leon A.A., Racelis A., Biotic and abiotic factors influencing infestation levels of the arundo leafminer, Lasioptera donacis, in its native range in Mediterranean Europe. Subtropical Agriculture and Environments. 69:8-18 2018.

Marshall M., Goolsby J., Vacek A., Moran P., Kashefi J., Densities of the arundo wasp, Tetramesa romana (Hymenoptera: Eurytomidae) across its native range in Mediterranean Europe and introduced ranges in North America and Africa, Biocontrol Science and Technology 28(8):1-14 · July 2018

Chaskopoulou A., Miaoulis M., Kashefi J., Ground ultra-low volume (ULV) space spray applications for the control of wild sand fly populations (Psychodidae: Phlebotominae) in Europe. *Acta Tropica* 182 54–59 (2018).

CURRICULUM VITAE: René F.H. SFORZA

Senior Research Entomologist, PhD, HDR
(equivalent to DR in France)
Weeds Research Leader

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email: rsforza@ars-ebcl.org
https://orcid.org/0000-0002-1737-2033
https://www.researchgate.net/profile/Rene-Sforza

Degrees:

2017 HDR (Habilitation à Diriger des Recherches), UNIVERSITY of Montpellier, France entitled "A multidisciplinary approach of plant and ecosystem protection with classical biological control"

1998 Ph.D. Thesis in Entomology and Plant pathology, UNIVERSITY of Paris VI, France.

Work experience:

2000 - present: Research Entomologist at USDA-ARS-EBCL, Montpellier, France.

1999 Postdoctoral fellow at INRAe - Laboratory "Vine and wine" (Colmar) & Lab. Phytoparasitologie (Dijon) "Grapevine leafroll epidemiology: Search for natural scale insect vectors (Hemiptera: Coccoidea) and strategies in biological control, RT-PCR detection of closteroviruses in plants and insects."

1994-98 Ph.D. at INRAe, Phytoplasma Laboratory in Dijon, France. Contract with the Rhône valley's viticultural organisations. "Epidemiology of Bois noir disease of grapevine: search for insect vectors and biology of *Hyalesthes obsoletus* Sign. (Hemiptera: Cixiidae); spread and technical control."

Research topics and projects at EBCL (2000-present):

Topics:

1. Classical biocontrol of weeds and insect pests using arthropods and pathogens as specific agents;
2. Foreign exploration for the discovery of biological control agents;
3. Host specificity testing and evaluating ecological interactions;
4. Phylogeography of invasive target species.

Projects:

1. Current project on an insect pest:
    a. Bagrada bug (Bagrada hilaris - Pentatomidae) and the scelionid wasp, *Gryon aetherium*.

2. Current projects on weeds and their potential biocontrol candidates when known:
    a. Tree of heaven (*Ailanthus altissima*, Simaroubaceae) and the eriophyid mite *Aculus taihanganensis*,
    b. Stinkwort (*Dittrichia graveolens*, Inulaceae),
    c. Swallow worts (*Vincetoxicum rossicum* and *V. nigrum* - Asclepiadaceae) and the Chrysomelid beetle *Chrysochus asclepiadeus*,
    d. Sahara mustard (*Brassica tournefortii*, Brassicaceae),
    e. Medusahead (*Taeniatherum caput-medusae* - Poaceae), African wiregrass (*Ventenata dubia* Poaceae),
    f. French broom (*Genista monspessulana* - Fabaceae) and the psyllid *Arytinnis hakani* and the weevil *Lepidapion argentatum*.

Teaching experience:

Pluri-annual courses in Biological control and insect diversity at Supagro, Montpellier, and the University of Montpellier

Recent publications (2018-2024):

Martel, G., Hogg BN, Sforza RFH. Anticipating the arrival of a new stinkbug pest in continental Europe: what can we learn from preliminary host specificity tests for biocontrol? *Entomologia generalis* (Accepted).

Sforza, RFH (2023) La lutte biologique, un intérêt majeur pour la planète. La Recherche, Sept-Dec 2023; 575. p76-83.

Bon MC, Guermache F, Kashefi J, Sforza RFH (2023) How sweet is the extrafloral nectar secreted by the hellish invasive Tree of heaven, *Ailanthus altissima* Mill.? International Journal of Plant Biology & Research 11(1): 1135. https://doi.org/10.47739/2333-6668/1135.

Mainardi CE, Peccerillo C, Paolini A, Cemmi A, Sforza RFH, Musmeci S and Cristofaro M (2023) Using the irradiation technique to Predict the sperm competition mechanism in *Bagrada hilaris* (Burmeister) (Hemiptera: Pentatomidae): Insights for a future management strategy Insects 14(8): 681. .

Lesieur V, Sforza RFH, Shaw R, Sheppard A (2023) Prioritising invasive alien plants affecting the environment for classical biological control in Europe: a reanalysis. *Weed Research,* 63(4), pp. 218-231.

Martel, G., Sforza, R.F.H. (2023) Development, survivorship and reproduction of *Gryon aetherium* Talamas (Hymenoptera: Scelionidae), an egg parasitoid of Bagrada hilaris (Burmeister) (Hemiptera: Pentatomidae) under eight constant temperatures. *Biological Control,* 180: 105185. https://doi.org/10.1016/j.biocontrol.2023.105185

Hoelmer KA, Sforza RFH, and Cristofaro M (2023) Accessing biological control genetic resources: the United States perspective. BioControl 68, 269–280. https://doi.org/10.1007/s10526-023-10179-5

Cristofaro M, Sforza RFH, Roselli G, Paolini A, Cemmi A, Musmeci S, Anfora G, Mazzoni V, Grodowitz M. (2022) Effects of Gamma Irradiation on the Fecundity, Fertility, and Longevity of the Invasive Stink Bug Pest *Bagrada hilaris* (Burmeister) (Hemiptera: Pentatomidae). *Insects*. 13(9): 787. DOI 10.3390/insects13090787

Borowiec N & Sforza RFH (2022). Classical biological control. pp31-42. In "Extended Biocontrol. Eds: X. Fauvergue et al. Springer. 327p.

Sforza RFH (2021) The diversity of biological control agents. Chapter 1, pp-1-36. In: *Biological control: Global Impacts, Challenges and Future Directions of Pest Management* (Ed. P.G. Mason). CSIRO Publishing, Melbourne, 626p.

Martel G and Sforza RFH (2021) Evaluation of three cold storage methods of Bagrada hilaris (Hemiptera: Pentatomidae) and the effects of host deprivation for an optimized rearing of the biocontrol candidate Gryon gonikopalense (Hymenoptera: Scelionidae). *Biological Control. Vol 163.* https://doi.org/10.1016/j.biocontrol.2021.104759

Kerdellant E, Thomann T, Sheppard A., Sforza R.F.H (2021) Impact and host specificity of *Lepidapion argentatum* (Col., Brentidae), a promising candidate for the biological control of the invasive *Genista monsspessulana* (Fabaceae). *Insects*, *12*(8), 691. https://doi.org/10.3390/insects12080691

Marini F, Profeta E, Vidović B, Petanović R, de Lillo E, Weyl P, Hinz HL, Moffat CE, Bon MC, Cvrković T, Kashefi J, Sforza RFH, M Cristofaro (2021) Host range evaluation of *Aculus mosoniensis* (Acari: Eriophyidae), a biological control agent of the tree of heaven (*Ailanthus altissima*). *Insects*, *12*(7), 637. https://doi.org/10.3390/insects12070637

Martel G, Scirpoli F and RFH Sforza (2021) How an egg parasitoid responds to a rare stinkbug oviposition behavior: the case of Gryon gonikopalense Sharma (Hymenoptera: Scelionidae) and Bagrada hilaris Burmeister (Hemiptera: Pentatomidae). *Entomologia Generalis.* 42: 3, p. 457 - 470 10.1127/entomologia/2021/1256

Gaskin JF, Endriss SB, Fettig CE Hufbauer RA, Norton AP, RFH Sforza (2021) One genotype dominates a facultatively outcrossing plant invasion. *Biological Invasions* 23 (1): 1901–1914. https://doi.org/10.1007/s10530-021-02480-0

Martel, G., Sforza, R.F.H. (2021) Catch me if you can: novel foraging behavior of an egg parasitoid, *Gryon gonikopalense*, against the stinkbug pest, *Bagrada hilaris*. Journal of Pest Science 94(4): 1161–1169. https://doi.org/10.1007/s10340-020-01325-4

Conti E., G. Avila, B. Barratt, F. Cingolani, S. Colazza, S. Guarino, K. Hoelmer, R.A. Laumann, L. Maistrello, G. Martel, E. Peri, C. Rodriguez-Saona, G. Rondoni, M. Rostas, P. Roversi, RFH. Sforza, L. Tavella & E.

Wajnberg (2020) Biological control of invasive stink bugs: global state and future. Entomologia Experimentalis et Applicata, 169: 28–51.https://doi.org/10.1111/eea.12967

Pervukhina-Smith, I., Sforza, R.F.H., Cristofaro, M., Novak, SJ. (2020) Genetic analysis of invasive populations of Ventenata dubia (Poaceae): an assessment of propagule pressure and pattern of range expansion in the Western United States. Biological Invasions 22, 3575–3592. .https://doi.org/10.1007/s10530-020-02341-2

Borowiec N & Sforza RFH (2020) Lutte biologique par acclimatation. In "Biocontrole: Eléments pour une protection agroécologique des cultures. Eds: X. Fauvergue et al. QUAE. 376p.

Wanjiru Clarke C, Calatayud PA, Sforza RFH, Ngeh Ndemah R, Nyamukondiwa C (2019) Parasitoids' Ecology and Evolution Editorial - Population and Evolutionary Dynamics, Front. Ecol. Evol., 7:485. doi: 10.3389/fevo.2019.00485

Sforza, RFH (2019) La lutte biologique contre les adventices est-elle possible? Cahier de Réussir Fruits & legumes, Prosp&ctives, 399: 66-70.

Milbrath LR, Dolgovskaya R, Volkovitsh M, Sforza RFH, Biazzo J (2019) Photoperiodic Response of Abrostola asclepiadis (Lepidoptera: Noctuidae), a Candidate Biological Control Agent for Swallow-worts (Vincetoxicum, Apocynaceae). The Great Lakes Entomologist, 52: 71-77.

Martel G, Augé M, Talamas E, Roche M, Smith L, & R.F.H. Sforza (2019) First laboratory evaluation of Gryon gonikopalense (Hymenoptera: Scelionidae), as potential biological control agent of Bagrada hilaris (Hemiptera: Pentatomidae). *Biological Control*. 135: 48-56 https://doi.org/10.1016/j.biocontrol.2019.04.014

Kerdellant E, Thomann T., Vitou J, Sheppard , Giusto C, Simonot O, Sforza RFH. (2019) The double life cycle of *Lepidapion argentatum*, a potential candidate for the biological control of French Broom. *Biocontrol Science and Technology*, 29(8): 773-785.

Bitume EV, Moran PJ, Sforza RFH (2019) Impact in quarantine of the galling weevil Lepidapion argentatum on shoot growth of French broom (*Genista monspessulana*), an invasive weed in the western U.S. *Biocontrol Science and Technology*, 29(7): 615-625.

Shaw RH, Ellison CA, Marchante H, Pratt CF, Schaffner U, Sforza RFH & Deltoro V (2018) Weed biocontrol in the EU: from serendipity to strategy. *BioControl* 63(3) 333-347 DOI: 10.1007/s10526-017-9844-6

Daane KM, Middleton MC, Sforza RFH, Kamps-Hughes N, Watson GW, Almeida RPP, et al. (2018) Determining the geographic origin of invasive populations of the mealybug *Planococcus ficus* based on molecular genetic analysis. PLoS ONE 13(3): e0193852. https://doi.org/10.1371/journal.pone.0193852

RODRIGO KRUGNER, Ph.D

Supervisory Research Entomologist/Research Leader

United States Department of Agriculture - Agricultural Research Service, San Joaquin Valley Agricultural Sciences Center, 9611 S. Riverbend Ave, Parlier, California 93648. Phone: 559-596-2887, E-mail: Rodrigo.Krugner@usda.gov.

EDUCATION

UNIVERSITY OF CALIFORNIA, RIVERSIDE
Ph. D., Entomology (2007)
Dissertation title: *Population Ecology of Homalodisca vitripennis (Germar) (Hemiptera: Cicadellidae) and Host-Finding and Selection Behaviors of Associated Egg Parasitoids*

CALIFORNIA STATE UNIVERSITY, FRESNO
M. S., Plant Science (2003)
Thesis title: *Macrocentrus iridescens (Hymenoptera: Braconidae), a Parasitoid of the Obliquebanded Leafroller (Lepidoptera: Tortricidae)*

UNIVERSIDADE DE SÃO PAULO, ESCOLA SUPERIOR DE AGRICULTURA "LUIZ DE QUEIROZ"
B. S., Agronomist Engineer (2000)

PROFESSIONAL EXPERIENCE

SUPERVISORY RES. ENTOMOLOGIST, USDA-ARS (March 2022 – Present)
RESEARCH ENTOMOLOGIST, USDA-ARS (September 2007 – March 2022)
GRADUATE STUDENT RESEARCHER (August 2003 – September 2007)
DEPARTMENT OF ENTOMOLOGY, UNIVERSITY OF CALIFORNIA – RIVERSIDE.
STAFF RESEARCH ASSOCIATE I (August 2001 – August 2003)
LABORATORY ASSISTANT II (August 2000 – August 2001)
DIVISION OF INSECT BIOLOGY, DEPARTMENT OF ENVIRONMENTAL SCIENCE, POLICY, AND MANAGEMENT. UNIVERSITY OF CALIFORNIA – BERKELEY.

SHORT BIO

Dr. Krugner has a multidisciplinary approach for understanding plant-pathogen-insect interactions, elucidating insect behaviors controlled by chemo- and physico-sensory activation, and developing novel methods of pest control. His research was the first to disentangle complex aspects of drought-mediated effects on the spread of a vector-borne plant pathogen. He identified crop irrigation regimes that reduce vector populations, alter vector dispersal behaviors, and decrease or increase vector transmission efficiency of *X. fastidiosa*. Dr. Krugner identified almond nursery stock as a pathway for *X. fastidiosa* infection of almond and provided solutions such as controlling specific host plants in vector habitats and using *X. fastidiosa*-resistant rootstocks. The latter was demonstrated to be an important factor for nursery plant protection against bacterial infection and also as a curative factor in orchards. To thrive in crops, the glassy-winged sharpshooter (GWSS, *Homalodisca vitripennis*) must communicate by exchanging vibrational mating signals. Sophisticated efforts in vibrometry elucidated GWSS communication and designed candidate disruptive signals for playback interference under field conditions. Dr. Krugner discovered that vibrational signals emitted by a dominant GWSS female suppress signaling activity of other females on the plant. Playback of such signal through vineyard trellis significantly reduced mating of GWSS on grapevines compared to control, cementing vibrational disruption as a novel control method available for integration with current GWSS management tools in vineyards. In biological control research, Dr. Krugner has worked on hymenopteran parasitoids focusing on host finding and acceptance behaviors, population ecology, and biology. The following are the most relevant publications for this project.

RELEVANT PEER REVIEWED MANUSCRIPTS

Krugner, R., Daane, K.M., Lawson, A., and Yokota, G.Y. Biology of *Macrocentrus iridescens* (Hymenoptera: Braconidae), a parasitoid of the obliquebanded leafroller (Lepidoptera: Tortricidae). Environ. Entomol. 34:336-343. 2005.

Krugner, R., Daane, K.M., Lawson, A.B., and Yokota, G.Y. Temperature-dependent development of *Macrocentrus iridescens* (Hymenoptera: Braconidae) as a parasitoid of the obliquebanded leafroller (Lepidoptera: Tortricidae): Implications for field synchrony of parasitoid and host. Biol. Cont. 42:110-118. 2007.

Krugner, R., Johnson, M.W., Groves, R.L., and Morse, J.G. Host specificity of *Anagrus epos*: A potential biological control agent of *Homalodisca vitripennis*. Biocontrol 53:439-449. 2008.

Krugner, R., Johnson, M.W., Daane, K.M., and Morse, J.G. Olfactory responses of the egg parasitoid, *Gonatocerus ashmeadi* Girault (Hymenoptera: Mymaridae) to host plants infested by *Homalodisca vitripennis* (Germar) (Hemiptera: Cicadellidae). Biol. Cont. 47:8-15. 2008.

Krugner, R., Groves, R.L., Johnson, M.W., Flores, A.P., Hagler, J.R., and Morse, J.G. Seasonal population dynamics of *Homalodisca vitripennis* (Hemiptera: Cicadellidae) in sweet orange trees maintained under continuous deficit irrigation. J. Econ. Entomol. 102:960-973. 2009.

Krugner, R., Johnson, M.W., Morgan, D.J.W., and Morse, J.G. Production of *Anagrus epos* Girault (Hymenoptera: Mymaridae) on *Homalodisca vitripennis* (Germar) (Hemiptera: Cicadellidae) eggs. Biol. Cont. 51:122-129. 2009.

Krugner, R. Differential reproductive maturity between geographically separated populations of *Homalodisca vitripennis* (Germar) in California. Crop Prot. 29:1521-1528. 2010.

Wang, X.G., Johnson, M.W., Opp, S.B., Krugner, R., and Daane, K.M. Honeydew and insecticide-bait as competing food resources for a fruit fly and common parasitoids. Entomol. Exp.

Appl. 139:128-137. 2011.

Hagler, J.R., Blackmer, F., Krugner, R., Groves, R.L., Morse, J.G., and Johnson, M.W. Gut content examination of the citrus predator assemblage for the presence of *Homalodisca vitripennis* remains. Biocontrol 58:341-349. 2012.

Krugner, R., Hagler, J.R., Groves, R.L., Sisterson, M.S., Morse, J.G., and Johnson, M.W. Plant water stress effects on the net dispersal rate of the insect vector *Homalodisca vitripennis* (Hemiptera: Cicadellidae) and movement of its egg parasitoid, *Gonatocerus ashmeadi* (Hymenoptera: Mymaridae). Environ. Entomol. 41:1279-1289. 2012.

Krugner, R. Suitability of non-fertilized eggs of *Homalodisca vitripennis* for the egg parasitoid *Gonatocerus morrilli*. Biocontrol 59:167-174. 2014.

Krugner, R., Sisterson, M.S., Chen, J.C., Stenger, D.C., and Johnson, M.W. Evaluation of olive as a host of *Xylella fastidiosa* and associated sharpshooter vectors. Plant Dis. 98:1186-1193. 2014.

Krugner, R., Wallis, C.M., and Walse, S.S. Attraction of the egg parasitoid, *Gonatocerus ashmeadi* Girault (Hymenoptera: Mymaridae) to synthetic formulation of a (E)-β-ocimene and (E,E)-α-farnesene mixture. Biol. Cont. 77:23-28. 2014.

Burbank, L., Rogers, E.E., Sechler, A.J., Magdeleno, M., and Krugner, R. Experimental infection of California Ripe Olive cultivars with *Xylella fastidiosa* subspecies pauca De Donno and acquisition by glassy-winged sharpshooter. Phytofrontiers: https://doi.org/10.1094/PHYTOFR-04-24-0037-R. 2024.

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Education:

University of California, Berkeley	Entomology	Ph.D., 1988
University of California, Santa Barbara	Zoology	B.A., 1979

Professional Experience:

Assistant (2000-02), Associate (2002-06), and Professor Cooperative Extension Specialist (2006-present). Department of Environmental Science, Policy, and Management, UC Berkeley.
Assistant (1989-92), Associate (1992-98) and Research Specialist (1998-2000). Division of Biological Control, UC Berkeley. Stationed at the Kearney Agricultural Center, Parlier, CA.
Lecturer (1989). Department of Conservation and Resource Studies, UC Berkeley.
Post Doctorate Researcher (1988-89). Division of Biological Control, UC Berkeley.
Graduate Student Research Assistant (1984-88). UC Berkeley.
Research Assistant III (1982-83). California Department of Food and Agriculture.
Laboratory Assistant I (1980, 1981). Department of Entomological Sciences, UC Berkeley.

Committee/Organization Participation highlights:

Co-Chair, Center for Biological Control (2000-2024); Co-Chair, Van Den Bosch Memorial Scholarship (2005-present); Chair, Kearney Research and Extension Center Research Advisory Committee (2000–2005, 2016-present); Liaison, UC Research Advisory Committee, Calif. Table Grape Comm. (2005-2023); Member, ESA Awards Committee (2019-2023); Member, Calif. Dept. Pesticide Regulation Alternatives to Chlorpyrifos (2019-2022); Member, Kearney Agricultural Research and Extension Center – Strategic Plan (2014-2017); Member, National Institute of Food and Agriculture Grant Panel (2010); Chair, Insectary and Quarantine Oversight Committee (2003-present); Co-chair: DANR "Biological Control" Workgroup, (2001-2007); Co-chair: California Conference on Biological Control (2000-2012); Chair, UC Statewide Integrated Pest Management Project Biological Controls Workgroup (1997-2000); Member (Secretary–2002, President–2003), Western Regional Bio-Control; Co-organizer: Entomological Society of America Symposia (numerous dates)

Awards and Honors:

2023 Lifetime Achievement Award San Joaquin Valley Wine Grape Association • 2023 Entomological Society of America's P-IE Integrated Pest Management Team Award: "Sustainable SWD Management Team" • 2023 Entomological Society of America, Journal of Economic Entomology – Publication Editors' Choice Award (Abram et al. 2022). 2022 Entomological Society of America, Journal of Economic Entomology – Publication Editors' Choice Award (Tait et al. 2021). 2021 C.W. Woodworth Award, Pacific Branch Entomological Society of America • 2021 Friends of IPM Award, Southern IPM Center for "Sustainable SWD Management Team" • 2020 Distinguished Scientist Award, International Organization of Biological Control (Nearctic Region) • 2017 International IPM Award of Recognition (European Grapevine Moth) • 2017 California Environmental Protection Agency (DPR) "IPM Achievement Award" Virginia Creeper Leafhopper • 2015-2016 University of California Cooperative Extension Distinguished Service Award "Outstanding Team" • Adjunct Faculty Essig Museum of Entomology (2017-present) • Adjunct Faculty Dept Plant Science, CSU Fresno • 2005 Distinguished Achievement Award in Extension, Entomological Society of America (Pacific Branch) • 1998 Griswold Lecture, Cornell University • 1988 Magy Memorial Graduate Student Scholarship, UC Berkeley • 1987 Outstanding Graduate Instructor, UC Berkeley

Professional Organizations:

Entomological Society of America • Pacific Coast Entomological Society • International Organization for Biological Control • Association of Applied Insect Ecologists • Sigma Xi Society • California Certified Organic Farmers

Professional Activities:

Publications: ca. 230 peer-reviewed (journal-only, Google Scholar H-index 54) and >350 grower-oriented reports and publications • Presentations: >830 grower-oriented and research symposia • Grants: > $35 million received as PI or co-PI (1990-present) • Reviewer: > 300 Manuscripts, > 250 Competitive grants • Associate Editor: *California Agriculture* (2009-2023); Subject Editor *Journal of Pest Science* (2020-present); Editorial Board *Biological Control* (2012-present), *Insects* (2018-present), *Entomologia* (2016-2018), co-editor special issue *Chemical Ecology of Parasitic Hymenoptera* (2014-2015).

Program Overview: Research and Outreach Activities

Kent Daane has laboratories on the Berkeley Campus and the Kearney Agricultural Center. His laboratory members develop IPM and sustainable programs for insect and mite pests, primarily in vineyards and orchards (pistachio, almond, stone fruit, and olive). Research on and extension of arthropod pest management systems is the primary objective. Biological control, insect ecology, natural enemy biology, and pheromone chemistry are the lab's primary contributions to IPM. In vineyards, Kent has worked with mealybugs, leafhoppers and vineyard pathogens and their vectors; in pistachios and almonds, navel orangeworm, Lygus, San Jose scale, stink bugs, leaffooted bugs, and the obliquebanded leafroller have been key insects studied; in stone fruit work focused on oriental fruit moth, peach twig borer and omnivorous leafroller. Over the past 30 years, he participated in programs receiving >$37,000,000, with $23,000,000 directed to his lab. Based both on the Berkeley campus and Kearney has provided an excellent opportunity to work with many excellent growers, commodity organizations, PCAs and industry representatives and researchers in and outside of the University of California system.

Commonly Cited Articles (Google Scholar H-Index 63; i10-index 233, top 30 in BioControl):

Asplen , M. K., Anfora, G., Biondi, A. Choi, D.-S. Chu, D., Daane, K. M. et al. 2015. Invasion biology of spotted wing drosophila (*Drosophila suzukii*): a global perspective and future priorities. *Journal of Pest Science* 88(3): 469–494.

Daane, K. M., and Johnson, M. W. 2010. Olive fruit fly: Managing an ancient pest in modern times. *Annual Review of Entomology* 55: 151-169.

Tauber, M. J., Tauber, C A., Daane, K. M., and Hagen, K. S. 2000. New tricks for old predators: implementing biological control with *Chrysoperla*. *American Entomologist* 46(1): 26-38.

Haye, T., Girod, P., Cuthbertson, A. G. S., Wang, X. G., Daane, K. M., Hoelmer, K. A., Baroffio, C., Zhang, J. P., and Desneux, N. 2016. Current SWD IPM tactics and their practical implementation in fruit crops across different regions around the world *Journal of Pest Science* 89(3): 643-651.

Daane, K. M., Wang, X.-G., Biondi, A., Miller, B. E., Miller, J. C., et al. 2016. First foreign exploration for Asian parasitoids of *Drosophila suzukii*. *Journal of Pest Science* 89(3): 823-835.

Daane, K. M., Sime, K. R., Fallon, J., and Cooper, M. L. 2007. Impacts of Argentine ants on mealybugs and their natural enemies in California's coastal vineyards. *Ecological Entomology* 32: 583-596.

Tsai, C.-W., Rowhani, A., Golino, D. A. Daane, K. M., and Almeida, R. P. P. 2010. Mealybug transmission of grapevine leafroll viruses: an analysis of virus-vector specificity. *Phytopathology* 100(8): 830-834.

Millar, J. G., Daane, K. M., McElfresh, J. S., Moreira, J., Malakar-Kuenen, R., Guillen, M., and Bentley, W. J. 2002. Development and optimization of methods for using sex pheromone for monitoring the mealybug *Planococcus ficus* (Homoptera: Pseudococcidae) in California vineyards. *Journal of Economic Entomology* 95(4): 706-714.

Hogg, B. N., Bugg, R. L., and Daane, K. M. 2011. Attractiveness of common insectary and harvestable floral resources to beneficial insects. *Biological Control* 56: 76-84.

Costello, M. J., and Daane, K. M. 1998. Influence of ground covers on spider (Araneae) populations in a table grape vineyard. *Ecological Entomology* 23: 33-40.

SERGUEI V. TRIAPITSYN

A. Vita

Principal Museum Scientist
Entomology Research Museum
Department of Entomology
University of California
Riverside, CA 92521
Tel.: (951) 827-7817
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E-mail: serguei.triapitsyn@ucr.edu

Education:
Moscow Timiriazev Agricultural Academy, USSR, Ph.D., 1991, Agricultural Entomology.

Professional Experience:
Principal Museum Scientist, University of California, Riverside, July 1994 to present.
Postdoctoral Researcher, University of California, Riverside, November 1991 to June 1994.

Current Job Responsibilities:
Management of the UCR Entomology Research Museum and the UCR Quarantine Facility; research on taxonomy and biology of parasitic Hymenoptera.

Professional Interests:
Systematics and biology of parasitic Hymenoptera, particularly the families Mymaridae, Aphelinidae, Encyrtidae, Trichogrammatidae, and thrips-attacking Eulophidae; classical biological control, foreign exploration, and introduction of natural enemies, especially of egg parasitoids.

B. Publications (over 360 including 9 monographs and over 250 scientific articles in peer reviewed journals)

Most recent publications:

Triapitsyn, S. V. & T. Adachi-Hagimori. 2024. A new record of the fairyfly *Cosmocomoidea tenuis* (Hymenoptera: Mymaridae) from Japan, with notes on its host associations. Applied Entomology and Zoology 59 (3): 279-285.

Triapitsyn, S. V. 2024. Review of *Cremnomymar* species (Hymenoptera: Mymaridae) in mainland South America, with a new generic synonymy. Zootaxa 5463 (1): 25-46.

Aguirre, M., G. Logarzo, S. Triapitsyn, H. Diaz-Soltero, S. Hight & O. Bruzzone. 2024. Effect of egg production dynamics on the functional response of two parasitoids. PLoS ONE, 19 (3): e0283916 (pp. 1-27). https://doi.org/10.1371/journal.pone.0283916.

Triapitsyn, S. V., Y. Yasuhara, T. Adachi-Hagimori & M. Tsukada. 2024. Fairyfly egg parasitoids (Hymenoptera: Mymaridae) of the invasive lace bug *Corythucha marmorata* (Uhler) (Hemiptera: Tingidae) in Japan. Journal of Asia-Pacific Entomology 27 (1): 102201 (8 pp.). https://doi.org/10.1016/j.aspen.2024.102201

O'Dea, J. K., J. M. Milnes, S. V. Triapitsyn & P. F. Rugman-Jones. 2023. *Baryscapus rugglesi* (Rohwer, 1919) (Hymenoptera: Eulophidae) discovered in western North America: Redescription, notes on biology, and implications as a parasitoid of its host, *Agrilus cuprescens* (Ménétries, 1832) (Coleoptera: Buprestidae). Pan-Pacific Entomologist 99 (4): 246-265.

Triapitsyn, S. V. 2023. Taxonomy of the Palaearctic species of *Anaphes* Haliday, 1833 (Hymenoptera: Mymaridae), with special focus on their identity and diversity in Finland. Annales Zoologici Fennici 60 (1): 127-197.

Kusuhara, H. & S. V. Triapitsyn. 2023. A new host record for the leafhopper egg parasitoid *Lymaenon aureus* (Girault, 1911) (Hymenoptera: Mymaridae). Japanese Journal of Entomology (New Series) 26 (1): 8-11.

Ortis, G., S. V. Triapitsyn & L. Mazzon. 2023. Two new host records for *Centrodora italica* Ferrière (Hymenoptera, Aphelinidae) from eggs of Tettigoniidae (Orthoptera, Ensifera) in northeastern Italy. ZooKeys 1156: 25-31.

Triapitsyn, S. V., H. Kusuhara, P. F. Rugman-Jones & T. Adachi-Hagimori. 2023. Revised molecular characterization of *Gonatocerus cincticipitis* Sahad (Hymenoptera: Mymaridae), an egg parasitoid of green rice leafhopper *Nephotettix cincticeps* (Uhler) (Hemiptera: Cicadellidae) in Japan. Journal of Asia-Pacific Entomology 26 (2): 102069 (pp. 1-12). https://doi.org/10.1016/j.aspen.2023.102069

Triapitsyn, S. V., P. F. Rugman-Jones, M. I. Abul-Sood & N. S. Gadallah. 2023. On the genetic identity of *Gonatocerus aegyptiacus* (Hymenoptera Mymaridae) from Egypt. Bulletin of Insectology 76 (1): 127-132.

Triapitsyn, S. V., P. F. Rugman-Jones, H. Kusuhara, R. Nakano, P. Janšta, S. Arikawa & T. Adachi-Hagimori. 2023. Genetic analysis reveals conspecificity of two nominal species of *Anaphes* fairyflies (Hymenoptera: Mymaridae), egg parasitoids of *Oulema* leaf beetle (Coleoptera: Chrysomelidae) pests of cereal crops in Europe and of rice in East Asia. PLOS One 18 (1): e0273823 (pp. 1-14). https://doi.org/10.1371/journal.pone.0273823.

Curriculum Vitae

Christos Vasilikiotis, MSc, PhD
Associate Dean for Research
Associate Professor
Chair of Sustainable Agriculture & Management Dept.
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EDUCATION & TRAINING

Bachelor of Science:
Biology Department, Aristotelian University, Thessaloniki – 1983

Doctoral Studies:
Department of Molecular and Cell Biology. Ph.D. degree in Molecular and Cell Biology, University of California at Berkeley, CA, U.S.A. 1991

Master of Science in Agronomy:
Dept. of Agronomy. M.Sc. in Agronomy, Iowa State University, U.S.A 2018

Training in Agronomy, Agroecology & Organic Agriculture:
Nova Scotia Agricultural College, online course on Organic Field Crop Management, Fall Semester 2003-2004.

Training as a visiting scholar in Agroecology:
Dept. of Environmental Science, Policy & Management. University of California at Berkeley, CA, U.S.A. 1997-1999. Completed the following Upper Division courses as a visiting scholar:

1. Terrestrial Resource Ecology
2. Plant Physiology
3. Insect Ecology
4. Biological Control
5. California Plant Life & Laboratory
6. Garden Ecosystems
7. Microbial Ecology & Laboratory
8. Soil Characteristics & Laboratory
9. Soil Microbiology
10. Statistics in Biology
11. Agroecology
12. International Development

Professional Development
Member of EU CAP Network Focus Group on "Regenerative agriculture for soil health: How can regenerative agriculture practices help farmers to restore, protect and improve soil health and productivity? https://eu-cap-network.ec.europa.eu/focus-group-regenerative-agriculture-soil-health_en. 2023-2024

Participation in an International training activity on Conservation Agriculture organized by the CAMA project (Conservation Agriculture in the Mediterranean Area) in Foggia, Italy. 15-18 March 2023.

TP Organics workshop on "Co-creating solutions for soil health in Living Labs – Soil Mission call 2023". 11 May 2023.

Wageningen University, Soil Biology Lab Skills Training Course. Wageningen, Netherlands

13–17 May, 2019.

Technological Educational Institute of Peloponissou & Center for Agricultural Entrepreneurship, Hydroponic Cultivations, Perrotis College, 4 & 5 February, 2014.

Technological Educational Institute of Kalamata & Alegre Grow Shop, Training workshop on hydroponics & aquaponics. Thessaloniki, 26 May, 2013.

Aristotle University of Thessaloniki, Life-Long Learning Program, Training course on "Production of Medicinal and Aromatic Plants (MAP) and Essential Oils". Course duration 103 hours (10 ECTS). August – September 2012.

Australia Felix Permaculture & Permaculture Greece. Athens, Greece, Permaculture Design Course Certificate, 6-21 September 2011.

European Sustainability Academy, Ecological Building Workshop, Crete, 22-36 June 2011.

Euroeducation Ltd. Thessaloniki, Workshop on "Microbiological Water Quality", 29-30 October 2007.

Chemical and Analytical Equipment Company MERCK Inc., Thessaloniki, Workshop on "Accreditation of an Microbiological Analysis Laboratory under EN ISO 19011", 2006.

Luis Bolk Institute, The Netherlands, EU QLIF (QualityLowInputFood), International Workshop on Soil Quality. 2-4 February, 2005.

Scholarships:
National Institute of State Scholarships, Greece. Academic Scholarships for three consecutive years, awarded to the top student of each class, Fall 1980-Spring 1983.

National Institutes of Health, United States Public Health Service. National Research Service Award, September 1984- September 1986.

RESEARCH

Undergraduate Research:
Department of Molecular and Cell Biology, University of California at Berkeley, CA, U.S.A. Research Assistant, 1985-1991

Ministry of Education, Athens, Greece. Internship, Summer 1983. "Ecological Study of the Impacts of Tourism on Agriculture in Skiathos and Skopelos"

Doctoral Research:
Department of Molecular and Cell Biology, University of California at Berkeley, U.S.A.- 1991
"The role of Heat Shock Proteins during initiation of DNA replication"

Academic Research:
Research coordinator and lead scientist from Perrotis College for the Research Program LILAS4SOILS HORIZON Europe, September 2024-2029.

Research project on the "Processing of two-phase olive mill solid waste through vermicomposting".

Perrotis College. Fall 2015-Today.

Research project on the "Evaluation of cover crop mixtures for weed management, soil fertility and carbon sequestration in organic agriculture". Perrotis College. Fall 2015-Today.

Research project on the effect of herbicides on mycorrhizae and soil microbial activity in orchards. Department of Plant Sciences, UC Davis. Fall 2018

Research coordinator and lead scientist, BalkanROAD Project "Towards farms with zero carbon-, waste- and water-footprint. Roadmap for sustainable management strategies for Balkan agricultural sector". INTERREG Balkan-Mediterranean 2018-2020.

Research project on "The role of mycorrhizae on almond production in California" as a visiting professor. Department of Plant Sciences, UC Davis. Spring and Summer 2016

Member of the Leadership Team and Associate Program Manager for the New Agriculture for a New Generation: Recharging Greek Youth to Revitalize the Agriculture and Food Sector of the Greek Economy Project, Stavros Niarchos Foundation's Recharging the Youth initiative, 2015.

Coordinator for three sectoral studies on "Aromatic & Medicinal Plants", "Alternative Fruit Crops for Greece" and "Viticulture, Table & Wine Grapes". Stavros Niarchos Foundation Project. "New Agriculture for a New Generation" Recharging the Youth initiative. December 2015.

Supervising professor for master thesis projects, Summer 2015 & Summer 2016. Master of Science degree Program on "Sustainable Agriculture", Mediterranean Agronomic Institute of Chania (MAICh).

Supervising professor for master thesis projects 2009-2010. Interdepartmental Master Program "Environment – New Technologies", Alexander Technological Educational Institute, Thessaloniki (ATEI) with topics:

1. "Organic waste management (plant residues, clippings and manures)"
2. "Management of biodegradable household wastes – composting with earthworms (vermicomposting)"

Research coordinator for Perrotis College participation in "Συνεργασία (Collaboration) 2011" Application of statE-of-the-art green technology for the development of hiGh Added value cosmeceuticals based on the GrEek flora (ENGAGE). 2013-2015.

Research coordinator and lead scientist, Research Program PAVET-NE-2004, Greek Secretariat for Research and Technology. Fertilization and plant protection methods study for organic orchards in Northern Greece. 2005-2007.

Community Gardening & Ecological Education Project, Dept. of Environmental Science, Policy & Management, University of California at Berkeley, CA, U.S.A. Researcher & Program Coordinator, 2000-2001

Agroecology Laboratory, Dept. of Environmental Science, Policy & Management, University of California at Berkeley, CA, U.S.A. Postdoctoral Research Associate in Agroecology, 1998-2002

S.A.N.E. (Sustainable Agriculture & Nutrition Education) Project, University of California

Cooperative Extension, Berkeley, CA, U.S.A. Designed S.A.N.E Program and served as Research Coordinator, 1998-2000

Urban Farm Cooperative, University of California Cooperative Extension, Berkeley, CA, U.S.A Sustainable Agriculture Researcher & Program Coordinator, 1996-1998

Dept. of Genetics and Plant Breeding, Thessaloniki, Greece. Aristotelian University of Thessaloniki, Postdoctoral Research Associate, 1995-1996 "Genetic Breeding of Sugarbeets"

Plant Physiology Laboratory, Department of Plant Biology, University of California at Berkeley, CA, U.S.A.
Postdoctoral Fellow 1992-1995 "Plant acclimation strategies to irradiance stress"

Melaleuca Inc., San Francisco, U.S.A. Research Consultant on plant derived pharmaceuticals, Fall 1991- Summer 1992.

PRACTICAL APPLICATIONS IN ORGANIC FARMING
Conversion of a conventional olive grove into organic management. Management of the olive grove for the production of organic olive oil. Polygyros, Chalkidiki, Greece. 2003-2013.

Conversion of a conventional farm into organic management. Management of the organic farm for the production of organic field crops. Stavros, Imathia, Greece. 2004- 2012.

BUSINESS EXPERIENCE
aCert - European Organization for Certification S.A. Certification Manager for Organic Agriculture Products. October 2008 – July 2010

Geotechnical Laboratory S.A., - Organic Certification Agency. Research Director and Quality System Manager, 2006-2008

Geotechnical Laboratory S.A., - Organic Certification Agency. Founding member and scientific advisor on Organic Agriculture, 2006

Geotechnical Laboratory - Soil and Water Analysis Laboratory. Research Director 2004-2005.

TEACHING EXPERIENCE
Perrotis College, Division of American Farm School -Thermi, Greece. Associate Professor. September 2020- Today

Perrotis College, Division of American Farm School -Thermi, Greece. Assistant Professor. October 2014- August 2020.

Master of Science degree Program on "Organic Farming", Aristotle University of Thessaloniki & RUDN University, Russia. Teaching the course "Introduction to Organic Farming". Fall 2018

Master of Science degree Program on "Sustainable Agriculture", Mediterranean Agronomic Institute of Chania (MAICh). Visiting professor, teaching one-week intensive course "Agro-ecosystems and population dynamics". Fall 2017 – Today

Master of Science degree Program on "Sustainable Agriculture", Mediterranean Agronomic Institute of Chania (MAICh). Visiting professor, teaching one-week intensive course "Agro-Environmental Impact Assessment & Farm Management". Fall 2010 – 2016

American Farm School, Adult Education Program, Instructor for the course on "Cultivation of endemic plants with floral and aromatic use". Spring 2013 – Today.

Perrotis College , American Farm School - Thessaloniki, Greece. Lecturer. Fall 2011- 2013:
1. Introduction to Agro-Environmental Systems (Theory & Laboratory)
2. Ecological Agriculture (Theory & Laboratory)
3. Plant Physiology (Theory & Laboratory)
4. Environmental Technology & Farm Machinery (Theory & Laboratory)

Plant production Department, Alexander Technological Educational Institute, Thessaloniki (ATEI). Lecturer 2004-2012:
1. Plant Anatomy – Morphology Laboratory
2. Plant Physiology Laboratory
3. Agrotourism

Erasmus Program, Tourism management Department, Alexander Technological Educational Institute, Thessaloniki (ATEI). Lecturer in Environmental Management. 2007 - 2012

European Masters Programme for Rural Animators (EMRA. Tutor-Trainer, Sustainable Agriculture Module. Mediterranean Agronomic Institute of Chania (MAICh), Crete, Greece, Fall 2011

CerOrganic Training of Trainers Summer School. "A School for Future Trainers of Organic Farmers". Leonardo da Vinci Programme. Tutor-Trainer, Mediterranean Agronomic Institute of Chania (MAICh), Crete, Greece, May 2011

Interdepartmental Master Program "Environment – New Technologies" Alexander Technological Educational Institute, Thessaloniki (ATEI). Lecturer in Agricultural and Industrial Pollution – Ecosystems – Management. 2008-2009

University of California at Berkeley, CA, U.S.A. Designed and instructed an international Web-based Agroecology Course in collaboration with the Agroecology Laboratory, (http://www.agroeco.org/agroecology) 2003 – 2008

Department of Earth Sciences, Stanford University, CA, U.S.A. Visiting Scholar in Agroecology, Spring 2002

Dept. of Environmental Science, Policy & Management, Agroecology Laboratory, University of California at Berkeley, CA, U.S.A. Taught the following courses as a Postdoctoral Fellow:
1. Organic Farming Research, 1998 & 1999
2. Agroforestry, 2001 & 2002
3. Agroecology, 2001 & 2002

The Foundation for Deep Ecology, & Agroecology Laboratory, University of California at Berkeley, CA, U.S.A. Designed and constructed a web page for the promotion of Sustainable Agriculture: http://www.agroeco.org/fatalharvest/. Spring 2002

The Foundation for Deep Ecology, & Agroecology Laboratory, University of California at Berkeley, CA, U.S.A. Organized a series of workshops on Sustainable Agriculture in Colleges and Universities in California on the "Ecological Impact of Industrial Agriculture and Agroecologically Based Solutions & Alternatives" 2002

Bay Area Seed Interchange Library & The Ecology Center, Seed Saving Workshop
October 13, 2001

D.E.I. Foundation Courses Biology Instructor, 1995-1996

Department of Athletics, University of California at Berkeley, CA, U.S.A. Instructor in Outdoor Education, 1988-2002

Department of Molecular and Cell Biology, University of California at Berkeley, CA, U.S.A
Teaching Assistant in Molecular Biology, 1987-1988

PUBLICATIONS

Articles in peer reviewed journals

1. Chatzistathis, T., Zoukidis, K., Vasilikiotis, C., Apostolidis, A., Giannakoula, A.E., Bountla, A. and Chatziathanasiadis, A., 2024. Plant-growth-promoting rhizobacteria and arbuscular mycorrhizal fungi may improve soil fertility and the growth, nutrient uptake, and physiological performance of Batavia lettuce (Lactuca sativa L. var. longifolia) plants. *Horticulturae*, *10*(5), p.449. **Impact Factor:** 3.1
2. Tripodi, P., Beretta, M., Peltier, D., Kalfas, I., Vasilikiotis, C., Laidet, A., Briand, G., Aichholz, C., Zollinger, T., Van Treuren, R. and Scaglione, D., 2023. Development and application of Single Primer Enrichment Technology (SPET) SNP assay for population genomics analysis and candidate gene discovery in lettuce. *Frontiers in Plant Science*, *14*, p.1252777. **Impact factor: 4.1**
3. Vasilikiotis, C., Li, M., Schmidt, J.E., Azimi, A., Garcia, J., Volder, A., Lampinen, B. and Gaudin, A.C., 2020. Orchard management practices affect arbuscular mycorrhizal fungal root colonisation of almond. Biological Agriculture & Horticulture, 36(4), pp.230-248. DOI: 10.1080/01448765.2020.1802777. **Impact factor: 1.674**
4. Vasilikiotis, Christos, (2018). "Evaluation of cover crop mixtures for weed management and soil fertility improvement in organic agriculture". MSc Creative Component https://lib.dr.iastate.edu/creativecomponents/111
5. Elissavet G. Ninou, Konstantinos A. Paschalidis, Ioannis G. Mylonas, Christos Vasilikiotis, and Athanasios G. Mavromatis. The effect of genetic variation and nitrogen fertilization on productive characters of Greek oregano. Acta Agriculturae Scandinavica, Section B — Soil & Plant Science Vol. 67 , Iss. 4, 2017 **Impact factor: 1.092**
6. Tertivanidis, K., Goudoula, C., Vasilikiotis, C., Hassiotou, E., Perl-Treves, R., and Tsaftaris, A. (2004). Superoxide dismutase transgenes in sugarbeets confer resistance to oxidative agents and the fungus Cercospora beticola. Transgenic Research 13, 225-233. **Impact factor: 2.19**
7. Vasilikiotis, C., Melis, A. The role of Chloroplast-encoded protein biosynthesis on the rate of D1 protein degradation in *Dunaliella salina*. Photosynthesis Research, v.45, n.2, (1995): 147-155. **Impact factor: 3.574**
8. Vasilikiotis, C., Melis, A. Photosystem II reaction center damage and repair cycle: Chloroplast acclimation strategy to irradiance stress. Proceedings Of The National Academy

Of Sciences Of The United States Of America. 1994. 91(15):7222-7226. **Impact factor: 9.58**

9. Wyman, C., Vasilikiotis, C., Ang, D., Georgopoulos, C., Echols, H. Function of the GrpE heat shock protein in bi-directional unwinding and replication from the origin of phage lambda. Journal Of Biological Chemistry. 1993. 268(33):25192-2519. DOI:10.1016/S0021-9258(19)74587-6 **Impact factor: 4.238**

Book Chapters

Gertsis A.C., Vasilikiotis C. (2018) The LISA and SOCRATEES© Approach for Sustainable Crop and Soil Management. In: Sengar R., Singh A. (eds) Eco-friendly Agro-biological Techniques for Enhancing Crop Productivity. Springer, Singapore

Publications in Conference proceedings

1. Chidiac, A., Roulias, I., Meis, S., Woodruff, J., Efstathiou, A. & Vasilikiotis, C., 2024. Integrating multi-species cover crops with organic no-till practices for enhanced soil health and quality. *Conference on Soil Health: Current Status and Future Needs*, Chania, Greece, 07-09 October 2024.
2. Zoukidis, K., Vasilikiotis, C., Gertsis, A., Scialletti, A., Strouthopoulos, G. & Vergos, E., 2024. Effect of an organo-mineral fertilizer produced by recovered sulphur & orange wastes on winter wheat growth as a sustainable mitigation of soil desertification. *Conference on Soil Health: Current Status and Future Needs*, Chania, Greece, 07-09 October 2024.
3. Karampetian, G., Zoukidis, K., Gertsis, A., Falaras, A., Vasilikiotis, C., Apostolidis, A., Vergos, E. & Tziachris, P., 2024. Estimation of soil organic carbon content using remote sensing and GIS techniques. *Conference on Soil Health: Current Status and Future Needs*, Chania, Greece, 07-09 October 2024.
4. Christos Vasilikiotis, Athanasios Gertsis, Konstantinos Zoukidis and Ali Nasrallah. 2015. Multi-species cover crop biomass evaluation using a hand-held Normalized Difference Vegetation Index (NDVI) sensor and Photosynthetically Active Radiation (PAR) sensor. HAICTA 2015
Proceedings of the 7th International Conference on Information and Communication Technologies in Agriculture, Food and Environment. Kavala, Greece, September 17-20, 2015.
5. Gertsis, A., Konstantinos Zoukidis , and Christos Vasilikiotis. 2015. Evaluation of an irrigation water treatment technology (MAXGROW) on its effects to vegetable species yield. 7th International Conference on Information and Communication Technologies in Agriculture, Food and Environment HAICTA 2015. Kavala, Greece (17-20 Sept. 2015 http://ceur-ws.org/Vol-1498/
6. M. Dajko and C. Vasilikiotis. 2015 Vermicomposting of two-phase olive mill waste (OMW) using the earthworm *Eisenia fetida* with the aim to reduce environmental pollution and produce of a high quality organic fertilizer and soil amendment. Fork to Farm: the International Journal of the American Farm School of Thessaloniki, Vol 2, No. 1, p
7. Dajko, M. A. Gertsis and C. Vasilikiotis. 2014. The effect of two-phase olive mill waste (TP-OMW) on the population growth of the earthworm *Eisenia fetida* and the quality of the produced vermicompost as organic fertilizer and soil amendment. In: Gertsis, A. 2014. (editor) ***Innovative olive production systems adapted for mechanical harvesting: holistic approaches for sustainable management.*** Book of Presentations. International Workshop/Conference, 12-15 Nov. 2014, Thessaloniki, Greece (ISBN 978-618-80868-2-1)

8. Casey Parker, Alex Chaskopoulou, Max Fotakis, Christos Vasilikiotis, Roberto Pereira1 & Philip Koehler, 2014. Activity patterns of *Aedes albopictus* within a diverse environment of residential and agricultural activity and introduction of a new lethal ovitrap for controlling wild vector populations. 19th European SOVE Conference, Thessaloniki, Greece. October 2014.
9. C. Verikoukis, A. Gertsis and C. Vasilikiotis, 2014, The effect of three types of dairy manure used as a feeding stock on the reproduction and growth of Eisenia fetida and on the quality of the produced vermicompost. XLIII Annual Meeting of the European Society for New Methods in Agricultural Research, Bolzano Italy September 2014.
10. D. Vournoukas, C. Vasilikiotis; A. Gertsis, 2014. Evaluating the Yield Potential of Salad Type Lettuce (*Lactuca sativa L.*) Hybrid Seed Offspring. Fork to Farm: the International Journal of the American Farm School of Thessaloniki, Vo1 1, No.1, p 1-6.
11. C. Vasilikiotis. Alternative Sustainable Farming Systems: The Importance Of Education, Training And Extension To Comply With Changing Regulatory Landscape And Consumer Demands In Greece. European Masters Programme for Rural Animators. International Conference. Mediterranean Agronomic Institute of Chania, Greece. July 2012.
12. G. Bardas, C. Vasilikiotis, P. Pantopoulos and K. Tzavela-Klonari. 2006. Disease control in apple and peach orchards with lime-sulfur under organic management. 13th Panhellenic Phytopathological Congress, Hellenic Phytopathological Society. October 2006.
13. Echols, H., Hoffmann, H., Lyman, S., Vasilikiotis, C., Dodson, M., Rajagopalan, M., Kwack, S., Lu, C. Multi-protein interactions initiating lambda DNA replication and replicative bypass of DNA lesions. Journal Of Cellular Biochemistry Suppl. 1992(16 PART B):4.

General Articles

1. Christos Vasilikiotis, 2016. New Farmers-Their contribution to the development of Greek Agriculture (Νέοι Γεωργοί – Η συνεισφορά τους στην ανάπτυξη της ελληνικής γεωργίας). Agricultural Economics Publication (Περιοδική Έκδοση Για Την Αγροτική Οικονομία ΕΠΙ ΓΗΣ), Pireus Bank (Τράπεζα Πειραιώς).
2. C. Vasilikiotis, M. Altieri. Promoting Urban-based Ecological Pest Management: Expanding Outreach in East Bay Schools and Community gardens. (2005)
3. Vasilikiotis, C. Organic Farming. Terrain, Summer 2000, Ecology Center, Berkeley, CA, U.S.A
4. Vasilikiotis, C. Can Organic Farming "feed the world"?. The Natural Farmer, Winter 2000 v.2, n. 47:16-18, Northeast Organic Farming Association, U.S.A.
5. Vasilikiotis, C. Cotton. Ecology Center Terrain, May 1992, Ecology Center, Berkeley, CA, U.S.A

CONFERENCES & TRAINING
A. Presentations & Posters
Conference presentation at the *Conference on Soil Health: Current Status and Future Needs*. Presentation topic: "Integrating multi-species cover crops with organic no-till practices for enhanced soil health and quality". Chania, Greece, 07-09 October 2024.

Conference presentation at the the 2nd International Yale Symposium on Olive oil and Health in Delphi, December 1-4, 2019

Conference presentation at the Agriculture & Food 7th International Conference. Presentation topic: “Evaluation of cover crop mixtures for weed management and soil fertility improvement in organic agriculture”. Burgas, Bulgaria, 25-29 June 2019.

Vasilikiotis, C. Zormpa, D. Conference Poster: GAP Analysis of the Balkan agricultural sector Gaps and barriers - the Greece case. Agriculture & Food 7th International Conference, Burgas, Bulgaria, 25-29 June 2019.

International Conference Climatico 2019: “Climate Change in the Mediterranean: Agriculture, Food and Health Impacts and Challenges.”. Presentation topic: Soil and water management strategies for low-footprint viticulture. Limassol Cyprus, April 11-12, 2019.

Conference presentation at the 1st International Olive Center Conference | Table Olives : Pursuing Innovation - Exploring Trends, 24-26 May 2018, Thessaloniki, Greece. Presentation topic: Organic cultivation and certification of table olives.

Vasilikiotis, C. Vergopoulos, A. and Zoukidis, K. Weed suppression by fall-sown legume and grass cover crop mixtures in organic agriculture. 18th European Weed Research Society Symposium, Ljubljana, Slovenia, June 2018.

Tamara McClung, Christos Vasilikiotis, Bruce Lampinen, Amélie Gaudin & Astrid Volder. Survey and Potential of Mycorrhizal Inoculation to Mitigate Water Stress in Almond. Almond Board of California Conference, December 6–8 2016, Sacramento, CA, U.S.A.

Panagiotis Paliouras, Konstantinos Zoukidis and Christos Vasilikiotis. 2016. Response of legumes to Inoculation with isolated native Rhizobium strains and N fertilization. Effect on crop growth, yield and N uptake. 45th Conference of the European Society for New Methods in Agricultural Research (ESNA). 6th - 8th September 2016, Belgrade, Serbia.

Ali Nasrallah, Konstantinos Zoukidis and Christos Vasilikiotis. 2016. The effect of multi-species cover crop systems on weed suppression and soil fertility in organic agriculture. 45th Conference of the European Society for New Methods in Agricultural Research (ESNA). 6th - 8th September 2016, Belgrade, Serbia.

Christos Vasilikiotis, Athanasios Gertsis, Konstantinos Zoukidis and Ali Nasrallah. 2015. Multi-species cover crop biomass evaluation using a hand-held Normalized Difference Vegetation Index (NDVI) sensor and Photosynthetically Active Radiation (PAR) sensor. HAICTA 2015 7th International Conference on Information and Communication Technologies in Agriculture, Food and Environment 17-20 September 2015, Kavala, Greece.

Dajko, M., Gertsis, A. and Vasilikiotis, C.- 2014 Poster presentation - “The effect of two-phase olive mill waste (TP-OMW) on the population growth of the earthworm *Eisenia fetida* and the quality of the produced vermicompost as organic fertilizer and soil amendment” International Workshop/Conference “Innovative Olive Production Systems Adapted for Mechanical Harvesting: Holistic Approaches for Sustainable Management”, Perrotis College, Thessaloniki, Greece. 12-15 November, 2014

I. Koutsoupias and C. Vasilikiotis. 2014 Evaluation of three cover crop systems for weed suppression and soil fertility. International Conference on Global Trends in the Agro-food Sector, Kalamata, Greece, 11 -13 of September, 2014.

M. Dajiko, A. Gertsis and C. Vasilikiotis. 2014 Vermicomposting of two-phase olive mill waste (TP-OMW) using the earthworm Eisenia fetida with the aim to reduce environmental pollution and produce of a high quality organic fertilizer and soil amendment. International Conference on Global Trends in the Agro-food Sector, Kalamata, Greece, 11 -13 of September, 2014.

Verikoukis Christos, Gertsis Athanasios, Vasilikiotis Christos, 2014. Poster presentation. The effect of three types of dairy manure used as a feeding stock on the reproduction and growth of Eisenia fetida and on the quality of the produced vermicompost. The XLIII Annual Meeting of the European Society for New Methods in Agricultural Research (ESNA). 3rd- 6th September 2014.

C. Vasilikiotis, P. Pantopoulos and I. Therios. Poster presentation. Evaluation of Organic Amendments on Soil Fertility and Nutrient Uptake in Commercial Organic Orchards in Northern Greece. ESNA European Society for New methods in Agricultural research. 4-8 September 2013.

European Masters Programme for Rural Animators. International Conference. Mediterranean Agronomic Institute of Chania, Greece. July 2012.

European Network for the Durable Exploitation of Crop Protection Strategies (ENDURE). Invited specialist on Agroecology for the Regional discussion meeting for European Crop protection in 2030 of the ENDURE Foresight Study – Mediterranean Section. MAICH, Chania, 24-25 September 2009.

International UNESCO conference and Aristotelian University of Thessaloniki. Cultivating Sustainable Lifestyles. 15-17 October, 2004.

E. Anastassopoulos, K. Tertivanidis, C. Goudoula, C. Vasilikiotis, E. Hassiotou, R.Perl-Treves, N.J. Panopoulos, and A.S. Tsaftaris (2002) A microplate freezing assay applied to transgenic sugarbeet transformed with superoxide dismutase transgenes. 13th Congress of the Federation of European Societies of Plant Physiology, Crete, Greece. Book of Abstracts Pg. 707.

Third Western Regional Photosynthesis Conference, Pacific Grove, California, U.S.A, January 4 - 7, 1994.

Second Western Regional Photosynthesis Conference, Pacific Grove, California, U.S.A, January 12-15, 1993.

Molecular Mechanisms in DNA Replication and Recombination, UCLA Symposia, U.S.A, March 27-April 3 1989.

Molecular Genetics of Bacteria and their Phages: Prokaryotic gene regulation, Cold Spring Harbor Laboratory, U.S.A, Aug 18-23, 1987

Mechanisms of DNA Replication and Recombination, UCLA Symposia, U.S.A, March 16-23, 1986

Molecular Genetics of Bacteria and Phages, Cold Spring Harbor Laboratory, U.S.A, Aug 20-25, 1985

B. Organizing & Presentations in Workshops & Public Events

Organizer and instructor of the Regenerative Agriculture Workshop. EIT Food, Co-funded by the

European Union. Perrotis College, Thessaloniki, 28 June, 2024.

Workshop on the EU Green Deal and the new Common Agricultural Policy for producers in Messinia, in collaboration with the Center for AgroFood Entrepreneurship of Messinia. 2 February 2023

Invited panelist for the Leaders Debate: “Synthetic Biology or Regenerative Agriculture?” at the Global Agripreneurs Summit, September 7-11, 2019

Trainer-Lecturer on 1st Mediterranean Summer school on Sustainable Development and Bioeconomy. Perrotis College, 9-12 July, 2019

Invited speaker at a workshop organized by the Mediterranean Agronomic Institute, Chania (MAICH), on Ecology, Sustainable Agriculture and Organic Farming. Chania, Crete, Greece, June 8, 2019.

Invited speaker at the “Agrocircle: The implementation of circular economy to agriculture and livestock farming”. Zootechnia Conference, Thessaloniki, Greece, 31 January-1 February 2019.

Invited speaker at the Circular Economy in Agriculture Conference, with a presentation on “Food and agricultural waste recycling – Closing the nutrient cycling loop through composting”. Thessaloniki, Greece, 30 January 2019.

Invited speaker to Evergreen State College in Washington State to present a Lecture on “Cover Crops as a tool for Climate Change Mitigation”. Olympia, Washington, USA. Fall 2018

Presented a series of Adult Education seminars on Aromatic and Medicinal Plant Production in Gjirokastër, Albania, 2-4 February 2018

Trainer-Lecturer on the Summer school on Integrated Weed Management: Principles and implementation, organized by the European Weed Research Society, the Greek Weed Science Society and the American Farm School (AFS)/Perottis College. Thessaloniki, Greece 22-25 June 2017

Moderator and presenter at the “Entrepreneurship and Employability in Agriculture; Utilizing the prospects in the area of aromatic plants and herbs, in production, processing and marketing”. American Farm School, Perrotis College, and DIO. Thessaloniki, Jan 14, 2016

Moderator and presenter at the Lecture series “Cultivating our cities: Creation of Green Spaces” in Thessaloniki and Athens organized by the Association of Friends and the American Farm School. 28-29 April, 2015.

Moderator and presenter at the Food Taste & Society 2015 “Gastronomic Greece: a fusion of ideas people and ingredients” American Farm School, Thessaloniki. 8-10 May, 2015.

C. Participation in Scientific Conferences

“1 Decembrie 1918” University, Alba Iulia, Romania, International Conference “Environmental Engineering And Sustainable Development”, 26-27 May 2011.

BENA Conference «Balkan Collaboration: the must for better life and environment in S.E. Europe», Florina, 6-9 November 2008.

2nd AGROTICA Conference on "Climate Change and Agriculture". Ministry of Agriculture & Aristotelian University of Thessaloniki, Agronomy Dept. 8-9 February, 2008.

The Colloquium of Organic Researchers (COR) What will organic farming deliver? Heriot-Watt University, Edinburgh, 18-20 September 2006.

Organic Farming Workgroup, Berkeley, California, USA, 15 July, 2004

California Conference on Biological Control, Berkeley, California, USA, 13-14 July, 2004

Ecological Farming Conference, Asilomar Conference Center,
Pacific Grove, California, U.S.A: 1999, 2000, 2001, 2002 and 2003

Short Curriculum Vitae

Name: **Athanasios C. Gertsis** (or **Gkertsis** based on latest EU name regulations)
Work address: Marinou Antypa 54, str, Perrotis College, American Farm School, P.O. Box 60097, GR 551 02, Thermi, Thessaloniki, Greece
Work tel: +30-2310-492 816 Fax: +30-2310-492815 Mobile phone: +30-6947 370 490
E-mail address: ***agerts@afs.edu.gr***
www.perrotiscollege.edu.gr

HIGHER EDUCATION:

Diploma, 1980. Higher School of Agricultural Technology (Crop Production curriculum, (KATEE- currently TEI)-), Thessaloniki, GREECE.

B.Sc. degree in Agronomy (Crop Science), 1982. Mississippi State University, Miss. State, MS, USA.

Certificate of Highest School of Pedagogical and Technological Education (***ASPAITE***-*former SELETE*), Thessaloniki, Greece, Sept. 2003 (*Certified teacher*).

POST-GRADUATE EDUCATION:

M. Sc. degree in Agronomy (Plant Physiology & Soil Science), 1985, Mississippi State University (MSU), Miss. State, MS, U.S.A. M.Sc. Thesis title: Validation of GLYCIM: a dynamic simulator of soybeans (*Glycine max* (L.) Merrill).

Ph. D. degree in Agronomy (Crop Physiology and Soil Science with emphasis on: Soil-plant-water relations and Agronomic crop simulation & modelling), 1992, Texas Tech University (TTU), Lubbock, TX- U.S.A. Ph.D. Dissertation title: Cotton (*Gossypium hirsutum* L.) plant water status as a function of water supply and evaporative demand in a semiarid environment.

Post-Doctoral: (Expert scientist- Scientific collaborator, 1994-1997) at the University of Thessaly, Volos, Greece, in a research project funded by European Union (Project AIR3-CT93-0936: *Evaluation and application of GOSSYM, a cotton crop simulation model, under the soil, climatic and cultural conditions of Greece and Spain*).

OTHER EDUCATION & NATIONAL CERTIFICATIONS

- Special training in agrochemicals at the NPTN (National Pesticide Telecommunications Network) as a Research Assistant, in the Medical School of Texas Tech University. Lubbock, TX, USA. 1987
- National Accreditation Centre for Continuing Vocational Training (EKEPIS-www.ekepis.gr), Greece, 2001 (*Accreditation of Trainers of Continuing Vocational Training) updated 2008*
- OECD Research Fellowship, at University of Georgia, Tifton, GA-USA, 2005.
- Quality assurance certification in Agro 2.1., 2.2 and EUREPGAP systems

ADDITIONAL SKILLS AND FOREIGN LANGUAGES:

Certificate of good knowledge of French language (*Certificat de bonne connaissance de la langue Française).* Obtained in 16-7-1979)

Excellent knowledge of computer operating systems (Windows and Apple Macintosh) and software applications **ECDL certification** (Word processing, databases, spreadsheets, graphics, statistical software (SPSS, JMP), presentation systems, Geographic Information Systems (Q.G.I.S.), Network communications, data acquisition systems and computer interfaces, crop simulation models etc.)

Scientific publications
In English (92) - In Greek (35) - Books & Chapters (6)
BSc and MSc Theses supervised: (In Greek State Universities and Perrotis College)
Approximately 146 as of early 2024

PROFESSIONAL ORGANIZATION MEMBERSHIPS:

- Agronomy Society of America (ASA) *formerly*
- International Soil Science Society (ISSS) *formerly*
- Crop Science Society of America (CSSA) *formerly*
- Soil Conservation Society of America (SCSA) *formerly*
- European Society of Soil Conservation (ESSC)
- Hellenic Chamber of Agriculturists (GEOTEE)
- Hellenic Scientific Society of Plant Breeding and Genetics (EEEGBF)
- Hellenic Soil Science Society (EEE)
- European Federation of Information Technology in Agriculture (EFITA) and local branch (HAICTA –Hellenic Association of Information and Communication Technology in Agriculture, Food and Industry)
- European Society for New Methods in Agricultural Research (ESNA)
- **International Society of Precision Agriculture (ISPA)**

Co-Supervision of International M.Sc. degree students (EU and USA)
1. University of Aberdeen, Scotland, UK
2. University of Georgia, Athens, USA
3. Wageningen UR, The Netherlands
4. University of Padua , Italy (DAFNAE)
5. International Hellenic University , Thessaloniki, GREECE

FUNDED RESEARCH PROJECTS AND OTHER ACADEMIC ACTIVITIES

- **Scientific Leader** –Post Doctoral (1994-1997) (Project AIR3-CT93-0936: *Evaluation and application of GOSSYM, a cotton crop simulation model, under the soil, climatic and cultural conditions of Greece and Spain*) funded by EOK –EU d429 ECUS=€) University of Thessaly, TEI Thessaloniki and SPAIN (cotton research Institute at Seville)
- **National participant & member of the Steering Committee** in the NUMALEC project (1999-2001): *Nutrient Management Legislation in European Countries*), FAIR concerted action , funded by EU (256.000 ECUS=€)
- **Project coordinator** (2000-2002) of the project: *Evaluation of sesame production under low input systems friendly to the environment*) funded by a private Ag Business HAITOGLOU Bros (6,000,000 G ~reek Drachmas 20.000 €)
- **Scientific Leader and Project Coordinator (2004-2007):** *Evaluation of zeolite tuffs, vermiculite, fly and bottom ash as soil improving materials*. Funded by European Union – Hellenic Ministry of Development and Institute of Geological and Mineralogical Research -3rd Framework Program
- **Scientific Leader and Project Manager (2006-2008):** *Digital Precision Agriculture*. funded by the Funded by European Union – Hellenic Ministry of Development, and the General Secretariat of Research and Development of Greece in the context of the Regional Poles of Innovations for Central Macedonia (269,000 €)

- **Project and Scientific Manager (2007-2010)** of the project: *Evaluation of residual paper pulp as a soil improving material*. Funded by a private Paper Mill Business (15,000 €)
- **Project and Scientific Manager (2015-2020) SANOVITA products** evaluation studies (25,000 €)
- **Project and Scientific Manager (2015-2020) BIOSOLIDS** - evaluation of organic soil improving materials (25.000 €)
- **Project and Scientific Manager (2012-2020)** Evaluation of **MAXGROW** electronic water treatment device (15.000€)
- **ERASMUS+ project partner (SMART FARMING, 2017-2020 & 2020-2023).** **https://www.isa-lille.com/international-programs/erasmus/smart-farming/** **(268.000,00** €) and https://erasmusplus-agreensmart.eu/

Additional academic & Administrative activities

- Associate Editor (from 2003 to 2014). RENEWABLE AGRICULTURE AND FOOD
- SYSTEMS (*formerly known as AMERICAN JOURNAL OF ALTERNATIVE AGRICULTURE*). John W. Doran (Editor-in-Chief)
- Journal reviewer: The Pedosphere
- Director, The KRINOS OLIVE CENTER-Perrotis College, AFS
- Quality Auditor – for GAB Hellas studies (a partner of Eurofins Agroscience Greece) https://www.eurofins.com/contact-us/worldwide-interactive-map/greece/eurofins-agroscience-greece/
- Chair of MSc program in Sustainable Agriculture & Management, Perrotis College

Special awards

Awarded by HE The President of the Hellenic Republic, Mr. Carolos Papoulias for the contribution in innovative technology transfer in the Greek Agriculture (October 2013)

Curriculum Vitae

Nicholas C. Manoukis

Contact Information

United States Department of Agriculture - Agricultural Research Service
European Biological Control Laboratory (EBCL)
810 avenue du Campus Agropolis 34980 Montferrier-sur-Lez FRANCE

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Email	nicholas.manoukis@usda.gov
Homepage	http://unitsci.org/manoukis.php
ORCID	0000-0001-5062-7256

Education

PhD	University of California, Los Angeles (Los Angeles, CA). Ecology and Evolutionary Biology, Dec 2006.
BA	Reed College (Portland, OR). Biology, Jan 1997.

Positions Held

Agricultural Research Service (USDA)

2024 – pres.	**Laboratory Director (GS-15)**	Lead USDA-ARS European Biological Control Laboratory (EBCL) near Montpellier, France and Thessaloniki, Greece. Manage laboratory budget and six scientists, plus technicians and staff. Conduct personal research on biological control, ensure administrative efficiency and financial stability of the laboratory while connecting with stakeholders in the US and around the world.
2016 – 2024	**Supervisory Res. Biologist/ Research Leader (GS-14, -15)**	Provide research and administrative leadership to the Tropical Crop and Commodity Protection Research Unit (Hilo, Hawaii), comprised of ten scientists plus staff. Manage unit budget and act as a research liaison to state, federal, university and other stakeholders. Conduct personal research on Tephritid fruit fly and other invasive pest biology and control.
2010 – 2016	**Research Biologist (GS-12, -13)**	Designed and implemented a research program on Tephritid fruit fly biology and control. Planed and executed research, supervised up to six employees and published impactful research results.

National Institutes of Health (NIH)

2007 – 2010	**Post-doctoral Fellow**	Researched *An. gambiae* ecology and evolution in the laboratory of Dr. José M.C. Ribeiro. Focused on mosquito sexual behavior and dry season ecology.

University of California, Los Angeles (UCLA)

2001 – 2006	**Graduate Student Researcher**	Conducted research under the supervision of Dr. Charles E. Taylor in mosquito ecology, evolution and malaria epidemiology.
2003 – 2004	**Systems and Integrative Biology Fellow**	Awarded NIH-funded fellowship to pursue courses in mathematical modeling and theory.

Long Beach Unified School District

1998 – 2000	**Teacher**	Provided instruction on all subject areas to Fifth graders who were English language learners (Garfield Elementary, Long Beach Unified School District).

University of Southern California

1997 – 1998	**Laboratory Assistant**	Prepared laboratories for introductory Life Science course.

Reed College

1995 – 1997	**Head Lab Assistant**	Maintained an amphibian colony, aided with experiments and assisted with training in the laboratory of Dr. Robert H. Kaplan.

Grants, Awards, and Memberships

2022-2024	Awarded $124,500 by USDA-APHIS for project "Advancing Male Annihilation Technique for Invasive Cuelure-Responding Fruit Flies."
2021-2022	Awarded $25,000 ARS "Innovation Fund" (Round 14).
2019-2021	Awarded $75,000 by California Dept of Food and Agriculture (CDFA) to support research on Male Annihilation Technique.
2016-2018	Awarded $140,000 ARS Administrator's Research Associate to support modeling (class of 2016).
2016-2017	Received $110,000 to support postdoctoral researcher to implement Coffee Berry Borer Area-Wide Research.
2016-2018	Cooperator on $170,000 Farm Bill section 10007 project to model Medfly risk in continental US.
2014-2015	Awarded $80,000 USDA grant to create GIS-based spatial model of Coffee Berry Borer on Hawaii Island.
2013-2015	Co-PI on $230,000 USDA-APHIS grant to support continuing work with Agent Based Simulation.
2013-2014	Awarded $13,000 contract to model increased efficiency trapping networks and parametrize model with field work by IAEA/FAO.
2022 – present	Sigma Xi, The Scientfic Research Honor Society, member.
2012–present	Hawaiian Entomological Society, member.
2011–present	Entomological Society of America, member.

2001–2021	American Association for the Advancement of Science, member.
2003–2004	Recipient of Systems and Integrative Biology Training Grant (NIH).
2000–2001	Recipient of California Genetic Resources Conservation Program Grant.
1995	Recipient of Reed College Independent Fieldwork Grant.

Invitations, Appointments, and Professional Service

Chief scientific investigator (agreement holder) on International Atomic Energy Agency Research Coordination Meeting (RCM) on improving Sterile Insect Technique (2021 - 2026).

President of the Hawaiian Entomological Society (2021 - 2022).

Nominated and selected to be a member of International Atomic Energy Agency/Food and Agriculture (IAEA/FAO) International Fruit Fly Steering Committee (2018-present).

Affiliate Faculty, Tropical Conservation Biology and Environmental Studies Program at the University of Hawaii, Hilo (2016-present).

Editorial board member, *Journal of Insect Behavior* (2018-present).

Editorial board member (Ecology and Evolution) *Scientific Reports* (2015-2017).

Member of Technical Advisory Group on tephritid trapping (New Zealand Fruit Fly Council; 2017-2018).

Member of the Entomological Society of America Diversity and Inclusion Committee (2015-2017).

Invited to National Taiwan University as featured speaker on trapping tephritids, Taipei, November 2016 (funding provided by host).

Participant in International Atomic Energy Agency Research Coordination Meeting (RCM) on Male mosquito biology and behavior in Petrolina, Brazil, March 2013 (funding provided by host).

Invited to participate in Degree-day modeling workshop at USDA-APHIS, Raleigh, NC, March 2011 (funding provided by host).

Invited to present "Adaption and diversification in *Anopheles gambiae*, and their implications for malaria transmission", special seminar at the Smithsonian National Zoo, Washington, DC, 2009.

Presented "Population size and migratory patterns of Anopheles gambiae in the Bancoumana region of Mali and their significance for efficient vector control." at the Fundação Luso-Americana (FLAD) Workshop on Malaria, Lisbon, Portugal, July 2009 (funding provided by host).

California State Science Fair Judge (1998–2002).

Reviewer for *Science*, *Theoretical Population Biology*, *J. of Economic Entomology*, *Acta Tropica*, *J. of Vector Ecology PLoS Neglected Tropical Diseases*, *Entomologia Experimentalis et Applicata*, *Insects*, *Physiological Entomology*, *Bioinformatics*, *PLoS ONE*, *Conservation Genetics Resources* and others, updated list see: `https://www.webofscience.com/wos/author/rid/V-9063-2019`

Teaching Experience

Spring 2013	Taught a graduate-level class titled "Computer Modeling and Simulation" in the Tropical Conservation, Biology and Environmental Science Department of the University of Hawaii, Hilo.
Fall 2012	Co-instructor of a one week bioinformatics workshop at University of Hawaii, Hilo.
Fall 2009	Taught a course titled "Introduction to Dynamic Biological System Modeling" in the Mathematics department of the Foundation for Advanced Education in the Sciences, National Institutes of Health, Bethesda Maryland.
2000-2005	Teaching assistant for various courses, including Introductory Biology, Evolutionary Biology and Biological Computation in the Organismic Biology, Ecology and Evolution Department at University of California, Los Angeles.
1998–2000	Fifth grade teacher at Garfield Elementary, Long Beach Unified School District, Long Beach, California.
1995 – 1997	Student teacher and first coordinator of Reed College Biology Outreach Program, Portland Oregon.

Field and Laboratory Work

- Conducting annual three-month sabbaticals at the ARS European Biocontrol Laboratory (EBCL) in Montpellier, France, to study parasitoids of *Bactrocera oleae* in quarantine and in field (2019-2023).
- Managed ecological surveys on the invasive Coffee Berry Borer (*Hypothenemus hampei*) on Hawaii Island (2016 – 2017).
- Led Mark-Release-Recapture experiments with fruit flies in Hawaii Island in tropical costal areas and arid lava flats (2011 – 2016).
- Conducted field work in Bamako and Niono, Mali (2002 – 2010).
- Proficient in population genetic molecular lab technique.
- Completed a demographic survey of threespine sticklebacks (*Gasterosteus aculeatus*) in Reed Canyon (1995).
- Conducted survey of the nectivorous bat *Glossophaga sorcina* in Rio de Janeiro, Brazil (1992).
- Fluent in Portuguese; Proficient in French and Spanish.

Computation Experience

- UNIX and GNU/Linux operating systems. Proficient programmer in Java, Python and R.
- Agent-based and matrix modeling.
- Computer Vision (CV).
- High Performance Computing (HPC) system design and administration. Was system admininstrator for HPC cluster moana.
- Data and metadata archiving and curation.
- GIS and spatial modeling.

Selected Presentations

[1] N. C. Manoukis. New technologies and approaches for controlling invasive *Bactrocera*: How we might build on our successes and address important gaps. Keynote presentation at 10^{th} Tephritid Workers of the Western Hemisphere Conference, Bogota, Colombia, 2020.

[2] N. C. Manoukis. New App for CBB Monitoring and Control: Best Beans. Coffee Berry Borer Area-Wide Program "Virtual Talk Story" (University of Hawaii) 16 April 2020.

[3] N. C. Manoukis. Climate and Mediterranean Fruit Fly Invasion Persistence: Insights from Agent-Based Simulations. Oral Presentation at Third FAO-IAEA International Conference on Area-wide Management of Insect Pests: Integrating the Sterile Insect and Related Nuclear and Other Techniques, Vienna, Austria, 2017.

[4] N. C. Manoukis, B. Hall and S. M. Geib. A Computer Model of Attractant-Based Fruit Fly Traps and its Utility for Optimizing Trapping Networks. Poster Presentation at 9^{th} International Symposium on Fruit Flies of Economic Importance, Bangkok, Thailand 2014.

[5] N. C. Manoukis. Effect of host *Bactrocera dorsalis* fruit fly sex on the parasitoid *Fopius arisanus*. Poster Presentation given at the Entomology 2013, Annual meeting of the Entomological Society of America, Austin TX.

[6] N. C. Manoukis. An Agent-Based Simulation of Tephritid Fruit Flies: Time to Extirpation of *Ceratitis capitata*. Oral Presentation given at the 8^{th} Meeting of the Tephritid Workers of the Western Hamisphere, Panama City, Panama 2012.

[7] N. C. Manoukis, S. Butail, D. A. Paley, A. S. Yaro, M. Diallo, S. F. Traoré, A. Dao, T. Lehmann and J. M. C. Ribeiro. Quantifying and analyzing the mosquito dance in mating swarms. Oral presentation given at the 2011 Entomological Society of America Pacific Branch Meeting, Waikaloa HI 2011.

[8] N. C. Manoukis, Y. Lee and C. E. Taylor. Detecting recurrent extinction in a metapopulation of *Anopheles gambiae*. Oral presentation given at the California Population and Evolutionary Genetics

Group (CalPEG), San Diego CA, 2004.

[9] N. C. Manoukis, M. A. Diuk-Wasser, M. B. Touré, G. Dolo, S. F. Traoré and C. E. Taylor. Malaria transmission and intraspecific competition in *Anopheles gambiae* at an irrigation project in Mali. Oral presentation given at 53^{th} Annual Meeting of the America Society of Tropical Medicine and Hygiene, Miami FL, 2004.

[10] N. C. Manoukis and C. E. Taylor. Effective Population Size of *Anopheles gambiae* s.s. in Mali: Some Implications for Malaria Control Through Genetically Modified Vectors. Oral presentation given at the Molecular and Population Biology of Mosquitoes Workshop (EMBO), Kolymbari, Greece, 2003.

Technical papers and other publications

B. Scalero and **N.C. Manoukis**. TGL-Lambda: An implementation of TrapGrid to estimate trap attractiveness from heterogeneous field data. arXiv:2408.05408 [q-bio.PE]. August 2024.

L. Stringer, R.C. Butler, **N.C. Manoukis**, T. Rahman, C. Weldon, S. Ndlela and M. Cristofaro. Combining lures for Mediterranean fruit fly surveillance. New Zealand Ministry of Primary Industries PFR SPTS No. 25478, May 2024.

A.N. Auclair, A. Perez de Leon, P.D. Teel, **N. C Manoukis**, M.T. Messenger and D.L. Bonilla. Prediction of Cattle Fever Tick Outbreaks in United States Quarantine Zone. Ag Data Commons https://doi.org/10.15482/USDA.ADC
December 2021.

N. C. Manoukis and M.P. Hill. Probability of Insect Capture in a Trap Network: Low Prevalence and Detection Trapping with TrapGrid. arXiv:2110.11432 [q-bio.QM], October 2021.

N. C. Manoukis. Drivers of Mosquito Mating. *Science*, 371:340 - 341, 2021.

B. P. Caton, H. Fang, **N. C. Manoukis** and G. R. Pallipparambil. How Effective is the 5-Mile-by-5-Mile Grid for Insect Trapping? A Simulation-based Investigation Plant Protection and Quarantine (PPQ) report, Animal and Plant Health Inspection Service, U.S. Department of Agriculture. Fall 2019.

J. D. Stark, L. Leblanc, R. F. L. Mau and **N. C. Manoukis**. In Memoriam: Roger Irvin Vargas (1947-2018). Proceedings of the Hawaiian Entomological Society v 50, 2018; Also published in American Entomologist, Spring 2019.

N. C. Manoukis. Description of a method for localizing swarming mosquitoes and other insects in 3d space with visualizations. Available from Nature Precedings, December 2008.

N. C Manoukis and E. C. Anderson. Guiliner: A configurable and extensible graphical user interface for scientific analysis and simulation software. arXiv:0806.0314v1 [cs.HC], June 2008.

N. C. Manoukis and D. K. Jacobs. Conservation of the California tree frog, *Hyla cadaverina*, from desert oasis areas in Joshua Tree National Park. Technical report, Genetic Resources Conservation Program, U.C. Davis, 2001.

Books and book chapters

N. C. Manoukis. Quantifying Insect Trap Network Captures Using TrapGrid *In:* Advances in Monitoring of Native and Invasive Insect Pests of Crops. M. Fountain and T. Pope (eds.), Burleigh Dodds Science Publishing, Cambridge UK, 2023.

N. C. Manoukis, A. Malavasi and R. Pereiria. Técnica de Aniquilção de Machos *In:* Moscas-das-frutas no Brasil - Conhecimentos Básicos e Métodos de Controle (Vol I). R.A. Zucchi (ed.), Fundação de Estudos Agrarios Luis de Queiroz, São Paulo, 2022.

N. C. Manoukis and T.C. Collier. Agent-based Simulations to Determine Mediterranean Fruit Fly Declaration of Eradication Following Outbreaks: Concepts and Practical Examples. *In:* Area-wide Integrated Pest Management: Development and Field Application. J. Hendrichs, R. Pereira, and M. J. B. Vreysen (eds.), CRC Press, Boca Raton Florida, USA, 2021.

Y. Itô, K. Yamamura and **N. C. Manoukis**. Role of Population and Behavioral Ecology in the Sterile Insect Technique. *In:* Sterile Insect Technique: Principles and Practice in Area-Wide Integrated Pest Management. 2^{nd} Edition, V. A. Dyck, J. Hendrichs and A. S. Robinson (eds.). CRC Press, Boca Raton Florida, USA, 2021.

R. I. Vargas and J. C. Pinero and L. Leblanc and **N. C. Manoukis** and R. F. L. Mau. Area-Wide Management of Fruit Flies in Hawaii *In:* Fruit Fly Research and Development in Africa - Towards a Sustainable Management Strategy to Improve Horticulture, S. Ekesi, S. Mohamed, and M. Meyer (eds.), Springer International, Switzerland, 2016.

H. J. Barclay, W. Enkerlin, J. Reyes-Flores and **N. C. Manoukis**. Guidelines for the Use of Mathematics in Operational Area-Wide Integrated Pest Management Programmes Using the Sterile Insect Technique with a Special Focus on Tephritid Fruit Flies IAEA, Vienna, Austria, 2013.

C. E. Taylor and **N. C. Manoukis**. Effective Population Size in Relation to Genetic Modification of *Anopheles gambiae* s.s. *In:* Ecological Aspects for Application of Genetically Modified Mosquitoes, W. Takken and TW Scott (eds.), Kluwer Dordrecht, The Netetherlands, 2003.

Peer-reviewed publications

[1] A. M. Welty Peachy, E. R. Moses, A. J. Johnson, M. M. Lehman, J. M. Yoder, S. G. De Faveri, J. Cheesman, **N. C. Manoukis**, and M. A. Siderhurst. Wind effects on individual male and female *Bactrocera jarvisi* (Diptera: Tephritidae) tracked using harmonic radar. *Environmental Entomology*, -:In Press, 2024.

[2] A. L. Hurst, A. L. O'Brien, N. D. Miller, A. M. Walty Peachy, J. M. Yoder, S. G. De Faveri, J. Cheesman, **N. C. Manoukis**, and M. A. Siderhurst. Tracking and modelingthe movement of Queensland fruit flies, *Bactrocera tryoni*, using harmonic radar in papaya fields. *Scientific Reports*, 14:17521, 2024.

[3] M. A. Johnson and **N. C. Manoukis**. Coffee berry borer (coleoptera: Scolytidae) population dynamics across Hawaii Island's diverse coffee-growing landscape: Optimizing location-specific pesticide applications. *Jounral of Economic Entomology*, 117:963 – 972, 2024.

[4] T. Fezza, T. E. Shelly, A. Fox, K. Beucke, C. Aldebron, and **N. C. Manoukis**. Less is more: Fewer attract-and-kill sites improve the male annihilation technique against *Bactrocera dorsalis* (Diptera: Tephritidae). *PLoS ONE*, 19:e0300866, 2024.

[5] J. Kean, **N. C. Manoukis**, and B. Dominiak. Review of surveillance systems for tephritid fruit fly threats in Australia, New Zealand, and the United States. *Jounral of Economic Entomology*, 117:8 – 23, 2024.

[6] D. G. Stockton, C. Aldebron, R. Gutierrez-Coarite, and **N. C. Manoukis**. Previously introduced braconid parasitoids target recent olive fruit fly (*Bactrocera oleae*) invaders in Hawaii. *Scientific Reports*, 13:22559, 2023.

[7] R. van Kinken, D. W. Gladdish, **N. C. Manoukis**, P. Caley, and M. P. Hill. Simulation to investigate site-based monitoring of pest insect species for trade. *Jounral of Economic Entomology*, 116:1296 – 1306, 2023.

[8] A. L. Roda, G. Steck, T. Fezza, T. Shelly, R. Duncan, **N. C. Manoukis**, L. A. Carvalho, P. Kendra, and D. Carillo. Sieving fruit pulp to detect immature tephritid fruit flies in the field. *Journal of Visualized Experiments*, -:e65501, 2023.

[9] B. P. Caton, H. Fang, G. R. Palipparambil, and **N. C. Manoukis**. Transect-based trapping for area-wide delimitation of insects. *Jounral of Economic Entomology*, 116:1002–1016, 2023.

[10] P. Liang, N. C. Ladizinsky, G. Asmus, L. J. Hamilton, A. L. Acebes-Doria, **N. C. Manoukis**, and P. A. Follett. Artificial fruits and nuts for studying predation of cryptic prey: A case of 3d-printed coffee berries for studying predation of coffee berry borer by flat bark beetles. *Entomologia Experimentalis et Applicata*, 171:716 – 720, 2023.

[11] **N. C. Manoukis**, J. Leathers, K. Beucke, and L. A. Carvalho. Jackson trap efficiency capturing *Bactrocera dorsalis* and *Zeugodacus cucurbitae* with male lures with and without insecticides. *Journal of Applied Entomology*, 147:231 – 238, 2023.

[12] N. D. Miller, T. J. Yoder, **N. C. Manoukis**, L. A. Carvalho, and M. S. Siderhurst. Harmonic radar tracking of individual melon flies, *Zeugodacus cucurbitae*, in Hawaii: Determining movement parameters in cage and field settings. *PLoS ONE*, 17:e0276987, 2022.

[13] D. G. Stockton and **N. C. Manoukis**. The comparative efficacy of a spider knottin insecticide, gs-omega/kappa-hxtx-hv1a, against four species of invasive tephritid fruit flies. *Journal of Applied Entomology*, 146(10):1311 – 1319, 2022.

[14] H. Fang, B. P. Caton, **N. C. Manoukis**, and G. R. Palipparambil. Simulation-based evaluation of two insect trapping grids for delimitation surveys. *Scientific Reports*, 12:11089, 2022.

[15] G. Desurmont, M. Tannières, M. Roche, A. Blanchet, and **N. C. Manoukis**. Identifying an optimal screen mesh to enable augmentorium-based enhanced biological control of the olive fruit fly *Bactrocera oleae* (diptera: Tephritidae) and the mediterranean fruit fly *Ceratitis capitata* (diptera: Tephritidae). *Journal of Insect Science*, 22(11):1 – 7, 2022.

[16] T. Shelly and **N. C. Manoukis**. Mating competitiveness of *Bactrocera dorsalis* (diptera: Tephritidae) males from a genetic sexing strain: Effects of overflooding ratio and released females. *Journal of Economic Entomology*, 115:799 – 807, 2022.

[17] S. B. Sim, K. M. Cubelo, **N. C. Manoukis**, and D. H. Cha. Evaluating *Bactrocera dorsalis* (hendel)(diptera: Tephritidae) response to methyl eugenol: Comparison of three common bioassay methods. *Journal of Economic Entomology*, 115:556 – 564, 2022.

[18] M. Johnson and **N. C. Manoukis**. Influence of seasonal and climatic variables on coffee berry borer (*Hypothenemus hampei* ferrari) flight activity in hawaii. *PLoS ONE*, 16:e0257861, 2021.

[19] B. P. Caton, H. Fang, **N. C. Manoukis**, and G. R. Palipparambil. Quantifying insect dispersal distances from trapping detections data to predict delimiting survey radii. *Journal of Applied Entomology*, 146:203 – 216, 2021.

[20] B. P. Caton, H. Fang, **N. C. Manoukis**, and G. R. Palipparambil. Simulation-based investigation of the performance of delimiting trapping surveys for insect pests. *Journal of Economic Entomology*, 114:2581–25, 2021.

[21] J. Hsu, M. Chou, R. Mau, C. Maeda, I. Shikano, **N. C. Manoukis**, and R. Vargas. Spinosad resistance in field populations of melon fly, *Zeugodacus cucurbitae* (Coquillett), in Hawaii. *Pest Management Science*, 77:5439 – 5444, 2021.

[22] L. Feugère, G. Gibson, **N. C. Manoukis**, and O. Roux. Mosquito sound communication: are male swarms loud enough to attract females? *Journal of the Royal Society Interface*, 18:20210121, 2021.

[23] B. Paranhos, S. Poncio, R. Morelli, D. Nava, L. Nogueira de Sá, and **N. C. Manoukis**. Non-target effects of the exotic generalist parasitoid wasp *Fopius arisanus* (Sonan) estimated via competition assays against *Doryctobracon areolatus* (Szepligeti) on both native and exotic fruit fly hosts. *BioControl*, 66:83–96, 2021.

[24] M. Johnson, C. Ruiz-Diaz, **N. C. Manoukis**, and J. Verle Rodrigues. Coffee berry borer (*Hypothenemus hampei*), a global pest of coffee: Perspectives from historical and recent invasions, and future priorities. *Insects*, 11:882, 2020.

[25] **N. C. Manoukis** and L. Carvalho. Flight burst duration as an indicator of flight ability and physical fitness in two species of Tephritid fruit flies. *Journal of Insect Science*, 20(5):11, 2020.

[26] M. Johnson and **N. C. Manoukis**. Abundance of coffee berry borer in feral, abandoned and managed coffee on Hawaii island. *Journal of Applied Entomology*, 144(10):920–928, 2020.

[27] M. Johnson, S. Fortna, and **N. C. Manoukis**. Evaluation of exclusion netting for coffee berry borer (*Hypothenemus hampei*) management. *Insects*, 11(6):364, 2020.

[28] A. M. Szyniszewska, N. C. Leppla, **N. C. Manoukis**, T. C. Collier, J. M. Hastings, D. J. Kriticos, and K. M. Bigsby. CLIMEX and MED-FOES models for predicting the variability in growth potential and persistence of Mediterranean fruit fly (Diptera: Tephritidae) populations. *Annals of the Entomological Society of America*, 113(2):114–124, 2020.

[29] M. Johnson, S. Fortna, R. Hollingsworth, and **N. C. Manoukis**. Post-harvest population reservoirs of coffee berry borer (*Hypothenemus hampei*) on Hawaii Island. *Journal of Economic Entomology*, 112(6):2833 – 2841, 2019.

[30] L. Hamilton, R. Hollingsworth, M. Sabado-Helpern, **N. C. Manoukis**, P. Follett, and M. Johnson. Coffee berry borer (*Hypothenemus hampei*) (coleoptera: Curculionidae) development across an elevational gradient on Hawaii Island: Applying laboratory degree-day predictions to natural field populations. *PLoS ONE*, 14(7):e0218321, 2019.

[31] **N. C. Manoukis**, R. Vargas, L. Carvalho, T. Fezza, S. Wilson, T. Collier, and T. Shelly. A field test on the effectiveness of male annihilation technique against *Bactrocera dorsalis* (diptera: Tephritidae) at varying application densities. *PLoS ONE*, 14(3):e0213337, 2019.

[32] K. Lehman, D. Barahona, **N. C. Manoukis**, L. Carvalho, S. De Faveri, J. Auth, and M. Siderhurst. Raspberry ketone trifluoroacetate trapping of *Zeugodacus cucurbitae* in Hawaii. *Journal of Economic Entomology*, 112(3):1306–1313, 2019.

[33] **N. C. Manoukis** and T. Collier. Computer vision to enhance behavioral research on insects. *Annals of the Entomological Society of America*, 112(3):227–235, 2019.

[34] P. Follett, **N. C. Manoukis**, and B. Mackey. Comparative cold tolerance in *Ceratitis capitata* and *Zeugodacus cucurbitae* (diptera: Tephritidae). *Journal of Economic Entomology*, 111(6):2632–2636, 2018.

[35] **N. C. Manoukis**, D. Cha, R. Collignon, and T. E. Shelly. *Terminalia* larval host fruit reduces the response of *Bactrocera dorsalis* (diptera: Tephritidae) adults to the male lure methyl eugenol. *Journal of Economic Entomology*, 111(4):1644–1649, 2018.

[36] M. A. Johnson, R. H. Hollingsworth, S. Fortna, and **N. C. Manoukis**. The Hawaii Protocol for scientific monitoring of coffee berry borer: A model for coffee agroecosystems worldwide. *Journal of Visualized Experiments*, 133:e57204, 2018.

[37] T. E. Shelly and **N. C. Manoukis**. Capture of melon flies, *Zeugodacus cucurbitae* (Diptera: Tephritidae), in a food-baited multilure trap: Influence of distance, diet, and sex. *Journal of Asia-Pacific Entomology*, 21(1):288–292, 2018.

[38] L. Aristizabal, M. A. Johnson, S. Shriner, R. H. Hollingsworth, **N. C. Manoukis**, R. Myers, P. Bayman, and S. Arthurs. Integrated pest management of coffee berry borer in Hawaii and Puerto Rico: Current status and prospects. *Insects*, 8:123, 2017.

[39] T. Collier and **N. C. Manoukis**. Evaluation of predicted Medfly (*Ceratitis capitata*) quarantine length in the United States utilizing degree-day and agent-based models. *F1000 Research*, 6:1863, 2017.

[40] E. B. Jang, R. V. Dowell, and **N. C. Manoukis**. Mark-release-recapture experiments on the effectiveness of methyl eugenol-spinosad male annihilation technique against an invading population of *Bactrocera dorsalis*. *Proceedings of the Hawaiian Entomological Society*, 49(1):37–45, 2017.

[41] M. A. Khan, **N. C. Manoukis**, T. Osborne, I. M. Barchia, G. M. Gurr, and O. L. Reynolds. Semiochemical mediated enhancement of males to complement sterile insect technique in management of the tephritid pest *Bactrocera tryoni* (Froggatt). *Scientific Reports*, 7:13366, 2017.

[42] J. Gaertner, V. B. Genovese, C. Potter, K. Sewake, and **N. C. Manoukis**. Vegetation classification of coffea on Hawaii island using Worldview-2 satellite imagery. *Journal of Applied Remote Sensing*, 11:046005, 2017.

[43] R. da Silva Gonçalves, **N. C. Manoukis**, and D. E. Nava. Effect of *Fopius arisanus* sonan oviposition experience on parasitization of *Bactrocera dorsalis* hendel. *BioControl*, 62(5):595 – 602, 2017.

[44] **N. C. Manoukis**, E. B. Jang, and R. V. Dowell. Survivorship of male and female *Bactrocera dorsalis* in the field and the effect of male annihilation technique. *Entomologia Experimentalis et Applicata*, 162:243–250, 2017.

[45] **N. C. Manoukis** and T. E. Mangine. Response of the pearly eye melon fly *Bactrocera cucurbitae* (coquillett) (diptera: Tephritidae) mutant to host-associated visual cues. *Proceedings of the Hawaiian Entomological Society*, 48:15–20, 2016.

[46] **N. C. Manoukis**. To catch a fly: Landing and capture of *Ceratitis capitata* in a Jackson trap with and without an insecticide. *PLoS ONE*, 11:e0149869, 2016.

[47] M. S. Siderhurst, S. J. Park, C. N. Suttles, I. M. Jaime, **N. C. Manoukis**, E. B. Jang, and P. W. Taylor. Raspberry ketone trifluoroacetate, a new attractant for the Queensland fruit fly (*Bactrocera tryoni* (Froggatt)). *Journal of Chemical Ecology*, 42:156–162, 2016.

[48] **N. C. Manoukis** and S. M. Gayle. Attraction of wild-like and colony-reared *Bactrocera cucurbitae* (Diptera:Tephritidae) to cuelure in the field. *Journal of Applied Entomology*, 140:241–249, 2016.

[49] **N. C. Manoukis**, M. Siderhurst, and E. B. Jang. Field estimates of attraction of *Ceratitis capitata* to trimedlure and *Bactrocera dorsalis* to methyl eugenol in varying environments. *Environmental Entomology*, 44:695–703, 2015.

[50] **N. C. Manoukis**, B. Hall, and S. M. Geib. A computer model of insect traps in a landscape. *Scientific Reports*, 4:7015, 2014.

[51] S. M. Geib, B. Calla, B. Hall, S. Hou, and **N. C. Manoukis**. Characterizing the developmental transcriptome of the oriental fruit fly *Bactrocera dorsalis* (Diptera: Tephritidae) through comparative genomic analysis with *Drosophila melanogaster* utilizing modENCODE datasets. *BMC Genomics*, 15:942, 2014.

[52] D. Shishika, **N. C. Manoukis**, S. Butail, and D. A. Paley. Male motion coordination in anopheline mating swarms. *Scientific Reports*, 4:6318, 2014.

[53] **N. C. Manoukis**, S. M. Geib, and R. I. Vargas. Effect of host *Bactrocera dorsalis* sex on yield and quality of the parasitoid *Fopius arisanus. BioControl*, 59:395–402, 2014.

[54] R. S. Lees, B. Knols, R. Bellini, M. Q. Benedict, A. Bheecarry, H. C. Bossin, D. D. Chadee, J. Charlwood, R. K. Dabiré, L. Djogbenou, A. Egyir-Yawson, R. Gato, L. C. Gouagna, M. M. Hassan, S. A. Khan, L. L. Koekemoer, G. Lemperiere, **N. C. Manoukis**, R. Mozuraitis, R. J. Pitts, F. Simard, and J. Gilles. Review: Improving our knowledge of male mosquito biology in relation to genetic control programmes. *Acta Tropica*, 132 Supplement:S2–S11, 2014.

[55] **N. C. Manoukis**, S. Butail, M. Diallo, J. M. Ribeiro, and D. A. Paley. Stereoscopic video analysis of *Anopheles gambiae* behavior in the field: Challenges and opportunities. *Acta Tropica*, 132 Supplement:S80–S85, 2014.

[56] **N. C. Manoukis** and K. Hoffman. An agent-based simulation of extirpation of *Ceratitis capitata* applied to invasions in California. *Journal of Pest Science*, 87(1):39–51, 2014.

[57] C. L. Chang, I. K. Cho, Q. X. Li, **N. C. Manoukis**, and R. I. Vargas. A potential field suppression system for *Bactrocera dorsalis* hendel. *Journal of Asia-Pacific Entomology*, 16(4):513–519, 2013.

[58] S. M. Gayle, M. McKenney, P. Follett, and **N. C. Manoukis**. A novel method for rearing wild tephritid fruit flies. *Entomologia Experimentalis et Applicata*, 148(3):297–301, 2013.

[59] R. I. Vargas, J. D. Stark, J. Banks, L. Leblanc, **N. C. Manoukis**, and S. Peck. Spatial dynamics of two Oriental Fruit Fly (diptera: Tephritidae) parasitoids, *Fopius arisanus* (Sonan) and *Diachasmimorpha longicaudata* (Ashmead) (Hymenoptera: Braconidae), in a guava orchard in Hawaii. *Environmental Entomology*, 42(5):880–901, 2013.

[60] S. Butail, **N. C. Manoukis**, M. Diallo, J. M. Ribeiro, T. Lehmann, and D. A. Paley. The dance of male *Anopheles gambiae* in wild mating swarms. *Journal of Medical Entomology*, 50(3):552–559, 2013.

[61] **N. C. Manoukis** and E. B. Jang. The diurnal rhythmicity of *Bactrocera cucurbitae* (diptera: Tephritidae) attraction to cuelure: Insights from an interruptable lure and computer vision. *Annals of the Entomological Society of America*, 106(1):136–142, 2013.

[62] R. I. Vargas, L. Leblanc, E. J. Harris, and **N. C. Manoukis**. Regional suppression of *Bactrocera* fruit flies (diptera: Tephritidae) in the pacific through biological control and prospects for future introductions into other areas of the world. *Insects*, 3(3):727–742, 2012.

[63] S. Butail, **N. C. Manoukis**, M. Diallo, J. M. Ribeiro, T. Lehmann, and D. A. Paley. Reconstructing the flight kinematics of swarming and mating behavior in wild mosquitoes. *Journal of the Royal Society Interface*, 7(9):2624–2638, 2012.

[64] **N. C. Manoukis**, I. Baber, M. Diallo, N. Sogoba, and J. M. Ribeiro. Seasonal climate effects anemotaxis in newly emerged adult *Anopheles gambiae* Giles in Mali, West Africa. *PLoS ONE*, 6(11):e26910, 2011.

[65] **N. C. Manoukis**, S. M. Geib, D. M. Seo, M. P. McKenney, R. I. Vargas, and E. B. Jang. An optimized protocol for rearing *Fopius arisanus*, a parasitoid of tephritid fruit flies. *Journal of Visualized Experiments*, 53:e2901, 2011.

[66] J. M. Ribeiro, J. M. Anderson, **N. C. Manoukis**, Z. Meng, and I. M. Francischetti. A further insight into the sialome of the tropical bont tick, *Amblyomma variegatum. BMC Genomics*, 12(136), 2011.

[67] I. Francischetti, J. M. Anderson, **N. C. Manoukis**., V. M. Pham, and J. M. Ribeiro. An insight into the sialotranscriptome and proteome of the coarse bontlegged tick, *Hyalomma marginatum* rufipes. *Journal of Proteomics*, 74(12):2892–2908, 2011.

[68] I. Baber, J. P. Tamby, **N. C. Manoukis**, D. Sangaré, S. Doumbia, S. F. Traoré, M. S. Maiga, and D. Dembélé. A python module to normalize microarray data by the quantile adjustment method. *Infection, Genetics and Evolution*, 11(4):765–768, 2011.

[69] I. Baber, M. Keita, N. Sogoba, M. Konate, M. Diallo, S. Doumbia, S. F. Traoré, J. M. Ribeiro, and **N. C. Manoukis**. Population size and migration of *Anopheles gambiae* in the Bancoumana region of Mali and their significance for efficient vector control. *PLoS ONE*, 5(4):e10270, 2010.

[70] A. Diabaté, A. Dao, A. S. Yaro, A. Adamou, R. Gonzalez, **N. C. Manoukis**, S. F. Traoré, R. W. Gwadz, and T. Lehmann. Spatial swarm segregation and reproductive isolation between the molecular forms of *Anopheles gambiae. Proceedings of the Royal Society B*, 276(1676):4215–4222, 2009.

[71] **N. C. Manoukis**, A. Diabaté, A. Abdoulaye, M. Diallo, A. Dao, A. S. Yaro, J. M. Ribeiro, and T. Lehmann. Structure and dynamics of male swarms of *Anopheles gambiae. Journal of Medical Entomology*, 46(2):227–235, 2009.

[72] **N. C. Manoukis**, J. R. Powell, M. B. Touré, A. Sacko, F. E. Edillo, M. B. Coulibaly, S. F. Traoré, C. E. Taylor, and N. J. Besansky. A test of the chromosomal theory of ecotypic speciation in *Anopheles gambiae*. *Proceedings of the National Academy of Sciences, USA*, 105(8):2940 – 2945, 2008.

[73] **N. C. Manoukis**. FORMATOMATIC: A program for converting diploid allelic data between common formats for population genetic analysis. *Molecular Ecology Notes*, 7(4):592 – 593, 2007.

[74] J. Marshall, K. Morikawa, **N. C. Manoukis**, and C. E. Taylor. Predicting the effectiveness of population replacement strategy using mathematical modeling. *Journal of Visualized Experiments*, 5:07/04/2007, 2007.

[75] R. Calsbeek, C. Bonneaud, S. Prabhu, **N. C. Manoukis**, and T. B. Smith. Multiple paternity and sperm storage lead to increased genetic diversity in *Anolis* lizards. *Evolutionary Ecology Research*, 9(3):495 – 503, 2007.

[76] **N. C. Manoukis**, M. B. Touré, I. Sissoko, S. Doumbia, S. F. Traoré, M. A. Diuk-Wasser, and C. E. Taylor. Is vector body size the key to reduced malaria transmission in the irrigated region of Niono, Mali? *Journal of Medical Entomology*, 43(5):820–827, 2006.

[77] M. A. Diuk-Wasser, G. Dolo, M. Bagayoko, N. Sogoba, M. B. Touré, M. Moghaddam, **N. C. Manoukis**, S. Rian, S. F. Traoré, and C. E. Taylor. Patterns of irrigated rice growth and malaria vector breeding in Mali using multi-temporal ERS-2 synthetic aperture radar. *International Journal of Remote Sensing*, 27:535–548, 2006.

[78] M. A. Diuk-Wasser, M. B. Touré, G. Dolo, M. Bagayoko, N. Sogoba, S. F. Traoré, **N. C. Manoukis**, and C. E. Taylor. Vector abundance and malaria transmission in rice-growing villages in Mali. *American Journal of Tropical Medicine and Hygiene*, 16:725–731, 2005.

Research Proposal for the California Olive Committee

Project Title:

Improved monitoring of olive fruit fly through gamma-hexalactone lures and improved attract-and-kill control of olive fruit fly through the combination of gamma hexalactone and GF-120.

Principal Investigator:

R. A. Van Steenwyk (Corresponding PI)

130 Mulford Hall,

Dept. of E.S.P.M.

University of California,

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bobvanst@berkeley.edu

Cooperators:

Spencer Walse, USDA-ARS, lure production advisor

Daniel Kuzmich, Kuzmich AG-Pheromone Resources, lure production and analysis

Cindy Kron, IPM Advisor UCANR Santa Rosa, research site selection

Idongesit Mokwunye, IPM Advisor UCANR Fresno, research site selection

Objectives of the Proposed Research:

1. Development of a gamma (γ)- hexalactone lure for improved monitoring of olive fruit fly (OLFF).
2. Development of an attract-and-kill control program for OLFF based on a γ-hexalactone and GF-120 combination.

Justification and Importance of the Proposed Research:

The olive fruit fly (*Bactrocera oleae*) is the most damaging direct fruit pest of table olives and is a serious pest of olives throughout the world. OLFF is native to eastern Africa and has been a devastating pest in the Mediterranean region for over 2,000 years. It was first found in the United States in Los Angeles County in 1998 and rapidly spread throughout California. Monitoring of OLFF populations has relied on yellow panel sticky traps or McPhail traps. The yellow panel traps are less effective than the McPhail traps but much easier to maintain. The yellow panel traps are baited with ammonium carbonate lures with or without a spiroketal sex-pheromone while McPhail traps are baited with Torula yeast pellets with or without a spiroketal sex-pheromone. Monitoring of OLFF adult populations is important to determine the need and timing of control.

For years, the primary material utilized for OLFF control has been GF-120 (spinosad) in an attract-and-kill strategy. GF-120 is applied by ground as large droplets by a modified weed sprayer mounted on an ATV. Because of the repeated use of GF-120 with little or no material rotation, many growers have been reporting decreased efficacy of GF-120. Alternative materials to GF-120 are Kaolin clay and Danitol. Kaolin clay is organically acceptable but needs to be removed from the fruit after harvest. Danitol can result in flare-ups of olive scale by suppressing beneficial insects. Thus, studies have been conducted and are currently in trial to develop additional insecticides for control of OLFF (Van Steenwyk et al. 2018 and Thayer et al. 2024). Recently, a new aggregation pheromone (γ-hexalactone) that could have a significant impact on both improved efficacy of GF-120 and monitoring has been discovered (Lopez et al. 2024). Lopez et al. 2024 showed that traps baited with γ-hexalactone and ammonium bicarbonate captured significantly more male and female OLFF than traps baited with ammonium bicarbonate alone. Also, traps baited with γ-hexalactone and ammonium bicarbonate captured significantly more male and female OLFF than baited with spiroketal sex-pheromone and ammonium bicarbonate. Thus, trapping protocols that use commercially available food-grade γ-hexalactone instead of the synthetically prepared sex-pheromone (spiroketal) may be more cost effective.

The discovery by Lopez et al. 2024 opens up the possibility of greatly improving captures of OLFF on yellow panel traps by developing a γ-hexalactone lure that can be used in conjunction with an ammonium carbonate lure, i.e. the traps would contain both lures. This would result in ca. 50 to

100% greater fly captures over ammonium carbonate alone based on the work of Lopez et al. 2024. Also, γ-hexalactone could be incorporated into GF-120 which could greatly improve the attractiveness of the GF-120 in the attract-and-kill strategy. Current studies with delta (δ)-heptalactone/δ-hexalactone on walnut husk fly (WHF), *Rhagoletis completa*, has resulted in the development of a new lactone lure and when combined with GF-120 has shown significant improvement in the attractiveness of GF-120 (Van Steenwyk et al. 2024). Both OLFF and WHF are in the Tephritidae family of insects but are in different genera. Thus, it is speculated that many members of the Tephritidae family use the lactones as an aggregation pheromone to form leks for the purpose of mating. In addition to γ-hexalactone, Lopez et al. 2024 also reported the presence of δ-hexalactone in the head capsule of OLFF. Δ-hexalactone is also found in the head capsule of WHF (Sarles et al. 2018). The exact role of δ-hexalactone is unknown in OLFF, but in WHF, δ-hexalactone has activity in that a 50:50 combination of δ-hexalactone and δ-heptalactone captures more flies than either compound alone (Van Steenwyk et al. 2024). Lopez et al. 2024 reported no activity from δ-hexalactone but provided no data. However, it may have activity when combined with γ-hexalactone. Unfortunately, Lopez et al. 2024 did not provide a ratio of γ-hexalactone to δ-hexalactone. A one year study is proposed to develop a γ-hexalactone lure and determine the potential of adding γ-hexalactone to GF-120 to improve the efficacy of GF-120.

Procedures to Accomplish Objectives:

Lure Development

1. Efficacy of lures: Polymeric plug lures will be constructed by Kuzmich AG-Pheromone Resources in Fresno to contain 100 mg, 200 mg, and 400 mg of γ-hexalactone. There will be 12 plug lures of each concentration. There will be 7 treatments replicated 6 times. The 7 treatments will be: 1) 100 mg γ-hexalactone plus ammonium carbonate (AC), 2) 100 mg γ-hexalactone without AC, 3) 200 mg γ-hexalactone plus AC, 4) 200 mg γ-hexalactone without AC, 5) 400 mg γ-hexalactone plus AC, 6) 400 mg γ-hexalactone without AC, 7) AC alone, and 8) a blank trap. The plugs will be attached to yellow panel traps and will be

monitored weekly for 8 weeks. The trap locations will be rotated weekly. The number of males and females will be recorded separately. Fly catch will be analyzed statistically using mean separation tests. This data will provide information on the utility of the lures under field conditions.

2. Longevity of lures: Polymeric plug lures will be constructed by Kuzmich AG-Pheromone, Resources in Fresno to contain 100 mg, 200 mg, and 400 mg of γ-hexalactone. There will be 24 plugs of each concentration. The plugs will be attached to yellow panel traps and all deployed at the same time. Four lures of each concentration will be removed weekly for 6 weeks and stored at -20ºC at UCB. After 6 weeks, all lures will be transported to the Kuzmich AG-Pheromone Resources facilities and the amount of γ-hexalactone remaining in each lure will be determined by analytical gas chromatography methods. The amount of γ-hexalactone remaining in the lures will be analyzed statistically using mean separation tests. This data will provide information on the longevity of the lures.

3. Ratio of γ-hexalactone and δ-hexalactone: Polymeric plug lures will be constructed by Kuzmich AG-Pheromone Resources in Fresno to contain a total of 200 mg of γ-hexalactone and δ-hexalactone in the ratios of 100/0, 90/10, 75/25, 50/50 γ-hexalactone/δ-hexalactone and a blank trap. The plugs will be attached to yellow panel traps and will be monitored weekly for 5 weeks. The trap locations will be rotated weekly. The number of males and females will be recorded separately. Fly catch will be analyzed statistically using mean separation tests. This data will provide information on the utility of the lures under field conditions.

Attract-and-Kill Development

1. Improvement of attractiveness of GF-120 with γ-hexalactone. Six treatments will be replicated six times. The six treatments will be: dental wicks containing 2 ml of 1 to 1.5 GF-120 to water solution plus 1 ml of hexane containing 50 mg, 25 mg, 10 mg and 5 mg γ-hexalactone, no γ-hexalactone and a blank yellow panel trap. The dental wicks will be attached to yellow panel traps. The traps will be monitored for 5 consecutive days and trap

locations will be rotated after each reading. The number of males and females will be recorded separately. Fly catch will be analyzed statistically using mean separation tests. This data will provide information on the amount of γ-hexalactone needed and the longevity of the GF-120/γ-hexalactone combination.

Timetable for Project: This is a one-year project in which we will develop a new OLFF lure and evaluate the potential of using γ-hexalactone to enhance the efficacy of GF-120. However, Corteva Agriscience, the producer of GF-120 would need to be willing to modify their product before any additional work could proceed.

Present Outlook and Estimated Success in Accomplishing Objectives:
The one-year deliverables will be a commercial γ-hexalactone lure for monitoring OLFF and the amount of γ-hexalactone needed to be added to GF-120 to enhance the efficacy of GF-120.

Budget Support Summary by Objectives:
Total Budget Request for 1/1/25 to 12/31/26:
Van Steenwyk, UC Berkeley

Salaries and Benefits:			
Lab. Assist II, Step 1.5 (6 months = 1/2 year)		$	24,526
Subtotal	Sub 2	$	24,526
Employee benefits	Sub 6	$	13,370
	Subtotal	$	37,896
Supplies and Expenses:			
Yellow panel traps, ammonium carbonate lures, field & lab supplies, γ-hexalactone and production of γ-hexalactone lures from Kuzmich AG-Pheromone Resources ($17,000 of total).	Sub 3	$	22,000
Travel (40 trips at 250 miles/trip at 0.67/mile)	Sub 5	$	6,700
	TOTAL	$	66,596

Literature Cited:

Lopez, S., P. Actin, A. Gomez-Zubiaur, C. Corbella-Martorell, and C. Quero. 2024. A shift in the paradigm? A male-specific lactone increases the response of both sexes of the olive fruit fly *Bactrocera oleae* to the food lure ammonium bicarbonate. J. Pest Science. 965-978.

Sarles, L., B. Fassotte, A. Boullis, G. Lognay, A. Verhaeghe, I. Marko, and F. Verheggen. 2018. Improving the monitoring of the walnut husk fly (Diptera: Tephritidae) using male-produced lactones. J. Econ. Entomol. 111:2032-2037.

Thayer, M. R., C. Kron, and R. A. Van Steenwyk. 2024. Control of olive fruit fly in olives, 2022. Arthropod Management Test 49:D. https://doi.org/10.1093/amt/tsae080

Van Steenwyk, R. A., D. M. Lightle, C. S. Cabuslay, and J. M. Choi. 2018. Control of olive fruit fly in olive, 2017. Arthropod Management Tests. 43 D https://doi.org/10.1093/amt/tsy044

Van Steenwyk, R. A., S. S. Walse, D. Kuzmich, M. R. Thayer, and J. Rijal. 2024. Preliminary findings on the improved monitoring of walnut husk fly (*Rhagoletis completa*) through a new pheromone lure. Acta Hortic. (In press).

Approved by:

Michael J. Mascarenhas
Dept. Chair E.S.P.M.

Ernie Simpson

Project Plan/ Research Grant Proposal

Project Year: 2025

Project Leader: Ernie Simpson
Mailing Address: 320 County Road 15 Orland, California 95963
Phone: 530-865-9829 Cell: 530-518-4685
E-mail: ernsimp17@sbcglobal.net

Commodity: Olive

Problem and its Significance:

Since the detection of Olive Fruit Fly in California in 1998, it has been a concern to olive growers in commercial orchards; preventative sprays are necessary. Trapping to monitor the Olive Fruit Fly populations in individual orchards is recommended. This will allow growers and PCA's to follow trends to their orchards and help evaluate spray program efficacy. Having an idea of area-wide population trends will help growers and PCA's interpret the results from their orchards.

Objectives:

1: Provide timely information to area growers regarding area-wide olive fruit fly population trends.
2: Continue to develop a historical perspective of olive fruit fly populations for the area.

Plans and Procedures:

Starting in late March, plastic McPhail traps using Torula yeast tablets dissolved in water as the bait will be placed in one tree at 12 sites (6 in Glenn County and 6 in Tehama County). The same sites that have been used in previous years will be monitored again to allow for comparison of current years trap catches to previous years. Earlier work in Glenn and Butte Counties has shown that the plastic McPhail traps catch more flies than the commonly used yellow panel trap. Traps will be checked and flies counted weekly. The results and field observations will be reported via email to the COC for further distribution. Trapping results will be reported as male and female flies for individual traps and combined and averaged by site for a graphic presentation of the data. Trapping and reporting will be continued through November or until trap catches decline for the year.

Budget Request

Budget Year: 2025

Funding Source: California Olive Committee
Salaries plus insurance and vehicle cost________________________________**$9800**
Supplies and Expenses: Trapping Supplies______________________________**$800**
Travel 3120 mi. @ $.61/mi._______________________________________**\$1900**

Total____**$12500**

Originator's Signature____

Ernie Simpson

PROJECT PLAN/RESEARCH GRANT PROPOSAL

Project Year: 2025 Anticipated Duration of the project: April-October 2025

Project Leader: Bert Quezada **Project Co-Leader:** Jim Stewart

Location: Tulare County

Mailing Address: PO Box 1095, Exeter CA 93221

Phone: Bert: (559) 936-0102 Jim: (559) 730-6243

Email: bertq@agipmc.com jrs@agipmc.com

Project Title: Southern San Joaquin Valley Olive Fruit Fly Monitoring Project

Cooperating Personnel: Andrew Quezada & Laura Lampe

Keywords: Olive Fruit Fly, Monitoring, Traps

Commodity: Olive

PROBLEM AND ITS SIGNIFICANCE:

The monitoring of Olive Fruit Fly (OLFF) in commercial olive groves in the Southern San Joaquin Valley started in 2001. OLFF is potentially the most significant insect pest in commercial Olive.

OBJECTIVES:

The objective of this project would be to continue the monitoring program of adult OLFF in commercial olive groves in the Southern San Joaquin Valley. Detection and seasonal monitoring of OLFF and the accurate timing of control measures, primarily bait sprays, would be the goal of this project. Correlation of fly collections with fruit susceptibility to infestation would indicate to growers when initial bait treatments should be applied. In addition, monitoring would continue to give growers information on the general OLFF population. This information would be specific for only the groves being monitored and would be available to growers to aid in making OLFF management decisions in their respective groves in the area being trapped.

PLANS AND PROCEEDURES:

The locations will be Ivanhoe, Woodlake, Exeter, South Exeter, Tonyville, West Lindsay, Strathmore, Porterville, and Terra Bella. In addition, a site in the city of Visalia would also be monitored. All of these sites are in Tulare County where a high percentage of the commercial olives are located in the Southern San Joaquin Valley. Some of the sites have been monitored starting in 2001. All traps will be in place by March 28, 2024. The traps will be read and reported by April 4, 2025, and continue on a weekly basis. Two yellow panel traps with ammonium carbonate bait and male pheromone will be used per site. Traps will be serviced and OLFF counted weekly. Reports detailing the number of male and female Olive Fruit Fly found at each location will be submitted to the California Olive Committee and interested parties within 48 hours on a weekly basis during the project. The program will end October 31, 2025.

BUDGET REQUEST

Budget year: April 1, 2025-October 31, 2025

Funding Source: California Olive Committee

Ag IPM Consultants, Inc.

Salaries and benefits:	$9,000.00
Supplies:	
Traps, bait, and pheromone	$1,000.00
Travel:	
Mileage to trap sites	$2,000.00
Equipment:	$0.00
TOTAL	$12,000.00

Total funding from the California Olive Committee would be: $12,000.00

Bert Quezada
Project Leader
Ag IPM Consultants, Inc
PO Box 1095, Exeter CA 93221
Phone: (559) 936-0102

James R. Stewart
Project Co-Leader
Ag IPM Consultants, Inc
PO Box 1095, Exeter CA 93221
Phone: (559) 730-6243

******ACTION REQUIRED******

FROM: COC RESEARCH SUBCOMMITTEE

SUBJECT: APPROVAL OF AUTHORITY TO THE EXECUTIVE DIRECTOR AND CHAIRMAN TO APPROVE NO-COST EXTENSIONS.

BACKGROUND: Each year, researchers will request a no-cost extension should their program run past the fiscal year. COC staff asks that the Committee grant authority to the Executive Director in conjunction with the Chairman to approve requests for no-cost extensions.

The below three projects are requesting No-Cost Extensions:

1. Evaluation of effects of Acceede (ACC) on tree architecture, and harvester type on enhancing horticultural maturity and abcission zone development and commercial trunk shaking efficiency in table olives.
2. Integrating Alternate Bearing Mitigation Strategies in a Commercial Table Olive Orchard.
3. Survey of Xylella fastidiosa diversity within California olive trees.

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DEPARTMENT OF PLANT SCIENCES
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COLLEGE OF AGRICULTURAL AND
ENVIRONMENTAL SCIENCES
AGRICULTURAL EXPERIMENT STATION
COOPERATIVE EXTENSION

October 31, 2024

Elise Oliver
Program Supervisor ,
California Olive Committee
2565 Alluvial Avenue, Suite 152
Clovis, CA 93611

RE: Agency/SPO Awards # SP0A243762-TASK01

Dear California Olive Committee:

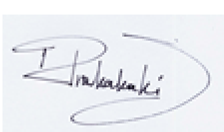
I would like to request a 6 month no-cost extension of funds in support of olive project entitled "Evaluation of effects of Accede® (ACC), tree architecture, and harvester type on enhancing horticultural maturity and abscission zone development and commercial trunk shaking efficiency in table olives" Jan 1- Dec 31, 2024.

This request is not for additional funds and we request that the project be extended until June 30 , 2025.

This extension is requested because the sample harvest just took place and expenses have not been yet registered. Thus, we need time to process and analyze the samples process the expenses.

Thank you for your time and consideration.

Sincerely,



__________________________ ___10- 31-2024________
Georgia Drakakaki, Professor Date

Agency Approval:

__________________________ ____________
Date

UC RIVERSIDE
UNIVERSITY OF CALIFORNIA

College of Natural and Agricultural Sciences
UC Division of Agriculture and Natural Resources
Agricultural Experiment Station and Cooperative Extension

Department of Botany and Plant Sciences-072
Riverside, CA 92521-0124

October 15, 2024

To: California Olive Committee

Attention: Elise Oliver, Program Supervisor
California Olive Committee
2565 Alluvial Avenue, Suite 152
Clovis, CA 93611
T: (559) 456-9096 | F: (559) 456-9099
www.calolive.org

From: Carol J. Lovatt
Professor of Plant Physiology, Emerita, and Plant Physiologist
Professor in the Graduate Division
Department of Botany & Plant Sciences-072
University of California-Riverside

Re: Request for a No-Cost Extension (NCE) through June 30, 2025

Project: Integrating Alternate Bearing Mitigation Strategies in a Commercial Table Olive Orchard

Elizabeth Fichtner and I are requesting a no-cost extension (NCE) for our current 2024 COC-funded project, "Integrating Alternate Bearing Mitigation Strategies in a Commercial Table Olive Orchard", through June 30th, 2025.

We request a no-cost extension for the following reasons:
1) Our olive research again this year, as in previous years, runs from pre-bloom of the funded year (2024) to full bloom (April/May) of the following year (2025).
2) The objectives for funded year 2024 of our COC project include collecting the second half of the data on recovery of shoot growth the following spring (April 2025) for trees that were treated in 2024. These data will be collected in the orchard in Exter only, due to the need to replace the Woodlake orchard as a result of poor fruit set and low yields in this orchard.
3) The objectives for the 2024 funded year of our COC project also require the evaluation of bloom the following spring (2025) to quantify the efficacy of the treatments applied in 2024 on return bloom for the orchard in Exeter. In addition, we will use these funds to collect the 2025 bloom estimates in the replacement orchard.
4) In 2024, despite a heavy ON-crop bloom, there were few fruit set in our Woodlake research orchard, so we did not apply two of the seven treatments and there was not enough fruit to obtain meaningful yield data to warrant harvesting the experiment at Woodlake. There were no problems at the Exeter site and the research was conducted as planned. As a result, we did not expend our full budget. With the NCE we will be able to use the 2024 funds to partially support the research proposed for 2025. We plan to submit a reduced budget for our 2025 proposal.
5) Additionally, it is anticipated that there will be a small amount of funds left over in the 2023 COC funds at UCR that support the independent contractor, who is the statistician for our

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College of Natural and Agricultural Sciences
UC Division of Agriculture and Natural Resources
Agricultural Experiment Station and Cooperative Extension

Department of Botany and Plant Sciences-072
Riverside, CA 92521-0124

project. We would like your permission to move these funds into the 2024 COC funds. This will assist us with our plan to reduce the budget for our 2025 proposal.

Thank you for your assistance with our request.

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******ACTION REQUIRED******

FROM: COC RESEARCH SUBCOMMITTEE

SUBJECT: APPROVAL OF 2025 BUDGET

RECOMMENDATION: THAT the Subcommittee approve various research projects for 2025.

BACKGROUND: Each year the Research Subcommittee approves various research projects funded by the Full Committee. The Subcommittee must determine which proposed projects to recommend to the Full Committee for funding. An estimated budget of **$493,218** (with no-cost extensions) is proposed based on the submitted projects.

#	Researcher	Project	Amount
1	Dr. Jim Adaskaveg*	Epidemiology and management of olive knot caused by Pseudomonas savastanoi pv. Savastanoi (year 1)	$15,490
2	Rodrigo Almeida	Survey of Xylella fastidiosa genetic diversity within California olive trees (year 2)	$30,361
3	Georgia Drakakaki and Becky Wheeler-Dykes**	Evaluation of effects of Accede® (ACC) at two different application rates on enhancing horticultural maturity and abscission zone development and commercial trunk shaking efficiency in table olives (year 3)	$115,129.20
4	Dr. Carol Lovatt and Elizabeth Fichtner	Integrating Alternate Bearing Mitigation Strategies in a Commercial Table Olive Orchard (year 3)	$29,156
5	Elizabeth Fichtner	Designing a web app for predicting risk of olive fruit fly—a tool for California olive growers and pest control advisors	$19,860
6	Franklin Lewis	Development of screening tools to determine Xylella fastidiosa tolerance in olives	$10,443
7	Joel Kimmelshue-Land IQ***	Olive Acreage Mapping	$30,700
8	Nicholas Manoukis	New prospects for the control of black scale in California olive groves	$73,000
9	R.A. Van Steenwyk	Improved monitoring of olive fruit fly through gamma-hexalactone lures and improved attract-and-kill control of olive fruit fly through the combination of gamma hexalactone and GF-120.	$66,596
10	Ernie Simpson	Sacramento Valley Olive Fruit Fly Monitoring Project	$12,500
11	Jim Stewart	Southern San Joaquin Valley Olive Fruit Fly Monitoring Project	$12,000
	2024 NCE-Carol Lovatt	Integrating Alternate Bearing Mitigation Strategies in a Commercial Table Olive Orchard	$13,530
	2024 NCE-Drakakaki	Evaluation of effects of Acceede (ACC) on tree architecture, and harvester type on enhancing horticultural maturity and abcission zone development and commercial trunk shaking efficiency in table olives	$52,552
	2024 NCE-Rodrigo Almeida	Survey of Xylella fastidiosa diversity within California olive trees	$11,900.80
		Contingency Fund	
		Total with No-Cost Extensions	**$493,218**
		Total without No-Cost Extensions	**$415,235.20**

*The COC traditionally co-funds this project with the OOCC.
**The COC will ask Valent if they are able to contribute any funds to reduce the fiscal impact as they did in 2024.
***The COC will also present this proposal to the OOCC to co-fund.

****ACTION REQUIRED****

FROM: COC RESEARCH SUBCOMMITTEE

SUBJECT: INTER-ITEM TRANSFERS OF THE RESEARCH BUDGET

RECOMMENDATION: THAT the Committee grant authority to the Executive Director and Chairman for inter-item transfers of the Research Budget.