

# AGENDA

### California Avocado Commission Production Research Committee Meeting

#### **Meeting Information**

Date: Thursday, April 3, 2025 Time: 8:00 a.m. Location: Hybrid Meeting

Physical Meeting Location: South Coast Research & Extension Center Annex Building Conference Room 7601 Irvine Blvd Irvine, CA 92618

Web Conference URL:

<u>n=81418771286</u>

Conference Call Number: (669) 900-6833 Meeting ID: 537 583 6823 Passcode: 348652

Meeting materials will be posted online at least 24 hours prior to the meeting at:

 $\underline{https://www.californiaavocadogrowers.com/commission/meeting-agendas-minutes}$ 

#### **Committee Member Attendance**

As of Friday, March 28, 2025, the following individuals have advised the Commission they will participate in this meeting:

- Danny Klittich, PRC Chair
- Allisen Carmichael
- Jim Davis
- Darren Haver
- Leo McGuire
- Daryn Miller
- Ryan Rochefort

| Time       | ltem |  |
|------------|------|--|
| 8:00 a.m.  | 1.   | Call to Order<br>a. Roll Call/Quorum   |
| 8:05 a.m.  | 2.   | <b>Opportunity for Public Comment</b><br>Any person may address the Committee at this time on any subject within the<br>jurisdiction of the California Avocado Commission. |
| 8:10 a.m.  | 3.   | <ul> <li>Approval of Minutes         <ul> <li>a. Consider approval of Production Research Committee Meeting Minutes of February 17, 2025</li> </ul> </li> </ul>            |
| 8:15 a.m.  | 4.   | Research Program Director's Report   |
| 8:20 a.m.  | 5.   | Action Items<br>a. Consider research proposals submitted for funding in response to<br>request for proposals   |
| 12:00 p.m. | 6.   | Adjourn Meeting  |
|            |      |  |

#### Disclosures

The times listed for each agenda item are estimated and subject to change. It is possible that some of the agenda items may not be able to be discussed prior to adjournment. Consequently, those items will be rescheduled to appear on a subsequent agenda. All meetings of the California Avocado Commission are open to the public and subject to the Bagley-Keene Open Meeting Act.

All agenda items are subject to discussion and possible action. For more information, or to make a request regarding a disability-related modification or accommodation for the meeting, please contact April Aymami at 949-341-1955, California Avocado Commission, 12 Mauchly, Suite L, Irvine, CA 92618, or via email at <u>aaymami@avocado.org</u>. Requests for disability-related modification or accommodation for the meeting should be made at least 48 hours prior to the meeting time. For individuals with sensory disabilities, this document is available in Braille, large print, audiocassette or computer disk. This meeting schedule notice and agenda is available on the internet at <u>https://www.californiaavocadogrowers.com/commission/meeting-agendas-minutes</u> and <u>http://it.cdfa.ca.gov/igov/postings/detail.aspx?type=Notices</u>.

If you have questions on the above agenda, please contact Tim Spann at <u>tim@spannag.org</u> or 423-609-3451.

Summary Definition of Conflict of Interest

It is each member's and alternate's responsibility to determine whether they have a conflict of interest and whether they should excuse themselves from a particular discussion or vote during a meeting. To assist you in this evaluation, the following *Summary Definition of Conflict of Interest* may be helpful.

A Commission *member or employee* has a conflict of interest in a decision of the Commission if it is reasonably foreseeable that the decision will have a material effect, financial or otherwise, on the member or employee or a member of his or her immediate family that is distinguishable from its effect on all persons subject to the Commission's jurisdiction.

No Commission member or employee shall make, or participate in making, any decision in which he or she knows or should know he or she has a conflict of interest.

No Commission member or employee shall, in any way, use his or her position to influence any decision in which he or she knows or should know he or she has a conflict of interest.

#### CALIFORNIA AVOCADO COMMISSION PRODUCTION RESEARCH COMMITTEE MEETING MINUTES

#### February 17, 2025

A meeting of the Production Research Committee (PRC) of the California Avocado Commission (CAC) was held on Monday, February 17, 2025, with the following people participating:

#### MEMBERS PARTICIPATING:

Danny Klittich, Chair Victor Araiza Allisen Carmichael Jim Davis Consuelo Fernandez Leo McGuire Daryn Miller Ryan Rochefort

#### **CAC STAFF PARTICIPATING:**

Ken Melban April Aymami

OFFICIALLY PARTICIPATING: Dr. Tim Spann, Spann Ag Research & Consulting

### GUESTS PARTICIPATING:

John Berns

#### CALL TO ORDER

Danny Klittich, Production Research Committee (PRC) Chairman, called the meeting to order at 2:01 p.m. with a quorum present.

#### **OPPORTUNITY FOR PUBLIC COMMENT**

There were no public comments.

#### APPROVAL OF MINUTES OF JANUARY 29, 2025 PRODUCTION RESEARCH COMMITTEE MEETING

#### <u>MOTION</u>

To approve the minutes of the January 29, 2025 Production Research Committee meeting.

(Davis/Rochefort) MSC unanimous

Motion 25-2-17-1

#### ACTION ITEMS

# A. Consider proposal "Surveys for avocado fruit feeding insect pests in Guatemala"

Chairman Klittich reminded the Committee that the Board had previously approved the concept of reevaluating the fruit feeding pests in Guatemala and it was the Committee's role to evaluate the scientific merit of the proposal being considered. Discussion ensued and there was general agreement that the proposal was scientifically sound and would achieve the stated objectives. Questions were posed about what data Guatemala has and if any of that information could be shared. Dr. Spann reminded the Committee that the Pest Risk Assessment (PRA) for Guatemala that was compiled by the USDA listed 10 fruit feeding pests of concern, six of which were identified by Dr. Hoddle during his previous CAC-funded project in 2006-09, and it was unlikely that Guatemala had done any work themselves since that time.

It was mentioned that USDA's standard procedure when a country asks for access is to ask the country for whatever information they have and then supplement that data with what is present in the scientific literature. Thus, it falls on the California industry to help populate the literature with accurate information.

The idea of working with the international avocado community to establish an insect taxonomy lab in Mexico was discussed. Although this would not address the current issue with Guatemala, it would help to further develop the database of avocado pests present in the native range of avocado.

The consensus of the Committee was that it was just a matter of time before pests appeared in the U.S. through fruit shipments and being prepared and able to quickly identify the pests and act would be beneficial to all avocado growers.

#### <u>MOTION</u>

To recommend funding the proposal "Surveys for avocado fruit feeding insect pests in Guatemala" as submitted for a total cost of \$510,202.

#### (Davis/Miller) MSC unanimous

#### Motion 25-2-17-2

#### B. Consider proposal "Chemical Synthesis and Field Evaluation of an Enantiopure (+)-Grandisol, the Putative Avocado Seed Weevil (*Heilipus lauri*) Aggregation Pheromone"

The Committee was aware of recent weevil finds in fruit in packinghouses in Mexico and immediately moved into discussion. The consensus was that the proposal was scientifically sound. Despite previous issues with trying to synthesize the pheromone, the chemist working on the project has two viable pathways to follow to overcome the challenges. There was agreement that having the pheromone would allow for a monitoring program in the U.S. for early detection of the seed weevils if they were to

arrive. But the pheromone was also a tool for use in countries of export for monitoring in export groves and packinghouses.

The Committee also considered the growing restrictions on pesticide use in California and the benefits of a tool like this pheromone will be essential if we ever need to manage this pest in California.

The question was asked whether the field testing of the pheromone could be conducted in Guatemala in conjunction with the pest survey work rather than having to make separate trips to Mexico for testing. Dr. Spann agreed to ask Dr. Hoddle to consider this option.

#### <u>MOTION</u>

To recommend funding the proposal "Chemical Synthesis and Field Evaluation of an Enantiopure (+)-Grandisol, the Putative Avocado Seed Weevil (Heilipus lauri) Aggregation Pheromone" as submitted for the total amount of \$349,212.

(McGuire/Davis) MSC unanimous

Motion 25-2-17-3

#### C. Consider proposal "Delimiting cryptic species within avocado seed moth, *Stenoma catenifer* for improved management and control of an economically important pest"

There was general agreement among the Committee that we don't fully understand what is going on with this pest and the species complex that composes it. However, there was some dismay at why no other avocado producing countries seem to be willing to help support this work and why they don't see the importance of it.

The question came up about whether fully understanding the species complex would allow for better pest management should these pests arrive in the U.S. Similarly, would this information be of benefit from a regulatory standpoint or would it be better to have a general classification of this pest and place the burden on the importing country to prove what species may be present there?

The consensus was that, although this is important work, it likely would not affect how we would manage the pest should it arrive in California and could be conducted at a later date. There was no support to recommend funding for this project.

#### ADJOURN MEETING

Danny Klittich, Production Research Committee (PRC) Chairman, adjourned the meeting at 2:54 p.m.

Respectfully submitted,

Timothy Spann

#### EXHIBITS ATTACHED TO THE PERMANENT COPY OF THESE MINUTES

- EXHIBIT A February 17, 2025, Production Research Committee AB 2720 Roll Call Vote Tally Summary
- EXHIBIT B Proposal, "Surveys for avocado fruit feeding insect pests in Guatemala"
- EXHIBIT C Proposal, "Chemical Synthesis and Field Evaluation of an Enantiopure (+)-Grandisol, the Putative Avocado Seed Weevil (Heilipus lauri) Aggregation Pheromone"
- EXHIBIT D Proposal, "Delimiting cryptic species within avocado seed moth, Stenoma catenifer for improved management and control of an economically important pest"

#### California Avocado Commission Research Proposal

**Project Title**: Validation of the use of flowers at the cauliflower stage for nutrient analysis to better time fertilizer applications

Anticipated project duration: July 1, 2025 – October 31, 2028

**Total Budget Requested:** \$265,213

Project Lead: Mary Lu Arpaia, Professor of Extension Dept. of Botany and Plant Sciences University of California Riverside, CA 92521 mlarpaia@ucanr.edu 559-646-6561

Project Cooperators: Peggy Mauk, Professor of Extension Dept. of Botany and Plant Sciences University of California Riverside, CA 92521 peggy.mauk@ucr.edu 951-827-4274

> Marllon Soares dos Santos, Post-doctoral Scholar Dept. of Botany and Plant Sciences University of California Riverside, CA 92521 <u>marllon.soaresdossantos@ucr.edu</u> 951-329-8573

Ben Faber, Farm Advisor – Ventura/Santa Barbara Counties University of California, Agricultural and Natural Resources (UCANR) 669 County Square Dr., Suite 100 Ventura, CA 93003 <u>bafaber@ucdavis.edu</u> 805-645-1462

Benjamin R. Waddell, Vice President Fruit Growers Laboratory Director of Agricultural Services Corporate Offices & Laboratory 853 Corporation Street Santa Paula, CA 93060 <u>benrw@fglinc.com</u> 805-392-2092

#### **Executive Summary**

Traditionally, leaf analysis is used as a tool to guide the application of nutrients to the tree. Previous research by UC Riverside (Embleton, et al, 1959) showed that collection of spring flush leaves from non-bearing shoots in the Fall of the same year were good at predicting nutritional tree status. There are several reports from the literature where it has been suggested that flower analysis may be more precise

in predicting nutrient status or yield potential. Specifically for avocado, Razeto and Salgado (2004) examined the use of this approach in managing nitrogen status for 'Hass' avocado. In 2017, Campisi Pinto et al. (2017) proposed using cauliflower stage inflorescences (CSI) as an alternative to fall leaf analysis. In this study, the authors showed: "nutrient concentrations of cauliflower stage inflorescences (CSI) collected in March proved better predictors of yield than inflorescences collected at full bloom (FBI) in April, fruit pedicels (FP) collected at five different stages of avocado tree phenology from the end of fruit set in June through April the following spring when mature fruit enter a second period of exponential growth, or 6-month-old spring flush leaves (LF) from nonbearing vegetative shoots collected in September (California avocado industry standard). For CSI tissue, concentrations of seven nutrients, nitrogen (N), phosphorus (P), potassium (K), magnesium (Mg), sulfur (S), zinc (Zn), and copper (Cu) were predictive of trees producing greater than 40 kg of fruit annually." However, the authors concluded that additional work would be needed to verify this approach before suggesting that the industry switch to this method. This original research utilized plant tissue from 6 orchards with varying tree age and rootstock. The results of Campisi Pinto et al (2017) were reported across varying rootstocks. Mickelbart et al (2007) demonstrated at the same research site in Irvine, CA that clonal avocado rootstocks can vary in their nutrient uptake on 'Hass' avocado as measured by fall leaf analysis. These differences in nutrient uptake, could influence the interpretation of the inflorescence nutrient data.

The proposed research aims to verify the 2017 published research over a 3-year period. We propose to collect tissue samples from 2 phenological stages, 6-month-old spring flush leaves (LF) and cauliflower stage inflorescences (CSI) from five groves each in San Diego, Ventura and Santa Barbara Counties. Individual trees at each site will serve as replicates (30 trees per site) so that yield data may also be collected. We will also collect samples as outlined in the 2017 trial, using 'Hass' avocado trees located on the UC, Riverside campus where we will also be able to verify fruit size distribution related to nutrient analysis and the potential impact of rootstocks on nutritional status.

#### Project Objectives

We wish to answer the following questions:

- A. Is there a correlation between inflorescence (50% cauliflower stage) and yield?
- B. Is inflorescence analysis superior or equivalent in predicting tree nutrient needs compared to traditional fall leaf analysis?
- C. How do both methods perform as a predictor of yield for individual trees? Can this be extended to sampling across a block of trees?

#### **Project Deliverables**

- A. Confirm observations by Campisi Pinto et al (2017) that inflorescence analysis can be used as a predictor of yield potential the following year.
- B. Understand the relationship between spring inflorescence analysis and fall leaf analysis on an individual tree basis.
- C. Can spring inflorescence analysis replace fall leaf analysis or should it be considered an additional tool in a grower's toolbox?
- D. Deliver information to the grower community to provide guidance on the feasibility of using spring inflorescence analysis to help guide yield predictions and nutrient needs through a series of industry articles, in-person presentations and webinars

#### Work Plan

**Deliverable A.** Confirm observations by Campisi Pinto et al (2017) that inflorescence analysis can be used as a predictor of yield potential the following year.

**7/01/25** – **10/31/25**. We will initiate the project in Spring 2025 using established research sites from currently funded CAC project, "Does artificial pollination improve yield of 'Hass' and 'GEM' avocado?". We have 6 research sites identified in Ventura County that include both the 'Hass' and 'Gem' and know tree age and rootstock have been documented. Inflorescence samples will be collected in March 2025 from 30 individual trees and prepared for subsequent analysis after the start of the project (7/1/25). Leaf samples will be collected from the same trees in September 2025 for standard leaf analysis. Tree canopy volume data will be collected from all trees in October 2025.

**11/01/25 – 10/31/26.** An additional 6 sites (4 'Hass', 2 'GEM') will be identified in Riverside/San Diego Counties for the project. Tree health ratings and soil analysis samples will be conducted at these and the Ventura County sites. We will collect inflorescence samples, collected from the 13 total sites, in Spring 2026 and leaf samples will be collected in Fall 2026. Yield data will be collected from the Ventura County sites in Spring (approximately) 2026. An additional 50 trees will be identified from a 'Hass' rootstock trial located at the University of California, Riverside Citrus Research Center – Agricultural Experiment Station (UCR). The UCR tree study will use 10 trees of 5 rootstock (Toro Canyon, Dusa, Zerala, and 2 experimental with pending release,). Inflorescence and leaf samples will be collected in the Spring and Fall, respectively. Tree canopy volume data will be collected from all trees in October 2026.

**11/01/26 – 10/31/27.** Inflorescence sampling and leaf sampling will occur at all sites in the Spring (2027) and Fall (2027) as outlined above. Yield data will be collected from individual data trees from all commercial sites depending on the cooperator's harvest schedule. Tree yield at UCR will occur in spring 2027. Tree canopy volume data will be collected from all trees in October 2027.

11/01/27 – 10/31/28. Yield data will be collected from all sites.

**Deliverable B.** Understand the relationship between spring inflorescence analysis and fall leaf analysis on an individual tree basis.

Throughout the 3 years of the project, as data from Deliverable A is collected, we will initiate statistical analysis of the data. The first meaningful analysis will only be attained following the first harvest of the Ventura County trees in 2026, but this data will provide preliminary insights into understanding the relationship between inflorescence and leaf analysis. Final data analysis will be undertaken when the 2027/2028 yield data is completed and will incorporate the canopy volume data to calculate yield efficacy (lb/m<sup>3</sup>).

**Deliverable C.** Can spring inflorescence analysis replace fall leaf analysis or should it be considered an additional tool in a grower's toolbox?

Data analysis will be ongoing throughout the project but final conclusion for this portion of the project will be completed during the project's final year after completion of the tree harvest.

**Deliverable D.** Deliver information to the grower community to provide guidance on the feasibility of using spring inflorescence analysis to help guide yield predictions and nutrient needs through a series of industry articles, in-person presentations and webinars.

This is the outreach part of the project. The project team will work with CAC, UCCE and the California Avocado Society to insure during the project's duration to extend pertinent data from the project through whatever means is most appropriate for the stage of the project.

#### **Methods**

**Inflorescence Sampling and Analysis.** As outlined by Campisi Pinto et al (2017) we will collect inflorescence samples (whole panicles) at the cauliflower stage when 50% of the trees at an individual site has 50% of the tree at Stage 8 based on the floral development scale of Salazar-Garcia et al (1998).

We will collect one to two panicles from either side of the tree, down the row) will be collected. The samples will be analyzed at Fruit Growers' Laboratory in Santa Paula, CA using standard protocols.

**Leaf Sampling and Analysis.** Leaf samples will be collected using mature spring flush leaves in early Fall from individual trees. Samples will be analyzed at Fruit Growers' Laboratory in Santa Paula, CA using standard protocols.

**Yield data.** Total fruit mass (lb/tree) and fruit count will be collected from each data tree at all commercial cooperator sites. At the UCR site when the trees are harvested the fruit will also be graded by size to examine the relationship between yield, fruit size distribution, canopy volume and inflorescence and leaf analysis. Additionally, UCR data will be analyzed to determine if there is a rootstock effect. We will also calculate the Alternate Bearing Index for all trees after the final year of data collection using the same approach as outlined by Campisi Pinto et al (2017).

**Statistics.** The data will be analyzed using regression models to assess the relationship between nutrient concentrations and yield. A Generalized Linear Model (GLM) will be employed to control for random effects, considering rootstock, location, and optimal nutrient concentration for productivity. Principal Component Analysis (PCA) will be used to enhance data visualization and identify key nutrient predictors. Additionally, the Alternate Bearing Index (ABI) will be calculated to quantify yield fluctuations over the study period. This part of the project will be overseen by Dr. Soares dos Santos.

**Potential Challenges/Obstacles That Could Delay or Prevent the Success of the Research.** The field work in this project is very straightforward, however, excellent communication with the field cooperators will be key to ensure that harvest data is collected in a timely manner and no mistakes are made that would prevent the collection of the yield data. We cannot foresee untimely weather events, but a freeze or extreme heat event may jeopardize the collection of data. We will have weather stations at all sites to ensure we can capture any data that could indicate the timing and/or magnitude of such an event. It is possible that the statistical analysis may be more challenging than anticipated but any problem such as this can be addressed. At worst case, this may cause slight delays in the final outcome of the project. By constantly updating our database, we should be able to anticipate any such problems.

#### Anticipated timeline for the project.

Year 1 (07/1/25 – 10/31/25). The activities for Year 1 will include:

- July Analyze Spring 2025 inflorescence samples (collected prior to funding) collected from Ventura County research sites (7)
- September Collect leaf samples from Ventura County research sites
- October collect canopy volume data from Ventura County research sites

Year 2 (11/01/25 – 10/31/26). The activities for Year 2 will include:

- October December: Identify cooperators (6) for Riverside/San Diego counties and quantify information such as historical yield, tree age, rootstock, tree health, and soil analysis. Collect same information from UCR research site
- March: collect inflorescence samples from all sites (Ventura, 7 sites; Riverside/San Diego, 6 sites, UCR)
- April June (approximate): collect yield from Ventura County research sites
- September: collect leaf samples from all sites
- October: collect canopy volume data from all research sites (14)

Year 3 (11/01/26 – 10/31/27). The activities for Year 3 would include:

• Collect yield data in collaboration with individual cooperators (13) and at UCR

• Repeat activities from Year 2 for third year of data collection

Year 4 (11/01/27 – 10/31/28). The activities for Year 3 would include:

- Collection of yield data
- Summation and analysis of data.

Statistical Analysis: Will be ongoing throughout project.

**Outreach Activity:** Will be ongoing throughout project and beyond.

#### Project Outreach

The 3 lead collaborators on this project (Arpaia, Mauk, Faber) all have Cooperative Extension appointments and are recognized to be committed to extending the information and conclusions from this project. We will use appropriate written, oral and web-based outlets such as articles in *From the Grove, CAS Quarterly, Topics in Subtropics Newsletter*, speaking at grower events such as the CAC/CAS/UCCE meetings and other venues and webinars such as Avocado Café.

#### **References**

Campisi-Pinto, S., Y. Zheng, P.E. Rolshausen, D.E. Crowley, B. Faber, G. Bender, M. Bianchi, T. Khuong, C.J. Lovatt. 2017. Optimal Nutrient Concentration Ranges of 'Hass' Avocado Cauliflower Stage Inflorescences—Potential Diagnostic Tool to Optimize Tree Nutrient Status and Increase Yield. HortScience. 52(12):1707-1715.

Embleton, T.W., W.W. Jones, and M.J. Garber., 1959. Leaf analysis as a guide to nitrogen fertilization of the Hass avocado. Calif. Avocado Soc. Yrbk. 43:94–95.

Razeto, B. and J. Salgado. 2004. The inflorescence and fruit peduncle as indicators of nitrogen status of the avocado tree. HortScience. 39:1173–1174.

Salazar-García, S., E.M. Lord, and C.J. Lovatt. 1998. Inflorescence and flower development of the 'Hass' avocado (*Persea americana* Mill.) during ''on'' and ''off'' crop years. J. Amer. Soc. Hort. Sci. 123:537–544.

Mickelbart, M. V., G. S. Bender, G. W. Witney, C. Adams and M. L. Arpaia. 2007. Effects of clonal rootstocks on 'Hass' avocado yield components, alternate-bearing, and nutrition. J. Hort. Sci. and Biotech. 82(3): 460-466.

#### Milestone Table

| Milestone | Activity  | Estimated<br>Complete Date | Estimated Budget<br>Allocated |
|-----------|---|----------------------------|-------------------------------|
| 1         | Completion of Year 1 activities (sampling, tissue<br>analysis, tree measurements, preliminary data<br>analysis)                   | 10/31/2025                 | \$16,147                      |
| 2         | Completion of Year 2 activities (sampling, tissue<br>analysis, yield collection, tree measurements,<br>preliminary data analysis) | 10/31/2026                 | \$83,303                      |
| 3         | Completion of Year 3 activities (sampling, tissue<br>analysis, yield collection, tree measurements,<br>preliminary data analysis) | 10/31/2027                 | \$77,791                      |
| 4         | Completion of Year 4 activities (yield collection, final data analysis)   | 10/31/2028                 | \$87,972                      |

#### Project Proposal Budget 7/1/25 – 10/31/28

|  | Year 1    | Year 2       | Year 3       | Year 4       |         |
|--|-----------|--------------|--------------|--------------|---------|
|  | 7/01/25 – | 11/01/2026 - | 11/01/2027 – | 11/01/2027 — |         |
|  | 10/31/25  | 10/31/2027   | 06/30/2028   | 06/30/2028   | TOTAL   |
| Salaries and Benefits                    | 7,287     | 49,713       | 51,701       | 76,722       | 185,423 |
| Postdocs/Research Associates (M. Santos) | 2,225     | 17,352       | 18,046       | 37,535       |         |
| Research Specialist (R. Li)              | 3,000     | 18,720       | 19,469       | 20,248       |         |
| Benefits                                 | 2,062     | 13,641       | 14,187       | 18,940       |         |
| Supplies and Expenses                    | 300       | 1,000        | 500          | 500          | 2,300   |
| Equipment                                | 0         | 8,000        | 0            | 0            | 8,000   |
| Services                                 | 7,560     | 16,590       | 16,590       |              | 41,490  |
| Tissue Analysis (FGL); see budget        |           |              |              |              |         |
| justification for detail                 | 7,560     | 15,840       | 15,840       |              |         |
| UCR land recharges                       |           | 750          | 750          | 750          |         |
| Travel                                   | 1,000     | 8,000        | 9,000        | 10,000       | 28,000  |
| Total Amount Requested                   | 16,147    | 83,303       | 77,791       | 87,972       | 265,213 |

#### **Budget Justification**

#### Senior Personnel:

PIs, Mary Lu Arpaia and Peggy Mauk - Although no funds are requested for PI and Co-PI's, will be responsible for managing the project as described in the proposal, coordinating the field and laboratory research projects, analyzing the project data, supervising and advising the research staff, and reporting the results of this project.

#### Other Personnel: \$136,593

Post Doctoral Researcher Marllon Soares dos Santos at 25% for Years 1-3; 50% for Year 4 of the project. Rates are calculated on a base annual salary of \$58,478 in Year one.

Research Specialist Rui Li at 10% for Year one (7/2025-10/2025) and 20% for Years 2 and 3. Base salary is \$90,000 in Year 1.

Dr. Santos and Dr. Li will be responsible for the northern and southern avocado field plots respectively. The 2 scientists will coordinate on harvesting activities. Dr. Santos will be responsible for overseeing all aspects of the project and will be responsible for the data summation and analysis.

All salaries and wages were estimated using UC Riverside's academic and staff salary scales. Subsequent years include escalations of 4% based on recommendations by our campus administrative officials.

#### Fringe Benefits: \$48,830

Fringe benefit rates are calculated as a percentage of the gross salary as follows: M. Santos (actual rate) in Year 1 is 22.3%; R. Li (actual rate) in Year 1 is 43.7%. Subsequent years include escalations based on recommendations by our campus administrative officials.

#### TOTAL PERSONNEL: \$185,423

#### SUPPLIES AND EXPENSES: \$2,300

Materials/Supplies: - Supplies such as bags and field supplies to maintain plots and collect data.

#### EQUIPMENT: \$7,000

We will install Davis Instrument weather stations at all sites, including UCR. Weather stations have already been purchased for 6 of the 7 sites in Ventura County as part of the pollination project. We are requesting funds to purchase for the remaining 8 sites.

#### SERVICES: \$41,490

Tissue Analysis (Fruit Growers' Laboratory (FGL)). There will be a large number of tissue samples generated for analysis as listed in the table below. FGL has requested partial reimbursement for analysis. A complete nutrient analysis for either inflorescence or leaf tissue that will include chloride and sulfur is listed at \$132 per sample. Typically, FGL would give a 30% discount for the number of samples that will be generated reducing the price to \$92 per sample. FGL has proposed that they will do a 80/20 (FGL/CAC) cost share which reduces the cost per sample for the project to \$18 per sample. This is savings over the course of the project of **\$161,320** since the total cost at \$92 per sample would be \$200,560. Additionally, FGL have offered to do the soil sampling at no cost.

| Project Year            | Site                      | Inflorescence | Leaf | Total |
|-------------------------|---------------------------|---------------|------|-------|
| 1 (07/1/25 – 10/31/25)  | 7 sites – Ventura County; | 210           | 210  | 420   |
|                         | 30 trees per site         |               |      |       |
| 2 (11/01/25 – 10/31/26) | 7 sites – Ventura County  | 210           | 210  | 880   |
|                         | 6 sites – Riverside/San   | 180           | 180  |       |
|                         | Diego Counties (30        |               |      |       |
|                         | trees/site)               |               |      |       |
|                         | 5 rootstocks – UCR; 10    |               |      |       |
|                         | trees each                | 50            | 50   |       |
| 3 (11/01/26 – 10/31/27) | 7 sites – Ventura County  | 210           | 210  | 880   |
|                         | 6 sites – Riverside/San   | 180           | 180  |       |
|                         | Diego Counties (30        |               |      |       |
|                         | trees/site)               |               |      |       |
|                         | 5 rootstocks – UCR; 10    |               |      |       |
|                         | trees each                | 50            | 50   |       |
| 4 (11/01/27 – 10/31/28) |                           | 0             | 0    | 0     |
| TOTAL SAMPLES           |                           |               |      | 2,180 |

Summary table of Tissue Samples to be collected.

#### TRAVEL: \$28,000

Travel is requested for Dr. Li to travel to southern Riverside/northern California to monitor flowering and for collection of inflorescence samples in Spring of Years 2 - 3 and collection of Fall leaf samples at 6 research sites. Also included in travel for Dr. Li is 5 overnight trips to Ventura County to assist Dr. Santos in collection of yield data in years 2 - 4. Round trips vary; it is estimated 150 miles for trips to Riverside/San Diego Counties and 270 miles for trips to Ventura County. Overnight lodging estimated at \$180 per night and meals/incidentals at \$80 per trip. Cost also includes estimate for a rental from UCR Fleet Services when shared travel with Dr. Santos is not possible.

Travel is requested for Dr. Santos to travel to Ventura County to monitor flowering and for collection of inflorescence samples in Spring of Years 2 - 3 and collection of Fall leaf samples in Years 1 - 3. Also included in travel for Dr. Santos is 5 overnight trips to Riverside/San Diego Counties to assist Dr. Li in collection of yield data in years 2 - 4. Round trips vary; it is estimated 270 miles for trips to Ventura County and 150 miles for trips to the southern sites. Overnight lodging estimated at \$180 per night and meals/incidentals at \$80 per trip. Cost also includes an estimate for a rental from UCR Fleet Services when shared travel with Dr. Li is not possible.

Three trips per year are also budgeted for Drs. Arpaia and Mauk to visit research sites with Drs. Santos and Li. Mileage is calculated using UC Riverside as a home base at a rate of \$0.70 per mile (2025 IRS allowed rate).

Project title: Creating a Weather Station Network to Guide Irrigation Decision of Avocados

Project leads: Andre Biscaro, Ben Faber UC Cooperative Extension, Ventura County asbiscaro@ucanr.edu; bafaber@ucanr.edu

#### Executive summary:

The two most important decisions for improving irrigation efficiency and its effect on yield and plant health are when to start the irrigation, and how long to irrigate. While soil moisture sensors are effective at telling when to irrigate, evapotranspiration (ET)based scheduling is our best tool to determine how long (or how much) to irrigate. With many irrigations in a crop cycle, ranch managers and irrigators decisions of how long to irrigate are rarely data driven and are most commonly done on a calendar-basis.

While weather station data can provide fairly accurate information to guide irrigation decisions, it is essential that its data are representative of the area of interest. With several different microclimates and complex aspect situations based on landscape position in Ventura County and throughout California, increased numbers of stations are essential to ensure accuracy. This project proposal addresses two topics in irrigation management: the introduction of a network of weather stations managed and maintained by UC ANR, and to improve the accuracy of water and nutrient applications with the use of the Irrigation Calculator for example, which is currently funded by the Avocado Commission. Once the concept is implemented and tested in Ventura County, its expansion to other counties will be streamlined. This project proposal will also investigate how the accuracy of reference ET (ETo) data is compromised with decreased size of the grass area around the station. While the Department of Water Resources currently requires 8 acres of well-watered grass to site a CIMIS station, no information has been provided or is currently available to address the gains in accuracy with the increased size of the grass area. Most, if not all of the Department of Water Resource's CIMIS sites have considerably less grass footprint than 8 acres.

Therefore, the overall goal of this project proposal is to assess the viability of using a reduced size of grass for ETo weather stations, and to establish a network of weather stations that can improve the adoption of data-driven decisions to optimize irrigation water and maximize yield and plant health.

List of specific project objectives:

Identify three cooperating growers who, paid a fee, can establish and maintain a wellwatered grass area of 100x100ft to host a weather station.

Purchase and install the stations.

Make sure the station's data is available online, free of charge.

Connect the stations to the irrigation calculator.

Identify one cooperating grower who, paid a fee, can establish and maintain a well-watered grass area of 4 acres to host a weather station with mobile sensors used to assess the

difference in accuracy between ETo data collected from the center of the 4 acres vs different distances from the edge of the grass.

Analyze data from the grass area size comparison.

Extend the information and access to weather stations to growers.

#### List of project deliverables:

Free access to four weather stations' data.

Improved irrigation recommendations of the irrigation app addressing weather conditions in different micro-climates. That will most likely lead to increased adoption of the irrigation app among avocado growers.

Improved understanding of how different grass area sizes affect the accuracy of reference evapotranspiration (ETo) data, and therefore its impact on irrigation recommendations. This factor has a direct impact on the possibility of expanding ETo weather stations with grass area sizes that can be more easily accommodated by several growers (e.g.: 100x100ft, or even 50x50ft).

The deliverables described above are contingent on securing cooperating growers willing to host these stations (plant and maintain the grass areas).

#### Work Plan and Methods:

The locations for the three stations installed in 100x100ft (0.2 acre) grass area will be identified based on differences of microclimate where avocado is commonly grown, in addition to land availability and suitability. The location for the station with 4-acre grass area will be identified based on land availability and suitability, also in an area where avocado is commonly grown.

Hourly and daily ETo data will be compared between the station installed in the center of the 4-acre grass field (base station) and another mobile station placed at the following distances from the edge of the field, in the prevailing wind side: 50, 150 and 250ft. While the base station will be at the center of the field for the entire year, the mobile station will be moved among the three sites (50, 150 and 250ft) every 30 days, totaling 120 days at each of the three sites. Moving the mobile station monthly will allow the comparisons to include at least one month within all sites (3) and seasons (4). The accuracy assessment will be estimated with both hourly and daily ETo change from the base station's value. Irrigation recommendations will be created with data from both stations and compared to assess if the ETo differences are meaningful to growers in terms of total water recommendations.

The limitation of this method is that the wind will not always come from the prevailing direction (most mornings and during specific Santa Ana winds), and therefore the air flowing towards the sensors would have passed through different lengths of grass than expected for each site. This can be addressed by removing data for periods when the wind is not from the prevailing direction.

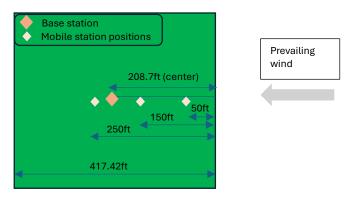


Figure 1. Illustration of how the base and mobile stations would be placed in the 4-acre grass field for the grass size assessment.

In addition to the method described above, ideally, one of the stations with the 0.2acre grass will be sited very close to the station with the 4-acre grass, allowing for another type of comparison: 4.0 vs 0.2 acre grass areas, where both stations will be at the center of their respective grass areas for the entire year.

The main challenge of this study is to find suitable sites and willing cooperators to host each station. The sites must be within certain proximity of avocado grown areas, without buildings and/or trees blocking the wind, and with a grower (or a landowner) willing to plant and maintain (irrigate, fertilize, mow etc) the grass field.

Answer to the reviewers' comments on the concept proposal: while we would want to capitalize on existing weather stations from growers, there are significant limitations on how the data can be used in terms of accuracy, considering none of these stations are surrounded by well-watered grass. However, we will assess if solar radiation from some of these stations can be used to estimate ETo using the remaining data from the stations from this project.

#### Project outreach:

The results of this project will be shared through grower meetings, field days hosted at one or all of the weather stations sites, an article in the California Avocado Commission magazine From the Grove, and a newsletter article.

| Milestone | Activities             | Scheduled  | Budget   |
|-----------|------------------------|------------|----------|
|           |                        | Completion |          |
| 1         | Meet with potential    | February   | \$88,375 |
|           | collaborators and      | 2026       |          |
|           | industry stakeholders  |            |          |
|           | to identify four sites |            |          |

#### Milestone Table:

|   | monthly, data analysis<br>Total   | Project Budget: | \$92,746 |
|---|---|-----------------|----------|
| 2 | • Data collection,<br>establish a<br>maintenance routine<br>for the stations, move<br>the mobile station    | June 2027       | \$4,368  |
|   | <ul> <li>where the stations can<br/>be installed.</li> <li>Purchase and install<br/>the stations</li> </ul> |                 |          |

#### Budget:

#### Estimated total project cost:

| \$     | Description                    |
|--------|--------------------------------|
| 32,052 | 3 x \$10,684 Campbell          |
|        | Scientific ETo stations        |
| 17,870 | 1 Campbell Scientific ETo      |
|        | station with mobile sensors    |
| 5,824  | SRA time: 128h @ \$26.86/h     |
|        | salary with 69.4% benefits;    |
|        | 32h for installation, and 8h   |
|        | per month to inspect and       |
|        | maintain/troubleshoot          |
|        | sensors x 12 months            |
| 2,000  | Travel expenses                |
| 20,000 | Grower incentive for           |
|        | planting and maintaining 4     |
|        | acres of grass (lease, water,  |
|        | labor, 1 year)                 |
| 15,000 | Grower Incentive for           |
|        | planting and maintaining       |
|        | 0.2 acre of grass (\$5,000 x 3 |
|        | sites x 1 year)                |
| 92,746 | Total Requested Funds          |

#### Budget Narrative:

#### Fiscal year 2025-26

 $32,052: 3 \times 10,684$  Campbell Scientific ETo stations from the Western Weather Group. These stations will be installed at the 0.2 acre grass sites.

\$17,870: 1 x Campbell Scientific ETo station with mobile sensors from the Western Weather Group. The base and mobile station will be installed at the 4-acre grass site to assess the grass area requirements.

\$1,456: Staff Research Associate time to support the installation of the stations: 32h @ \$26.86/h salary with 69.4% benefits.

\$20,000: Grower incentive for planting and maintaining 4 acres of grass (lease, water, labor, 1 year). It is possible that a grower will charge less for this, but we want to make sure the amount offered is attractive.

\$15,000: Grower incentive for planting and maintaining 0.2 acre of grass (\$5,000 x 3 sites x 1 year). Yearly cost will be renegotiated with cooperator and additional funds will be requested after the first year in case promising results are obtained in the first year.

#### Fiscal year 2026-27

\$2,000: Travel expenses. Funds will support travel expenses of UC Davis Biometeorologist Rick Snyder to assess project details after the installation of the stations and data analysis.

\$4,368: Staff Research Associate time to support monthly inspection, maintenance and troubleshooting of the stations: 96h @ \$26.86/h salary with 69.4% benefits.

Title: A pesticide resistance monitoring program for avocado thrips

PI: Hamutahl Cohen, Assistant Entomology Advisor, Ventura, UC ANR Cooperative Extension Co-PI: Bodil Cass, Assistant Subtropical Entomology Specialist, UC Riverside Co-PI: Laura Leger, Postdoctoral Researcher, UC Riverside Co-PI: Ben Faber, Subtropical Crops Advisor, Ventura, UC ANR Cooperative Extension

#### **Executive Summary**

Pesticide resistance is a major global challenge threatening food security and resulting in increased pesticide use. Our aim is to develop a regional resistance monitoring program for avocado thrips (Scirtothrips persea), a severe pest of avocado in Ventura County that is vulnerable to pesticide resistance due to its high fecundity, short life cycle, asexual reproduction, and cryptic behavior. Abamectin is the primary chemical control product for avocado thrips because it has strong efficacy and a limited impact on natural enemies - however, resistance with abamectin is likely because it has long a persistence inside leaf tissues which may subject sequential generations of thrips to the same chemical mode of action. Resistance is also likely because it is often applied more than once a year for control of both avocado thrips in the spring and persea mites later in the season. For avocado thrips, resistance monitoring has not been conducted in over 12 years. To obtain new baseline resistance data for avocado thrips, we will establish study sites in Ventura County and annually monitor avocados thrips for resistance at these sites using bioassays. This program will set the groundwork for offering growers resistance diagnostic services in the future wherein we could compare grower-submitted samples to baseline resistance levels at the nearest study site from this project. This program provides a critical contribution to the avocado industry in Ventura because it is unlikely that abamectin will be easily replaced if lost to resistance.

#### **Project Narrative**

Avocado thrips (*Scirtothrips persea*) arrived in California in 1996 from Mexico. Without available control mechanisms, heavily infested orchards in Ventura County experienced 50% to 80% crop damage in 1997, and much of the fruit was unmarketable (Hoddle et al. 2002). Today, avocado thrips are thought to infest 80% of the state's ~53,000 avocado acres (Hoddle et al. 2002). Because this pest lacks effective natural predators in California, the use of chemical control is one of the primary control options. Although growers rely on pesticide applications to control avocado thrips, this tool is threatened by the development of pesticide resistance. We are proposing to develop a pesticide resistance monitoring program for avocado thrips and disseminate best practices for the prevention of resistance development.

Avocado thrips are small, slender, straw-yellow insects that are a serious threat to avocado production. Adult females lay eggs on immature leaves and fruit. Thrips larvae and adults can build to high densities on young leaves during the spring, then move to fruits when the leaves harden off. Losses are caused by lesions from feeding. Thrips mouthparts consist of mandibular stylets that pierce plant tissue and result in deformation of the fruit in the form of elongated, ridged scarring that looks like "alligator skin" and can downgrade fruit at harvest and result in loss of value to the grower (Ávila-Quezada et al. 2005, Goldarazerna 2015).

To control avocado thrips, the chemical control option with the greatest IPM value is abamectin (Agri-Mek) because it is considered fairly innocuous to natural enemies and pollinators. This product is a macrocylic lactone, unstable in sunlight, exhibits translaminar activity, and must be used with oil. Thrips exposed to abamectin take 3-5 days to die and, thus, control can be somewhat slower than with faster acting insecticides. This material is quite persistent in leaves (Rugg et al. 2005) and treatments and can hold for 6-10 weeks or more. In 2022, California growers applied approximately 400lbs of the active ingredient abamectin to avocado for the control of avocado thrips and persea mite (California Department of Pesticide Regulation 2022).

The concern is that grower reliance on abamectin will result in pesticide resistance. Resistance is a phenomenon in which insect evolve physiological and chemical mechanisms to overcome pesticide exposure. These mechanisms include toxicodynamic and toxicokinetic changes, such as reduced penetration, activation, detoxification, and excretion. For thrips in particular, resistance is a global issue. There are over 150 worldwide cases of insecticide resistance associated with different thrips species, including products in seven chemical classes (Gao et al. 2012). This is because thrips species have short generation times, high reproductive fecundity, and a haplodiploid breeding system where resistance genes can be passed undiluted from females to their offspring as they do not require mating to reproduce. The likelihood of resistance developing is further exacerbated by reliance on a single active ingredient for control, which creates stronger selection pressure from repeated, successive applications.

The combination of thrips reproductive biology and the lack of other management options makes the California avocado system particularly at risk for developing avocado thrips resistance. If avocado thrips become resistant to available control tools, they will become increasingly difficult to control. Once resistance develops, product efficacy may be lost for years or indefinitely. This has been the case for sabadilla (Humeres & Morse, 2006). Because of environmental, economic, and health concerns, new insecticide chemistries can take many years to become available. It is therefore critical to monitor pesticide resistance to inform growers about thrips susceptibility and resistance. This information about local resistance levels helps growers distinguish control failures due to resistance, from control failures due to other causes including high pest pressure or application failures (timing, coverage, etc.), and inform management decisions moving forward.

#### Deliverables

They key deliverable of our project is **a resistance monitoring program** resulting in publicly accessible, easy-to-read results for our local avocado community shared online. We will obtain baseline resistance levels for avocado thrips in our county so that UCCE Ventura can offer diagnostic services in the future where thrips resistance can be compared to baseline data. We are focusing on avocado because the industry has specifically requested support for grower decision-making with regards to pesticide resistance and identified this as a project of interest.

#### **Objectives**

While growers *can* manage and prevent resistance, they need data on resistance development to inform practices such as reduced spraying or using alternative controls. We aim to deliver resistance data directly to growers. We will address three objectives: 1) pilot field and laboratory

protocols, 2) measure baseline pesticide resistance in avocado thrips, and 2) communicate results and strategies to reduce resistance to avocado industry stakeholders.

#### Work Plan & Methods

#### Pilot field and lab protocols (July 2025 – Oct 2025)

One of the primary challenges for implementing this project is that thrips are challenging to rear in a lab setting. Because thrips populations are lower in the late summer and Fall, we will utilize Year 1 of the project to trial the a) best methods for field collecting thrips, including how to transport thrips and store them prior to lab bioassays, and b) best methods for lab bioassays, including pesticide preparations, rearing receptacles, and timing and conducting mortality assessments. Protocols will be modified from existing literature (Morse et at. 2006). Cohen, Cass, and Leger will trial both field and lab protocols in Fall 2025 during secondary flush events. Year 1 of the project will also be used to identify 4-6 participating study sites for specimen collections and to train a Cooperative Extension Staff Research Associate (SRA) on field and lab protocols for project support.

## *Measuring resistance levels (Nov 2025- Oct 2026, Nov 2026- Oct 2027, Nov 2027- Oct 2028, Nov 2028 – Oct 2029)*

In Years 2-5 we will implement resistance monitoring at 4-6 sites and collect thrips twice annually at each site for resistance testing. Our initial goal is to monitor resistance for four growing seasons at the same set of sites to characterize base resistance levels across the region. In Years 4 and 5 of the project we will expand field collections to include additional grower sites based on grower interest and demand for diagnostic services – we should be able to assess resistance and compare resistance levels to baseline data. Field collections will occur during avocado flush in the early Spring in the Fall during secondary flushes. Field collection involves sealing young leaves with thrips into plastic bags, then storing in the fridge for a maximum of 24 hours before the lab bioassay. To conduct the lab bioassay, we will collect young avocado leaves with no prior pesticide exposure, treat them with different pesticide concentrations using a handheld stainless steel sprayer, and place them inside plastic modified Munger cells with 10-15 females second instar thrips in each cell. We will include a control with no exposure to pesticide. Munger cells will be kept at  $25C \pm 1$  °C with a 14:10 h light: dark photoperiod. We will assess thrips mortality after 48hr. under the microscope by counting thrips not exhibiting movement.

#### Analysis

We will calculate mortality of thrips as the number of thrips surviving after treatment, adjusted by the number of thrips in the control (Immaraju et al. 1990) as follows:

Adjusted % mortality = 
$$\frac{(PR \times C) - PS}{(PR \times C)} \times 100$$

Where PR is the number of thrips before treatment, PS is the number of thrips after treatment, and C is the number of thrips in the control after treatment. Probit analysis will be used to quantify the lethal concentration of abamectin that generates 50% mortality (LC50) in the population. Bioassays with control mortality >20% will be omitted from analysis. We will calculate resistance ratios for each avocado field site using the most susceptible LC50 value for abamectin.

#### Project Outreach

Starting in the second year of the project, we will work closely with the California Avocado Commission to share research progress and results to growers 1) annually through an oral presentation (e.g. at a field day or workshop) and 2) through an online, interactive web-based resource of resistance data (Figure 1). We will use ArcGIS Story Maps to share with growers the number of specimens tested for resistance in each year and categorical and numerical levels of resistance. The identity and location of participating growers will be anonymized by jittering data points, i.e. using an algorithm to provide random noise and displace locations while still preserving the pattern of the dataset. The UC ANR website hosting this map will include information on management practices that can prevent pesticide resistance, such as preventing product degradation, adjusting the pH of spray solutions, and timing applications. We will evaluate grower utilization and understanding of this data with a survey that will inform the continuation of this project. The baseline resistance data from this project can serve as a reference point for diagnostic assays provided to the grower community in the future.

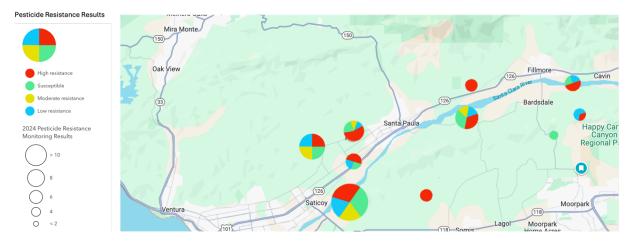


Figure 1. An example of the type of map that we can generate with ArcGIS Story Map for Ventura County resistance monitoring of avocado thrips. Each point on the map reflects categorical resistance levels and the number of thrips collected. Farm location will be anonymized by an algorithm that randomly moves the center of each sampling point to another. Each point on the map can be clicked on to obtain detailed, non-categorical number data about resistance levels.

| Outcome                         | Year 1    | Year 2   | Year 3   | Year 4   | Year 5   | Budget   |
|---------------------------------|-----------|----------|----------|----------|----------|----------|
|                                 | (July 25- | (Nov 25- | (Nov 25- | (Nov 25- | (Nov 25- | _        |
|                                 | Oct 25)   | Oct 26)  | Oct 26)  | Oct 26)  | Oct 26)  |          |
| Identify field sites, develop   | X         |          |          |          |          | \$9,411  |
| methods, obtain equipment       |           |          |          |          |          |          |
| Sample insects for establish    |           | X        | Х        |          |          | \$13,139 |
| baseline resistance levels      |           |          |          |          |          |          |
| Expand field collection sites   |           |          |          | Х        | Х        | \$5100   |
| and provide diagnostic services |           |          |          |          |          |          |
| Share results to growers at a   |           | X        | Х        | X        | X        | \$1,200  |
| field day or seminar            |           |          |          |          |          |          |
| ArcGIS Story Map                |           | Х        | Х        | Х        | Х        | \$2,610  |
| Publication                     |           |          |          | Х        |          | \$2,000  |

#### Milestone Table

#### **Budget & Budget Justification:**

| UC ANR (Cohen & Faber)   | Year 1<br>(July 25- | Year 2<br>(Nov 25- | Year 3<br>(Nov 25- | Year 4<br>(Nov 25-Oct | Year 5<br>(Nov 25-Oct |
|--|---------------------|--------------------|--------------------|-----------------------|-----------------------|
|  | Oct 25)             | Oct 26)            | Oct 26)            | 26)                   | 26)                   |
| Hand-held pesticide sprayer (B&G)  | \$450               |                    |                    |                       |                       |
| Munger Cells for bioassay  | \$150               |                    |                    |                       |                       |
| Misc. field & lab supplies (e.g. nitrile<br>gloves, distilled water, pesticide<br>product, beakers, fine sable brush,<br>forceps, aspirator, paraffin) | \$500               |                    |                    |                       |                       |
| Staff Research Associate (SRA, 10hr<br>in Year 1, 50hr Year 2-5 at \$51/hr for<br>salary + fringe)   | \$510               | \$2,550            | \$2,550            | \$2,550               | \$2,550               |
| Extension materials (printing, food for grower events)   |                     | \$300              | \$300              | \$300                 | \$300                 |
| ArcGIS Story Mapping and website<br>support from UC IGIS Center  |                     | \$1,260            | \$450              | \$450                 | \$450                 |
| Total  | \$1,610             | \$4,110            | \$3,300            | \$3,300               | \$3,300               |
|  |                     |                    |                    | TOTAL                 | \$15,660              |

| UC Riverside (Cass & Leger)   | Year 1 (July<br>25-Oct 25) | Year 2<br>(Nov 25-<br>Oct 26) | Year 3<br>(Nov 25-<br>Oct 26) | Year 4<br>(Nov 25-Oct<br>26) | Year 5<br>(Nov 25-Oct<br>26) |
|---|----------------------------|-------------------------------|-------------------------------|------------------------------|------------------------------|
| Travel from UC Riverside to<br>Ventura with vehicle (\$400) and 3<br>nights overnight stay each year<br>(\$200 x 3=\$600) | \$1,000                    | \$1,000                       | 000 20)                       | 20)                          | 20)                          |
| Postdoctoral Salary + Fringe<br>(1 month/annually)  | \$6,801                    | \$7,039                       |                               |                              |                              |
| Publication costs   |                            |                               |                               | \$2,000                      |                              |
| Total   | \$7,801                    | 8,039\$                       |                               | \$2,000                      |                              |
|   |                            |                               |                               | TOTAL                        | \$17,840                     |

Support from CAC is critical for the success of this project, which is currently unfunded. The research team includes early-career UCCE researchers proposing to advance integrated pest management of a key pest of avocado. Our budget includes requests for materials, labor, and travel.

Materials: In Year 1 of the project, we are requesting support for materials which can be used throughout the project duration, including materials to create munger cells, a small hand-held sprayer, and safety equipment such as gloves.

Labor: Because Year 1 only includes a few months, we are requesting only 10 hours of field work support for our staff research associate (SRA) at UC ANR to collect specimens. In Years 2-5, we are asking for 50 hours of field work support each year. The SRA will also assist in setting

up the bioassay and monitoring for mortality. We are requesting funds for our UCR postdoctoral researcher, Laura Leger, to travel to Ventura help us refine our bioassay in Year 1. In Year 2, Dr. Leger will work on designing and maintain the data management infrastructure for this project, analyzing preliminary data, writing reports, and disseminating results.

Mapping: In Year 2 of the project, Dr. Leger will work with the UC Informatics and GIS Center (IGIS) to develop the ArcGIS Story Map for this project. IGIS have provided a project estimate of 14 contracted hours for this project at \$90/hr. In Years 3-5, we are requesting 5 hours of each year for IGIS support in managing our map and providing refinement the map design.

Dissemination: In Years 2-5 we will host an annual presentation to update growers on our progress. We will use funds to provide lunch. In Year 4 we are requesting funding support to publish results in a peer-reviewed journal as we this data will additionally be of interest to the scientific community.

#### **D.** Curriculum Vitae or Resume:

#### Roles and Contribution:

Principal Investigator Cohen will serve as project leader and manager, overseeing day-to-day operations of the experiments, including communication with the participating growers, adhering to the project timeline, reporting deliverables, and organizing outreach activities. Co-PIs Cass, and Leger will be responsible for conducting laboratory work, participating in data analysis/interpretation, writing reports, and speaking at extension events. All team members will contribute to experimental design, project implementation in the field, data management, and report writing.

#### HAMUTAHL COHEN

Entomology Advisor University of California Cooperative Extension hcohen@ucanr.edu

#### **EDUCATION and POSITIONS**

| 2023-current | Extension Advisor, Entomology, University of California                   |
|--------------|---|
| 2021-2023    | Extension Agent, Commercial Horticulture, University of Florida IFAS      |
| 2018-2021    | Postdoctoral Researcher, Entomology, University of California, Riverside  |
| 2012-2018    | Ph.D., Environmental Studies, University of California, Santa Cruz        |
| 2009-2011    | B.S., Molecular Environmental Biology, University of California, Berkeley |

#### **SELECTED PUBLICATIONS (last 5 years)**

- Tsang, T. P., De Santis, A. A., Armas-Quiñonez, G., Ascher, J. S., Ávila-Gómez, E. S., Báldi, A., ...**Cohen, H**.... & Bonebrake, T. C. (2025). Land Use Change Consistently Reduces α-But Not β-and γ-Diversity of Bees. *Global Change Biology*, *31*(1), e70006.
- Smith, G.P., Cohen H., Zorn, J.F., McFrederick, Q.S., Ponisio, L.C. (2024). Plantpollinator network architecture does not impact intraspecific microbiome variability. *Molecular Ecology*. 33(7), e17306
- Ponisio, L.C., Cohen, H., Galbraith, S.M., Zorn, J.F., Zitomer, R.A., Rivers, J.W. (2023). Host and floral communities shape parasite prevalence and reproduction in intensively managed forests. *Ecosphere*. 15(1), e4709. doi: 10.1002/ecs2.4709
- Jha, S., Egerer, M.H., Bichier, P., Cohen, H., Liere, H., Lin, B.B., Lucatero, A., Philpott S.M. (2023). Multiple ecosystem service synergies and landscape-mediation of biodiversity in urban agriculture. *Ecology Letters*. 26(3), 369-383.
- Ong, T.W., Lin, B.B., Lucatero, A., Cohen, H., Bichier, P., Egerer, M.H., Danieu, A., Sha, J., Philpott, S.M., Lieri, H. (2022). Rarity begets rarity: Social and environmental of rare organisms in cities. *Ecological Applications*
- Cohen, H., Ponisio, L.C., Russell, K., Philpott, S.M., McFrederick, Q.M. (2022). Floral resources shape parasite and pathogen dynamics in bees facing urbanization. *Molecular Ecology* doi: 10.1111/mec.16374
- Ivers, N.A., Jordan, Z., Cohen, H., Tripodi, A., Brown, M.J.F., Lieri, H., Lin, B.B., Philpott, S., Jha, S. (2022). Parasitism of urban bumble bees influenced by pollinator taxonomic richness, local garden management, and surrounding impervious cover. Urban Ecosystems doi: 10.1007/s/11252-022-01211-0
- **Cohen, H.,** Egerer, M.H., Thomas, S-S., Philpott, S.M. (2022) Local and landscape features constrain the trait and taxonomic diversity of urban bees. *Landscape Ecology*
- Cohen, H., Smith, G., Zorn, J.F., Sardinas, H., McFrederick, Q.M., Woodard, S.H., Ponisio, L.C. (2021). Mass-flowering monoculture attracts bees, amplifying parasite prevalence. *Proceedings of the Royal Society B: Biological Sciences*. 288(1960)
- **Cohen, H.,** Philpott, S.M., Lin, B.B., Liere, H. & Jha, S. (2021). The relationship between pollinator community and pollination services is mediated by floral abundance in urban landscapes. *Urban Ecosystems*. 24(2), 275-90.
- **Cohen, H.,** McFrederick, Q.M., Philpott, S.M. (2020). Environment shapes the microbiome of the Blue Orchard Bee, *Osmia lignaria*. *Microbial Ecology*.

- **Cohen, H.,** Philpott, S.M., Lin, B.B., Liere, H. & Jha, S. (2020). Increasing pollinator abundance without enhancing diversity leads to pollination service dilution in floral-rich urban gardens. *Urban Ecosystems*. doi: 10.1007/s11252-020-01024-
- O'Connell, M., Jordan, Z., McGilvay, E., **Cohen, H.,** Cohen, R., Liere, Lina, B.B., Philpott, S.M., H., Jha, S, (2020). Reap what you sow: Local plant species composition drives pollinator foraging within urban garden landscapes. *Urban Ecosystems*.
- Egerer, M., Cecala, J., **Cohen, H.** (2020). Wild bee conservation within urban gardens & nurseries: effects of local & landscape management. *Sustainability*. 12(1), 293
- Philpott, S.M., Egerer, M.H., Bichier, P., Cohen, H., Cohen, R., Liere, H., Jha, S, & Lin, B.B. (2020) Gardener demographics, experience, and motivations drive differences in plant species richness and composition in urban gardens. *Ecology & Society*

#### **COMMUNICATION & EXTENSION**

**Presentations at Professional Meetings (2014-current):** 11 oral presentations & 10 posters at conferences such as Entomological Society of America & Ecological Society of America

Extension Classes (2020-current): >80 in-person and virtual presentations for the crop growers, horticultural professionals, pesticide applicators, and the public on topics related to bee conservation, agricultural sustainability, and IPM in commercial horticulture, subtropical crops, and vegetable production

**Extension Materials (2020-current):** 16 articles in blogs, newsletters, & magazines, 8 factsheets, 261 phone calls, walk-ins, e-mails, & visits, 927 followers on social media

#### **SELECTED GRANTS (last 5 years)**

**co-PI** CDFA Biologically Integrated Farming Systems "Improving sustainability of diamondback moth management in cruciferous vegetables" (2025-2030, \$1,000,000, subaward \$135,026)

**co-PI** Thelma Hansen Foundation "The effect of micro-sprinkler irrigation on predatory and pest mite populations in strawberry" (2024-2026, \$23,122)

**co-PI** Thelma Hansen Foundation "Influence of nitrogen application on western flower thrips populations in gerbera daisy production" (2024-2026, \$24,952)

**co-PI** Hrdy Foundation "Evaluation of climatic drivers of citrus pests using grower data" (2024-2028, \$125,000)

**co-PI** Agricultural Research Institute "Integrating vegetation into landscape scale pest management practices" (2023-2025, \$107,198, subaward \$28,000)

**co-PI** Agricultural Research Institute IPM "Landscape scale pest management practices in citrus and avocado" (2022-2024, \$803,635, subaward \$37,875)

PI Extension Foundation – USDA NIFA "Climate-smart landscapes" (2022, \$5,000)

#### LAURA LEGER

#### Postdoctoral Researcher Subtropical Fruit IPM Lab University of California Riverside, Department of Entomology Email: llege001@ucr.edu

| Educa                | ntion                           |  |  |  |  |  |
|----------------------|---------------------------------|--|--|--|--|--|
| 2017-2               |                                 | Ph.D. in Entomology  |  |  |  |  |
|                      |                                 | University of California, Riverside; Advisor: Dr. Quinn McFrederick                |  |  |  |  |
| 2015-2               | 2017                            | B.S. in Entomology   |  |  |  |  |
| 2010                 | 2015                            | University of California, Riverside  |  |  |  |  |
| 2010-2               | 2015                            | General Breadth requirements   |  |  |  |  |
|                      |                                 | Long Beach City College  |  |  |  |  |
| <u>Resea</u>         | rch Positions                   |  |  |  |  |  |
| 2024 -               | Present                         | Postdoctoral Researcher  |  |  |  |  |
|                      |                                 | University of California, Riverside; Advisor: Dr. Bodil Cass                       |  |  |  |  |
| Dubliz               | nations                         |  |  |  |  |  |
|                      | <u>cations</u><br>Leger, L., Da | rrow, C., Lam, C., Fournier, N., Vo, H., McFrederick, Q.S., "Bumble Bees           |  |  |  |  |
|                      |                                 | Multiple Simultaneous Stressors." In preparation.                                  |  |  |  |  |
| 2.                   |                                 | m, C., Melkonyan, M., Kyurklyan, R., Palmer-Young, E.C., and                       |  |  |  |  |
|                      | 0                               | Q.S., "Bumble Bee Parasite Strains Show Variation in Resistance to                 |  |  |  |  |
|                      |                                 | Remove Cadmium from their Growth Environment," In preparation.                     |  |  |  |  |
| 3.                   |                                 | , Russell, K.A., Leger, L., McFrederick, Q.S. and Graystock, P., (2020).           |  |  |  |  |
| 5.                   |                                 | ind indirect effects of environmental toxicants on the health of bumble bees       |  |  |  |  |
|                      |                                 | obiomes." <i>Proceedings of the Royal Society B: Biological Sciences</i> .         |  |  |  |  |
| 1                    |                                 |  |  |  |  |  |
| 4.                   | -                               | ner-Young, E.C., Nevarez, R.B., Russell, K.A., <b>Leger, L.</b> , Giacomini, S.J., |  |  |  |  |
|                      | -                               | go, M.S., Irwin, R.E. and McFrederick, Q.S., (2020). Cross-infectivity of          |  |  |  |  |
| 5                    | •                               | mble bee-associated parasites across three bee families. <i>Parasitology</i> .     |  |  |  |  |
| 5.                   |                                 | McFrederick, Q.S. (2020). "The Gut-Brain-Microbiome Axis in Bumble                 |  |  |  |  |
| <i>.</i>             | Bees." Insects                  |  |  |  |  |  |
| 6.                   |                                 | A., Leger, L., Kirkwood, J. S., & McFrederick, Q. S. (2019). "Cadmium and          |  |  |  |  |
|                      | -                               | sure affect the honey bee microbiome and metabolome, and bee-associated            |  |  |  |  |
|                      |                                 | potential for bioaccumulation." Applied and environmental microbiology.            |  |  |  |  |
| 7.                   |                                 | A., Leger, L., Graystock, P., Russell, K., & McFrederick, Q. S. (2019). "The       |  |  |  |  |
|                      | bumble bee m                    | nicrobiome increases survival of bees exposed to selenate                          |  |  |  |  |
|                      | toxicity." Env                  | ironmental microbiology.   |  |  |  |  |
| Follow               | vships, Grants                  | and Awards   |  |  |  |  |
| <u>renov</u><br>2022 |                                 | , and Awards<br>Research Grant   |  |  |  |  |
| 2022                 |                                 | California, Riverside; \$1900  |  |  |  |  |
| 2022                 |                                 |  |  |  |  |  |

2022 Dissertation Year Program Fellowship University of California, Riverside

| 2019 | National Science Foundation                               |
|------|---|
|      | Graduate Research Fellowship Program; \$134,000           |
| 2018 | Entomological Society of America Joint Annual Meeting     |
|      | Student Competition: 8 Minute Talk; 2 <sup>nd</sup> Place |
| 2017 | Deans Distinguished Fellowship Award                      |
|      | University of California, Riverside                       |
| 2017 | National Science Foundation                               |
|      | Graduate Research Fellowship Program; Honorable Mention   |
| 2016 | University Mini Grant for Undergraduate Students          |
|      | University of California, Riverside; \$700                |

#### BEN A. FABER

Soils/Water/Subtropical Horticulture Advisor University of California Cooperative Extension 669 County Square Dr. Ventura, CA 93003-5404 Phone: 805-645-1462 Fax: 805-645-1474 bafaber@ucanr.edu

#### **EDUCATION**

Ph.D. Soil Fertility, University of California, Davis. 1989 M.S. Soil Fertility, University of California, Davis. 1986 A.B. Biology, University of California, Santa Cruz. 1973

#### RESEARCH

Research experience in pest management, plant nutrition and soil management. Current research on irrigation requirements of avocado and citrus, methods of controlling groundwater nitrate pollution, effects of yardwaste mulches on citrus production, pest and disease management in avocado and citrus, citrus rootstock evaluation and citrus weed management.

#### SELECTED RECENT PUBLICATIONS

Lu, Jianhang; Wu, Laosheng; Newman, Julie; <u>Faber, Ben</u>; Gan, Jianying. 2006. Degradation of pesticides in nursery recycling pond waters. J. Agric. Food Chem. 54: 2658-2663.

Lu, J., L.; Wu, J. Newman, <u>B. Faber</u>, D. Merhaut, and J. Gan. 2006. Sorption and Degradation of Pesticides in Nursery Recycling Ponds. J. Env. Quality.35: 1795-1802

Grafton-Cardwell, E.E., 20 authors and <u>B. Faber</u>. 2006. Citrus leafminer. <u>http://www.ipm.ucdavis.edu/PMG/r107303211.html</u>. UC-IPM Guidelines. UC-ANR.

Faber, B.A., G.S. Bender, H.D. Ohr and J.A. Menge. 2007. Avocado-Diseases. UC IPM Pest Management Guidelines. UC ANR Pub 3436.

Phillips, P.A., <u>B.A. Faber</u>, J.G. Morse and M.S. Hoddle. 2007. Avocado-Insects and Pests. UC IPM Management Guidelines. UC ANR Pub 3436.

Faber, B.A., A.J. Downer, D. Holstege and M.J. Mochizuki. 2007. Accuracy varies for commercially available soil test kits analyzing nitrate-nitrogen, phosphorus, potassium and pH. HortTechnology 17(3): 358-362.

Crohn, David M., <u>Ben Faber</u>, A. James Downer and Oleg Daugovish. 2007. Probabilities for survival of glassy-winged sharpshooter and olive fruit fly pests in urban yard waste piles. Bioresource Technology. 99(5): 1425-32.

Grafton-Cardwell, E. E., D. A. Headrick, K. E. Godfrey, J. N. Kabashima, <u>B. A. Faber</u>, V. F. Lazaneo, P. A. Mauk, and J. G. Morse. 2007. Citrus Leafminer. UC IPM Pest Note 74137. 4 pp

Downer, A. J., D. Crohn, <u>B.Faber</u>, O. Daugovish, J.O. Becker, J.A. Menge, and M.J. Mochizuki. 2008. Survival of plant pathogens in static piles of ground green waste. Phytopathology 98:547-554.

Newman, J., S. Mangiafico, D. Merhaut, L. Wu, J. Lu, D. Haver, <u>B. Faber</u> and J. Gan. 2012. Mitigating pesticide runoff from nurseries. *In:* Pesticide mitigation strategies for surface water quality, K. Goh, B. Bret, T. Potter, and J. Gan. A. Chemical Soc. Vol. 1075. pp. 77-94.

Joy, M., M. Abit, D.L.Shaner, L.J. Krutz, C.M. Rainbolt, N.V. O'Connell, <u>B.A. Faber</u> and B.D. Hanson. 2012. Assessing simazine degradation patterns in California citrus orchards with different simazine histories. Air, Soil and Water Research (5) 69-78.

Eskalen, A., <u>B. Faber</u> and M. Bianchi. 2012. Spore trapping and pathogenicity of fungi in the Botryosphaeriaceae and Diaporthaceae associated with avocado branch canker in California. Plant Disease. Posted online at First Look 6 Sep, 2012.

Dreistadt, S., J. Clark, D. Martin, K. Al-Khatib, J. Strand, P. Goodell, J. Stapleton, E. Grafton-Cardwell, N. O'Connell, P. Phillips, J. Morse, <u>B. Faber</u>, J. Adaskaveg and A. Eskalen. 2012. Integrated Pest Management of Citrus. UC ANR 3303.

<u>Faber, B.A.</u>, and C.J. Lovatt. 2013. Use of foliar fertilization to offset effects on navel orange yield due to reduced water and fertilizer applied by partial root zone drying versus conventional irrigation. Proc.7<sup>th</sup> Int. Symp. Mineral Nutrition of Fruit Crops, Thailand 2012. pp.237-246.

Crane, J.H., G. Douhan, <u>B.A. Faber</u>, M.L. Arpaia, C.F. Balardi and A.F. Barrientos-Priego. 2013. Cultivars and Rootstocks. *In:* The Avocado: Botany, Production and Uses. Eds. Schaffer, Wolstenholme and Whiley. CABI. Pp. 247-278.

<u>Faber, B.A.</u> and C.J. Lovatt. 2014. Effects of applying less water by partial root zone drying versus conventional irrigation on navel orange yield. Acta Hort (ISHS) 1038: 523-530.

Faber, B. 2015. Irrigating citrus with limited water. UC Pub 8549. Pp. 6.

Vargas, R, S. Souder, J. Morse, E. Grafton-Cardwell, D. Haviland, <u>B. Faber</u>, B. Mackey, and P. Cook. 2015. Captures of wild Ceratitis capitate, Bactrocera dorsalis and Bactrocera cucurbitae (Diptera: Tephritidae) in traps with improved multilure TMR dispensers weathered in California. J. Econ Ent. 108(5): 34-4

#### E. References cited

Ávila-Quezada, G. D., Téliz-Ortiz, D., Vaquera-Huerta, H., González-Hernández, H., & Johansen-Naime, R. (2005). Progreso temporal del daño por trips (Insecta: Thysanoptera) en aguacate (Persea americana Mill.). *Agrociencia*, *39*(4), 441-447.

Gao, Y., Lei, Z., & Reitz, S. R. (2012). Western flower thrips resistance to insecticides: detection, mechanisms and management strategies. *Pest management science*, *68*(8), 1111-1121.

Goldarazena, A., Gattesco, F., Atencio, R., & Korytowski, C. (2012). An updated checklist of the Thysanoptera of Panama with comments on host associations. *Check List*, *8*(6), 1232-1247.

Hoddle, M. S., Morse, J. G., Phillips, P. A., Faber, B. A., & Jetter, K. M. (2002). Avocado thrips: new challenge for growers. *California Agriculture*, *56*(3).

Humeres, E. C., & Morse, J. G. (2006). Resistance of avocado thrips (Thysanoptera: Thripidae) to sabadilla, a botanically derived bait. *Pest Management Science: formerly Pesticide Science*, *62*(9), 886-889.

Immaraju, J. A., Morse, J. G., & Brawner, O. L. (1990). Evaluation of three bioassay techniques for citrus thrips resistance and correlation of the leaf dip method to field mortality. *J. Agric. Entomol*, *7*(1), 17-27.

Morse, J., Urena, A., Humeres, E., Robinson, L., Flores, P., & Watkins, P. (2006). Biology, management, and resistance monitoring of avocado thrips and persea mite. In *Proceedings, California Avocado Research Symposium* (pp. 16-24). Santa Ana, CA: California Avocado Commission.

Rugg, D., Buckingham, S. D., Sattelle, D. B., & Jansson, R. K. (2005). The insecticidal macrocyclic lactones.

#### **Bodil N. Cass**

Assistant Extension Specialist, Subtropical Fruit IPM Lab University of California Riverside, Department of Entomology <u>bodil.cass@ucr.edu</u> | 951-827-4454

#### Education

| University of Queensland | Brisbane, Australia | Genetics   | B.Sc. Hons., 2006 |
|--------------------------|---------------------|------------|-------------------|
| University of Arizona    | Tucson, AZ          | Entomology | Ph.D., 2015       |

#### Appointments

| 2023-present | Assistant Cooperative Extension Specialist, Department of Entomology, University of California-Riverside        |
|--------------|---|
| 2021-2023    | <b>Agricultural Scientist – Entomology</b> , Department of Agriculture, Weights & Measures, County of San Diego |
| 2016-2020    | <b>Postdoctoral Scholar,</b> Department of Entomology & Nematology, University of California, Davis             |

#### **Selected Publications**

Rosenheim JA, Cluff E, Lippey MK, Cass BN, Paredes D, Parsa S, Karp DS, Chaplin-Kramer R. 2022. Increasing crop field size does not consistently exacerbate insect pest problems. *Proceedings of the National Academy of Sciences* 119(37):e2208813119

- Kahl HM, Mueller TG, Cass BN, Xi X, Cluff E, Rosenheim JA. 2022. Herbivory by European Earwigs (*Forficula auricularia*; Dermaptera: Forficulidae) on *Citrus* Species Commonly Cultivated in California. *Journal of Economic Entomology* 115(3):852–862.
- Kahl HM, Mueller TG, Cass BN, Xi X, Cluff E, Grafton-Cardwell EE, Rosenheim JA. 2021. Characterizing herbivory by European Earwigs (Dermaptera: Forficulidae) on navel orange fruit with comparison to Forktailed Bush Katydid (Orthoptera: Tettigoniidae) herbivory. *Journal of Economic Entomology* 114(4):1722-32
- Cass BN, Kahl HM, Mueller TG, Xi X, Grafton-Cardwell EE, Rosenheim JA. 2021. Profile of fork-tailed bush katydid (Orthoptera: Tettigoniidae) feeding on fruit of clementine mandarins. *Journal of Economic Entomology* 114 (1), 215-224
- Cass BN, Hack LM, Buckmann D, Mueller TG, Grafton-Cardwell EE, Rosenheim JA. 2020. Arthropod infestation levels on mandarins in California. *Journal of Economic Entomology* 113(5):2335–2342
- Rosenheim JA, Cass BN, Kahl HM, Steinmann KP. 2020. Variation in pesticide use across crops in California agriculture: economic and ecological drivers. *Science of the Total Environment* 733:138683
- Cass BN, Grafton-Cardwell EE, Rosenheim JA. 2019. Resistance of fruits from a mandarin cultivar to feeding by fork-tailed bush katydids. *Journal of Economic Entomology* 112(6):2861–2871

- Mueller TG, Kahl HM, Cass BN, Grafton-Cardwell EE, Rosenheim JA. 2019. Differential impacts of citrus thrips across sweet orange and mandarin species. *Journal of Economic Entomology* 112(6):2767–2773
- Cass BN, Hack LM, Grafton-Cardwell EE, Rosenheim JA. 2019. Impacts of fruit-feeding arthropod pests on oranges and mandarins in California. *Journal of Economic Entomology* 112(5):2268–2277

#### Select recent extension presentations

- "Exploring Predators for Control of Banks Grass Mite in Dates", Citrus and Date Palm Seminars, Phoenix, AZ and Yuma, AZ, 12/2024
- "Determining ecological drivers of citrus thrips pressure using Ecoinformatics", UC ANR Fall Citrus Meeting, Exeter, CA, 10/2024
- "Determining the drivers of citrus pest populations for more region-specific management" California Citrus Conference, Citrus Research Board, Visalia, CA 10/2024
- "New Research at the Subtropical Fruit IPM Lab", Research Committee Meeting, Plant California Alliance, Irvine, CA, 09/2024
- "Citrus Pest Alerts", Citrus Roundtable, Association of Applied IPM Ecologists, Tulare, CA, 08/2024

"Avocado pest alerts", Avocado Field Day, California Avocado Society, Ventura, CA, 08/2024

- "Fruit Fly Invasions: What's going on in SoCal?" CAPCA, Ventura, CA, 07/2024 & Riverside, CA, 08/2024
- "Managing Insect Pests of Fruit Trees" 2024 Landscape IPM Workshop, UC Cooperative Extension and Port of San Diego, San Diego, CA, 06/2024
- "A new *Caloptilia* pest of avocados", Avocado Winter Workshop, California Avocado Society, Ventura, CA, 02/2024
- "Current Pest Alerts for Southern California", Professional Tree Care Association 33rd Annual Seminar and Field Day, San Diego, CA 08/2022

#### Synergistic activities

Member, Asian Citrus Psyllid Technical Review Team

Co-editor, UC ANR Topics in Subtropics quarterly grower newsletter

Reporting UC member, CDFA Citrus Pest and Disease Prevention Division Outreach Subcommittee



#### California Avocado Commission PROJECT PLAN / RESEARCH GRANT REQUEST FOR PROPOSAL

**<u>Project Title:</u>** Evaluating diverse avocado rootstocks for salinity using morphological, ionic, and physiological parameters

**Project Lead:** Jorge F.S. Ferreira, US Salinity Laboratory (USDA-ARS), 450 W Big Springs Rd., Riverside, CA 92507, Jorge.Ferreira@usda.gov,951-369-4832

*Co-PI*: Devinder Sandhu, US Salinity Laboratory (USDA-ARS), 450 W Big Springs Rd., Riverside, CA 92507, <u>Devinder.Sandhu@usda.gov</u>, 951-289-3627

Project Cooperator: Consuelo Fernandez, Brokaw Nursery LLC, Ventura, CA 93004, consuelof@brokawnursery.com, 805-218-3702

#### **Executive Summary:**

Water is the most limiting factor for any crop cultivated in semi-arid regions. California avocado-growing regions rely on a combination of Colorado River water, groundwater, and local reservoirs. Colorado water salinity has steadily increased to reach 1.0 dS/m (TDS = 700 mg/L) in California, and groundwater salinity can be even higher. Water salinity of 1.5 dS/m can kill avocado plants in less than two years but lower salinities, although symptomless, significantly decrease fruit yield and reduce farmers' profits and make California even more dependent on foreign fruits to meet its market demand. We propose to evaluate avocado rootstocks for salinity tolerance when irrigated with saline waters of different ionic compositions and dominant in Na<sup>+</sup>, Cl<sup>-</sup>, and SO<sub>4</sub>. Rootstock evaluation will be based on the leaf accumulation of those different ions (as minerals), the trunk diameter one year after plants have been irrigated with saline water, and physiological parameters (stomatal conductance, internal  $CO_2$ , and photosynthesis) measured in physiologically mature leaves. Ionic composition and balance are directly related to photosynthesis, nutritional balance, and growth, with excessive Na and Cl being the main culprits. Our experience of 8 years screening almond rootstocks clearly shows that efficiency in extruding Na<sup>+</sup> and Cl<sup>-</sup> is highly correlated with salttolerance index and survival. This also holds for crops that do not require rootstocks (strawberry, alfalfa, passion fruit, Jerusalem artichoke, etc.). Because there is no salinity tolerance database for avocado rootstocks, the information we will generate in this project will be extremely valuable for nurseries and farmers to determine which rootstock is more resilient to their water salinity.

#### **Background:**

Southern California avocado growers have more water available to their crops than pistachio and almond growers in the Central Valley. However, southern avocado farmers rely heavily on Colorado River water and groundwater, while northern avocado farmers rely on groundwater and local reservoirs (Spann, 2024). In 2023, the United States imported 81% of the total avocado yield from Mexico, valued at US\$ 2.84 billion, followed by Canada, Japan, and Spain (<u>https://apps.fas.usda.gov</u>). Mexico's 2024 fresh avocado yield exceeded two million metric tons (<u>https://www.statista.com/</u>) while California avocado yield in 2023/24 reached 164,926.2 metric tons (363.6 million pounds). Although much smaller than Mexico's yield, California's crop reached a value of US\$ 523,817,252,

according from the California Avocado Growers to report а (https://www.californiaavocadogrowers.com/industry/industry-statistical-data). This same report shows that, although the planted acres in 2023/24 were 1,200 acres more than in 2021/23, the average pounds of avocado per bearing acre increased from approximately 6,000 (2021/22) to 7,580 (2023/24). In Southern California, the water year (July 2022 - June 2023) had 28.4 inches (721 mm/year) of rain and the 2023/24 water year had 25.2 inches (640 mm/year) with the average rainfall recorded in downtown Los Angeles being approximately 14.25 inches/year (362 mm/year), but not evenly distributed through the year. Even if it were, irrigation below 540 mm/year is the threshold under which relative fruit yield starts to decrease (Kourgialas and Dokou, 2021). Thus, the short rain season of southern California requires supplemental irrigation, even on a wet year, with water from the Colorado River (TDS = 700 mg/L or  $EC_w = 1 \text{ dS/m}$ ) and from underground ( $EC_w$  can reach 2 dS/m or higher) (https://www.waterboards.ca.gov/gama/docs/coc salinity.pdf). Also, the water and nutrients must be evenly distributed throughout the crop cycle to guarantee the highest fruit-yield potential of each tree. The average California fruit yield per acre in the past four years was 6,000 lbs./acre, while the 4-year average for Mexico was 16,245 lbs./acre, with good years producing 20-25,000 lbs./acre, three- to four-fold higher than in Southern California. Although fruit yield depends on climate, soils with good drainage, pollinators, and good rootstocks and genotypes, Mexican farmer can count on 1,200 mm/year of evenly distributed rain (https://apps.fas.usda.gov). However, despite the fact that precision agricultural practices, such as AI-powered irrigation drone monitoring, and satellite imagery are growing in use, 1) avocado trees are highly sensitive to salinity, 2) the demand for avocados will continue to grow in the US and worldwide, 3) Under the currently imposed tariffs on Mexico, American consumers would benefit from a higher domestic yield, 4) irrigation-water salinity for Southern California growers (1-2 dS/m) is already over the salinity threshold ( $EC_e = 0.6$ dS/m or  $EC_w = 1.3 dS/m$ ) (Oster et al., 2007) under which fruit yield was reported to decrease significantly, and 5) our best and fastest resource to increase fruit yield under saline conditions rely on rootstocks that are more salt-tolerant. A USSL-UCR study on 13 avocado rootstocks found that, after 23 months of irrigation at  $EC_w=1.5 \text{ dS/m}$ , rootstocks with high Na and Cl accumulation had 100% mortality, while rootstocks that restricted salt transport survived (Celis et al., 2018). Subsequent research in Israel with two 'Hass'-avocado rootstocks at ECe=0.73 dS/m and Na and Cl concentrations of 22.3 and 17.6 mg/L confirmed these results (Lazare et al., 2021). Considering the reduced availability of low-salinity groundwater, the scarce and unevenly distributed precipitation in Southern California, the increase in avocado cultivation, and the increasing salinity of both Colorado River and groundwater, the use of reclaimed water seems inevitable (Harkness et al., 2023). The recent investments to expand the use of treated municipal wastewater (average  $EC_w = 1.1 \text{ dS/m or } 825 \text{ mg/L}$ TDS) in California underscores the urgent need for salt-tolerant avocado rootstocks.

Plants employ several mechanisms to cope with salinity stress and toxic ions, which include uptake or exclusion by roots, sequestration in vacuoles, root-to-shoot transport regulation, accumulation of compatible solutes, and tissue tolerance (Sandhu and Kaundal, 2018). Hence, understanding the roles of different component traits of plant salt tolerance mechanisms is critical.

Our salinity research facility is the best equipped in the U.S. to evaluate crops and rootstocks for salinity tolerance and support breeding programs to develop salt-tolerant varieties. We have extensive experience with fruit and nut crops, including almonds, peaches, and grapes. For the past eight years, we have received consistent funding from the Almond Board of California to advance rootstock and variety development for saline-affected regions. During this period, we assessed 16 commercial rootstocks and over 120 selections from USDA-ARS and UC Davis breeding programs. We demonstrated a significant variation for different components in diverse almond- rootstock genotypes. Our findings highlighted that Na, and to a lesser extent, Cl are the most critical toxic ions for almond rootstocks (Sandhu et al., 2020). Top-performing rootstocks under salinity had the lowest

concentrations of leaf and root accumulation of Na and Cl, suggesting that ion exclusion may be the main component trait of the salt tolerance mechanism in almonds. Furthermore, the expression analysis of salt tolerance genes revealed that treatments where Na and Cl were the main components of irrigation water led to the induction of most genes, suggesting the importance of both Cl and Na toxicities during salt stress in almonds.

Our previous studies with almond rootstocks provided a detailed analysis of the importance of different component traits and the knowledge generated by our approach can be readily transferred to develop a salt-tolerant rootstock database for avocadoes. Using this approach, new rootstocks can be quickly developed for vigor and tolerance under salinity, adding to other important features, such as root-rot tolerance.

The *rationale* of the study is that we identify salt-tolerant rootstocks that California avocado growers may be able to utilize the salt affected marginal lands and degraded waters for the avocado cultivation. Therefore, our approach to improving salt tolerance is very important to eventually increase the profitability of California avocado growers through higher fruit yields per acre. The resources to be created in this project will facilitate avocado research worldwide in preparation for ongoing global changes in terms of reduced availability of fresh water and increasing salinization of water resources.

# List of Project Objectives:

- 1. Screen 30 diverse avocado rootstocks for salinity tolerance based on morphological traits.
- 2. Screen 30 diverse avocado rootstocks for salinity tolerance based on ionomics.
- 3. Screen 30 diverse avocado rootstocks for salinity tolerance based on physiological parameters.
- 4. Evaluate diverse avocado rootstocks for salinity tolerance under mixed-salt combinations.

# List of Specific Project Deliverables:

- 1. Thirty avocado rootstocks will be evaluated for salinity tolerance based on morphological, ionic, and physiological parameters
- 2. Relative salt tolerance profiles for the screened rootstocks will be developed.
- 3. Correlation among morphological traits, ion tissue accumulation, and physiological parameters will be determined.
- 4. Different rootstocks will be characterized based on specific ion sensitivities by screening rootstocks under varying solution composition.
- 5. Oral and poster presentations in scientific and growers' meetings.
- 6. Articles will be published in peer-reviewed journals and commodity group magazines reporting identification of elite germplasm and characterization of mechanisms involved in salt tolerance.
- 7. Undergraduate students will be trained in field and laboratory techniques related to morphological, physiological, and ionomic responses of avocadoes to salinity stress.

# Work Plan and Methods:

**Obective 1.** Screen 30 diverse avocado rootstocks for salinity tolerance based on morphological traits

Elite rootstocks, tolerant to root-rot and for overall performance (e.g., Duke 7, Borchard, Dusa), have been developed by various nurseries and public breeding programs. We will collaborate with commercial nurseries to screen 30 elite rootstock for salinity tolerance. The experiment will be conducted at the U.S. Salinity Laboratory in Riverside, CA. One-year-old potted plants will be evaluated under field conditions with three replications using three plants per replicate (2 salts x 30

genotypes x 3 replicates x 3 plants per replicate = 540 trees). The experiment will be set up in a split-plot design, with replications as main plots, salinity levels as subplots, and genotypes as subsubplots. After transplanting into 3-gallon pots, plants will be allowed to acclimate for four weeks fertigated with municipal water and basic macro- and micronutrients ( $EC_{iw} = 1.2 \text{ dS m}^{-1}$ ). We will use mixed ion composition to represent a range of natural water compositions for our treatments, with Na<sup>+</sup> as the main cation and Cl<sup>-</sup> as the main anion (Appendix Table 1). Both control (Riverside municipal water;  $EC_{iw} = 1.2 \text{ dS m}^{-1}$ ) and moderate-salinity ( $EC_{iw} = 2.5 \text{ dS m}^{-1}$ ) treatments will have essential nutrients (Sandhu et al., 2020). Pots will be irrigated daily with pressure-compensated drippers to deliver excess water to reach a target leaching fraction of 0.3 or higher to prevent excessive salt buildup in the root zone (Ferreira et al., 2024).

Plants will be assessed for trunk diameter at the beginning and end of the experiment, 12 months later to calculate the change in trunk diameter. Also, the survival rate will be recorded, and the relative survival rate determined. The experiment will be repeated in the second year. We also plan to add rootstocks for salinity screening in subsequent years and develop a salinity-tolerance database for commercial rootstocks.

**Objective 2.** Screen 30 diverse avocado rootstocks for salinity tolerance based on ionomics

Building on the experimental setup in Objective 1, we will characterize genotypes based on ion accumulation in leaves. To understand ion uptake and homeostasis under saline conditions, ion analyses will be performed for Na, Cl, macronutrients (N, P, K, Ca, Mg, and S), and micronutrients (Fe, Mn, Mo, Co, Cu, and Zn). Samples of young and old leaves will be taken four weeks after the initiation of treatment to determine tissue ion composition. Young and old leaves will be analyzed separately to distinguish genotype allocation of toxic compounds to young leaves, as it is usually expected that toxic salts accumulate preferentially in old leaves. Tissue samples will be dried, digested in a Milestone Ethos EZ microwave digestion system, and analyzed by Inductively Coupled Plasma Optical Emission Spectrometry (ICP-OES, 3300DV, Perkin-Elmer Corp., Waltham, MA, USA) to determine the concentration of sodium, macro- and micronutrients. Chloride analysis will be carried out with a mercuric thiocyanate reaction in the presence of ferric nitrate in an AQ300 discrete analyzer (EPA600/4-79-020, 1983).

Statistical analysis will be performed with the SAS/STAT software package for analyses of variance, followed by Tukey and Tukey-Kramer pairwise comparison of means. Differences with an  $p \le 0.05$  or less will be considered significant. The relationship between change in trunk diameter and leaf-ion accumulation will be determined. We will calculate Pearson correlation coefficients to perform correlation analyses. Ion analysis will help characterize rootstocks based on their ability to regulate ions in leaves and roots, identifying key mechanisms of salinity tolerance. Correlating ion accumulation with morphological traits will provide insights into the role of ion exclusion, sequestration, and transport in stress adaptation. Understanding these relationships will enable the selection of rootstocks that minimize toxic ion buildup while maintaining growth and productivity under saline conditions. This knowledge can further facilitate development of more resilient avocado rootstocks for long-term agricultural sustainability.

**Objective 3.** Screen 30 diverse avocado rootstocks for salinity tolerance based on physiological parameters

Based on the experiment described in Objective 1, we propose determining the levels of proline, and physiological parameters such as photosynthetic activity, chlorophyll content, and stomatal conductance. These parameters will provide good insight into the mechanisms involved in salinity tolerance in avocado rootstocks.

Physiological parameters such as photosynthetic activity, chlorophyll content, and stomatal conductance will be measured at the youngest mature leaves, using a photosynthesis system (LI6400XT; LI-COR, USA). The measurements will be made between 9 and 11 am, using an artificial source of radiation (PAR of 1800  $\mu$ mol m<sup>-2</sup> s<sup>-1</sup>) and a CO<sub>2</sub> concentration of 400  $\mu$ mol mol<sup>-1</sup>. Leaf net photosynthetic rate and stomatal conductance will be measured using the LI-COR instrument 15 days after salt treatment and monthly after that. Proline will be quantified by the ninhydrin method used to measure free proline in salt-stressed plants (Huang et al., 2013). All measurements and analyses will be done in triplicates and analyzed statistically with SAS (version 9.3, SAS Institute, Cary, N.C.).

Correlating physiological parameters with morphological performance will help identify specific salinity tolerance mechanisms employed by different genotypes. Over time, these parameters may become essential tools for screening avocado germplasm for salinity tolerance. Insights gained from genotype evaluation will aid in selecting optimal parental combinations for future breeding programs, which will be crucial for understanding salt tolerance across different species and advance the development new rootstocks that will allow with higher salt tolerance and fruit yield.

**Objective 4.** Evaluate diverse avocado rootstocks for salinity tolerance under mixed-salt combinations.

Salt tolerance studies are generally undertaken using relatively simple single-salt (NaCl) or double-salt (NaCl+CaCl<sub>2</sub>) compositions. These studies are useful in that they standardize salt compositions among studies. However, for some species, especially those with low salt tolerance, the mechanism for salt tolerance is not osmotic stress but rather ion toxicity, triggered by Na or Cl toxicity, or both. Distinguishing among the specific ion toxicities can best be done using a variety of mixed salt compositions. Nine salt-tolerant rootstocks, selected based on results from Objectives 1-3, and one salt-sensitive rootstock as a control, will be evaluated in year 3 for their ion-specific response. The experimental set up will be a randomized complete block design with 3 replications, with 3 plants per replication. Irrigation waters will be designed into five treatments with different ion compositions mimicking groundwaters or Colorado River water used to irrigate avocado trees. In addition to a low-salinity control (riverside municipal water), we will use 1) a mixed cation composition (Ca=Mg=0.25 Na) with predominantly SO<sub>4</sub><sup>2-</sup> anion composition 2) a mixed cation composition (Ca=Mg=0.25 Na) with predominantly Cl<sup>-</sup> anion composition, 3) a mixed SO<sub>4</sub>-Cl anion composition with predominantly Na<sup>+</sup>, and 4) a mixed SO<sub>4</sub>–Cl anion composition with predominantly Ca<sup>2+</sup> and Mg<sup>2+</sup> cation composition, all added of a fixed NPK nutrition plus micronutrients. These mixtures will be designed to represent a range of natural water compositions.

Recent literature suggests that the more meaningful changes are the ones that take place days after initiation of stress; the changes that happen within the first few hours are normally associated with the shock response (Shavrukov, 2013). We will focus on screening genotypes at a relatively low water-salinity level ( $EC_{iw} = 2.5 \text{ dS/m}$ ) because avocados are sensitive to salinity and because at high salinity levels other mechanisms such as osmotic shock play a dominating role. These comparisons may help us dissect the mechanisms solely involved in salinity from the ones operating during osmotic shock. Different parameters analyzed will include survival rate, trunk diameter, height, leaf ion composition, stomatal conductance, and photosynthetic rate. The different parameters will be analyzed as described in Objectives 1-3.

#### **Project Outreach:**

The outcomes of this project will benefit avocado producers, public and private sector stakeholders including breeders, nurseries, and extension specialists. Results will be divulged through peer-reviewed journals, presentations for grower/stakeholder conferences, articles for *From the Grove* and reports to stakeholders, as appropriate.

| Timeline/Milestones  | Budget<br>ocation | Y | ear | r 1 | Yea | ar 2 | 2 | Yea | ır 3 | ; |
|--|-------------------|---|-----|-----|-----|------|---|-----|------|---|
| 1. Evaluation of 30 avocado rootstocks for salinity tolerance  | \$<br>70,800      |   |     |     |     |      |   |     |      |   |
| 2. Complete analysis of ions, proline,<br>chlorophyll, stomatal conductance,<br>photosynthetic rate. | \$<br>37,000      |   |     |     |     |      |   |     |      |   |
| 3. Development of a relative salt-tolerance profile for screened rootstocks.                         | \$<br>18,000      |   |     |     |     |      |   |     |      |   |
| 4. Evaluate selected avocado rootstocks under mixed-salt composition.                                | \$<br>60,000      |   |     |     |     |      |   |     |      |   |
| 5. Carry out the analysis for roles of different<br>ions during salinity stress in avocadoes         | \$<br>7,400       |   |     |     |     |      |   |     |      |   |

#### **Total budget**

#### \$ 193,200

#### References

- Celis, N., Suarez, D.L., Wu, L., Li, R., Arpaia, M.L., Mauk, P., 2018. Salt tolerance and growth of 13 avocado rootstocks related best to chloride uptake. HortScience 53, 1737-1745.
- EPA600/4-79-020 1983. Method 325.2. Chloride by Automated Colorimetry. In Methods For Chemical Analysis Of Water And Wastes (Cincinnati, OH, Environmental Protection Agency).
- Ferreira, J.F.S., Suarez, D.L., Sandhu, D., 2024. Critical questions about the origins and plant responses to soil salinity and sodicity, In: Clay, D.E., DeSutter, T.M., Clay, S.A., Nieya, T. (Eds.) Salinity and Sodicity: A Global Challenge to Food Security, Environmental Quality and Soil Resilience. John Wiley & Sons, Inc., pp. 143-155.
- Harkness, J.S., McCarthy, P.M., Jurgens, B.C., Levy, Z.F., 2023. Salinity trends in a groundwater system supplemented by 50 Years of imported Colorado River water. ACS ES&T Water 3, 3253-3264.
- Huang, Z.R., Zhao, L., Chen, D.D., Liang, M.X., Liu, Z.P., Shao, H.B., Long, X.H., 2013. Salt stress encourages proline accumulation by regulating proline biosynthesis and degradation in Jerusalem artichoke plantlets. PLOS ONE 8.
- Kourgialas, N.N., Dokou, Z., 2021. Water management and salinity adaptation approaches of Avocado trees: A review for hot-summer Mediterranean climate. Agric. Water Manag. 252, 106923.
- Lazare, S., Yasuor, H., Yermiyahu, U., Kuhalskaya, A., Brotman, Y., Ben-Gal, A., Dag, A., 2021. It takes two: Reciprocal scion-rootstock relationships enable salt tolerance in 'Hass' avocado. Plant Sci. 312, 111048.
- Oster, J.D., Stottlmyer, D.E., Arpaia, M.L., 2007. Salinity and Water Effects on 'Hass' Avocado Yields. J. Amer. Soc. Hort. Sci. 132, 253-261.
- Sandhu, D., Kaundal, A., 2018. Dynamics of salt tolerance: molecular perspectives, In: Gosal, S.S., Wani, S.H. (Eds.) Biotechnologies of Crop Improvement, Volume 3: Genomic Approaches. Springer International Publishing, Cham, pp. 25-40.
- Sandhu, D., Kaundal, A., Acharya, B.R., Forest, T., Pudussery, M.V., Liu, X., Ferreira, J.F.S., Suarez, D.L., 2020. Linking diverse salinity responses of 14 almond rootstocks with physiological, biochemical, and genetic determinants. Sci. Rep. 10, 21087.
- Shavrukov, Y., 2013. Salt stress or salt shock: which genes are we studying? Journal of experimental botany 64, 119-127.
- Spann, T. 2024. Do you have enough water to grow avocados?, Commission, C.A., ed. (From the Grove), pp. 22-26.

# **Budget Justification – Year 1-3**

# **Direct Costs - Personnel**

**Personnel:** Salaries and benefits are requested for a limited term employee (LTE) who will spend 50% of his/her time (1000 hrs/year) on this project. Fringe benefits are requested at 40% of his salaries. The LTE will be responsible for supervising two undergraduate students. He will also manage all experimental aspects in the lab including ion analysis, physiological measurements, and biochemical analysis. Salaries are requested for two UCR undergraduate students for 1,000 hours at \$17/hour. Undergraduate students will conduct all aspects of field-work including transplanting, watering, managing plants, sample collection and preparation for different analyses.

# **Direct Costs - Other**

**Materials and Supplies:** For each year we will need about 540 trees, with an average cost of a tree being \$10 that total cost will \$5400 per year. \$4,000 is requested each year to cover the costs of chemical reagents for ionic and mineral analyses, salinizing salts, pots, and potting soil mix. \$2,000 is requested each year for Tags, Tubes of tissue collection, Tips, glassware, plastic ware \$2000, \$500, and \$500 are requested for Y1, Y2 and Y3, respectively, to cover the cost of irrigation system setup.

#### 1. Personnel Total all Description 2026-27 2025-26 2027-28 vears 1 Lab/Field Technician - 1000 hours/yr @ \$25/hr \$ 25,000 \$ 25,000 \$ 25,000 \$ 75,000 2 Undergraduates Hourly - 500 hours/yr each @ \$ 17,000 \$ 17,000 \$ 17,000 \$ 51,000 \$17/hour Fringe Benefits for Lab/Field Technician @ 40% \$ 10,000 \$ 30,000 \$ 10,000 \$ 10,000 \$ 52,000 \$ 52,000 \$ 52,000 S **Personnel Subtotal** 156,000

# 2. Other operating Expenses

| Description  | 2025-26   | 2026-27   | 2027-28   | T  | 'otal all<br>years |
|--|-----------|-----------|-----------|----|--------------------|
| Cost of trees  | \$5,400   | \$5,400   | \$5,400   | \$ | 16,200             |
| Chemical reagents, salinizing salts, pots, potting soil mix          | \$4,000   | \$4,000   | \$4,000   | \$ | 12,000             |
| Tags, Tubes for tissue collection, Tips, glassware, plastic ware     | \$2,000   | \$2,000   | \$2,000   | \$ | 6,000              |
| Cost of irrigation system (hoses, pressure-<br>compensated drippers) | \$2,000   | \$500     | \$500     | \$ | 3,000              |
| Other Operating Expenses Subtotal                                    | \$13,400  | \$11,900  | \$11,900  | \$ | 37,200             |
| Grand Total  | \$ 65,400 | \$ 63,900 | \$ 63,900 | \$ | 193,200            |

**Matching funds:** US Salinity Laboratory will provide in-kind support in terms of personnel time and equipment maintenance support for the proposed study. In addition to the resources described in the budget section, this project will require part-time contributions of two USDA research assistants experienced in field and laboratory experimentation and analyses (Dr. Manju Pudussery, 10% time) and a soil-science technician (Layton Chhour, 10% time). The total expected value of the in-kind personnel support is ~\$20,000/year. Laboratory will also provide support by maintaining service contracts for ICP (\$27,000) and Carbon Nitrogen analyzer (\$26,000). The USSL will buy Argon gas (~\$1,000) for instrumental analysis and cover publications costs (~\$2,500).

Title: Continued Research at the San Luis Obispo Rootstock Trial Site (2025-2027)

# **Project Lead**

Lauren Garner Professor, Plant Sciences Department Cal Poly 1 Grand Avenue San Luis Obispo, CA 93407 Lgarner@calpoly.edu 805-756-2479

# **Project Cooperator**

Patty Manosalva UC Riverside pmanosal@ucr.edu

# **Executive Summary**

If approved for funding for 2025-2027, I propose to continue to maintain the orchard plot and collect and analyze the data required for the multi-site rootstock study and to build on this long-term, joint investment by continuing to keep the orchard plot well-maintained. This research plot could be utilized by other PIs as a northern site for any pest surveys and/or potential biocontrol releases that CAC may fund in other **priority topics** (e.g. 25, 28-30, and/or 39). All studies and data collection will be conducted at the rootstock trial plot at Cal Poly and will be overseen by a Master's student to be recruited for this purpose. That student will oversee undergraduate research assistants in data collection and entry and will work with me and Andrew Schaffner (Professor, Cal Poly Statistics Department) to analyze the data and to continue to prepare reports for the CAC and UCR and to co-author presentations and manuscripts for dissemination to growers and the wider scientific community. Additionally, the Master's student can work with any CAC-funded PIs to coordinate and/or conduct on-site pest surveys and/or biocontrol releases.

# Background

In 2019/2020, a collaboration began between Cal Poly, UCR, and the CAC, resulting in the establishment of a rootstock trial site on Cal Poly's campus in San Luis Obispo. This is the northern-most site in the statewide rootstock trial currently being conducted by the CAC and UCR. With financial and in-kind support from the CAC, members of the avocado industry, and Cal Poly, an avocado orchard was established at a site on campus with a documented and recent history of *Phytophthora* root rot (PRR). Trees of 'Hass' avocado grafted on 'Dusa', 'PP35, 'PP40', or 'PP45' were transplanted at the Cal Poly site on 24 June 2020 using a randomized complete block design with 10 replications of 8-10 trees per rootstock treatment in 3 blocks for a total of 384 trees, which are planted on berms at a 15' x 20' tree spacing.

In keeping with the protocols established for the statewide rootstock trial, all trees were measured and their health assessed 2 months after transplanting (August 2020) and during flushing in spring (March/April 2021-24), summer (July 2021-24), and fall (October 2021-2024), and harvest data was collected in 2023 and 2024. Our work to date has resulted in several presentations (at grower meetings and scientific conferences), contributions to all intermittent and annual reports required by me and/or Patty Manosalva to meet CAC milestones, one Master's thesis, and numerous undergraduate senior projects and class projects. Since planting, funding to support this research and maintain the orchard plot has come from ~\$85K from a grant I had from the Agricultural Research Institute (end date June 31, 2023) and from the California Avocado Commission (funding cycle November 2023 through October 2025).

# **Project Objectives**

- 1. Continue to collect and analyze tree growth, health, and yield data for the multi-site rootstock study
- 2. Continue to maintain the orchard plot to provide a well-maintained northern growing region study site for CAC-funded pest surveys and/or potential biocontrol releases

# **Project Deliverables**

### Objective 1

Reports will be submitted to the CAC. Data will be shared regularly with UCR as part of our continued participation in the multi-site rootstock study. Presentations and/or manuscripts will be prepared for dissemination to growers and the wider scientific community.

# **Objective 2**

The orchard will be maintained for continued use for the rootstock trial study, as a potential site for CAC-funded pest surveys and/or biocontrol releases and as a site for grower field days.

#### Workplan and Methods

# Objective 1

Data to track tree growth, health, and productivity will be collected during the spring (2026, 2027), summer (2026, 2027), and fall (2026, 2027) flushes, and during harvest (2026, 2027). Data collection will include tree height, trunk diameter, canopy volume, yield, and rating salinity damage, heat damage, vegetative flush and bloom. All data collection will be overseen by the Master's student to be recruited for this purpose. That person will oversee undergraduate research assistants in data collection and entry and will work with me and Andrew Schaffner (Professor, Cal Poly Statistics Department) to analyze the data and to continue to prepare reports for CAC and UCR and to co-author presentations and manuscripts for dissemination to growers and the wider scientific community.

#### Objective 2

In addition to employing students as research assistants, having student orchard assistants will allow us to dedicate weekly efforts to regular management and maintenance issues, including tasks such as pruning, weeding, walking irrigation lines, scouting, and harvesting. Additionally, Cal Poly's Plant Sciences Department has a long and successful history of collaborating with outside research entities to serve as a study site to monitor agricultural pests and for biocontrol releases. Our educational mission and fully functioning farm make us uniquely suited to such collaborations.

#### **Project Outreach**

Project results will be communicated to California avocado growers through presentations at grower meetings, on-site field days and direct interaction with industry members at meetings and visiting the campus site.

# **Budget:**

Total estimated 2-year cost (2025-2027): \$58, 065 See attached budget and budget justification.

#### **Milestones Table**

| Milestone    | Activities   | Scheduled<br>Completion            | Budget            |
|--------------|--|------------------------------------|-------------------|
| 1            | <ul> <li>Collect tree health<br/>data at Cal Poly<br/>orchard.</li> <li>Orchard<br/>maintenance</li> </ul>   | January 2026                       | \$6000            |
| 2            | <ul> <li>Collect tree health<br/>data at Cal Poly<br/>orchard.</li> <li>Orchard<br/>maintenance</li> </ul>   | April 2026                         | \$6000            |
| 3            | <ul> <li>Collect tree health<br/>and harvest data at<br/>Cal Poly orchard.</li> <li>Orchard<br/>maintenance</li> </ul>   | July 2026                          | \$9,500           |
| 4            | <ul> <li>Collect tree health<br/>data at Cal Poly<br/>orchard.</li> <li>Orchard maintenance</li> </ul>   | October<br>2026                    | \$7732            |
| Year 1 total | cost   |                                    | \$29,232          |
| Milestone    | Activities   | Scheduled<br>Completion            | Budget            |
|              |  |                                    |                   |
| 5            | <ul> <li>Collect tree health<br/>data at Cal Poly<br/>orchard.</li> <li>Orchard maintenance</li> </ul>   | January 2027                       | \$6000            |
| 5            | data at Cal Poly<br>orchard.   | January 2027<br>April 2027         | \$6000<br>\$6000  |
|              | <ul> <li>data at Cal Poly<br/>orchard.</li> <li>Orchard maintenance</li> <li>Collect tree health<br/>data at Cal Poly<br/>orchard.</li> <li>Orchard</li> </ul>   |                                    |                   |
| 6            | <ul> <li>data at Cal Poly<br/>orchard.</li> <li>Orchard maintenance</li> <li>Collect tree health<br/>data at Cal Poly<br/>orchard.</li> <li>Orchard<br/>maintenance</li> <li>Collect tree health<br/>and harvest data at<br/>Cal Poly orchard.</li> <li>Orchard</li> </ul>   | April 2027                         | \$6000            |
| 6<br>7       | <ul> <li>data at Cal Poly<br/>orchard.</li> <li>Orchard maintenance</li> <li>Collect tree health<br/>data at Cal Poly<br/>orchard.</li> <li>Orchard<br/>maintenance</li> <li>Collect tree health<br/>and harvest data at<br/>Cal Poly orchard.</li> <li>Orchard<br/>maintenance</li> <li>Collect tree health<br/>data at Cal Poly<br/>orchard.</li> <li>Orchard.</li> <li>Orchard<br/>maintenance</li> <li>Collect tree health<br/>data at Cal Poly<br/>orchard.</li> <li>Orchard maintenance</li> </ul> | April 2027<br>July 2027<br>October | \$6000<br>\$9,500 |

| Sponsor:                    |      | CA Avocado Commission        |               |          |          |          |          |
|-----------------------------|------|------------------------------|---------------|----------|----------|----------|----------|
| Title:                      |      | New and Continuing Avocad    | lo Research   | Trials   |          |          |          |
| Project Term:               |      | 11/1/25-10/31/27             |               |          |          |          |          |
| Proposal #:                 |      | 25_242                       |               |          |          |          |          |
|                             |      |                              |               |          | Quarter  | Quarter  |          |
|                             |      |                              |               |          | Year 1   | Year 2   |          |
| Personnel                   | WTUs |                              |               |          |          |          | Total    |
| Principal Investigator      | 0,0  | 0,00% Release @              | \$133,837     | /AY      | \$0      |          | \$0      |
| (no compensation)           |      | 0.00 months sum @            | \$16,730      | /MO      | \$0      |          | \$0      |
| Statistics Professor        | 0,0  | 0,00% Release @              | \$180,344     | /AY      | \$0      |          | \$0      |
|                             |      | 20.00 hours overload @       | \$86.70       | /HR      | \$1,734  |          | \$1,734  |
|                             |      | 15.00 hours overload @       | \$91,04       | /HR      |          | \$1,366  | \$1,366  |
| Undergraduate Student       |      |                              |               |          |          |          |          |
| Research Assistants         |      | 490 hours @                  | 18,50         | /HR      | \$9,065  |          | \$9,065  |
|                             |      | 490 hours @                  | 18.50         | /HR      |          | \$9,065  | \$9,065  |
| Graduate Student            |      |                              |               |          |          |          |          |
| Research Technician         |      | 129 hours @                  | 21.00         | /HR      | \$2,709  |          | \$2,709  |
|                             |      | 129 hours @                  | 21,00         | /HR      |          | \$2,709  | \$2,709  |
|                             |      |                              | Subtotal Pe   | ersonne  | \$13,508 | \$13,140 | \$26,648 |
| Fringe Benefits             |      |                              |               |          |          |          |          |
| Faculty summer & overload   |      | Statistics Professor         | 8.5%          |          | \$147    | \$116    | \$263    |
| Undergraduates              |      | Undergraduate Student Reseat | 2,5%          |          | \$227    | \$227    | \$454    |
| Graduate Students           |      | Graduate Student Research Te | 8.5%          |          | \$230    | \$230    | \$460    |
|                             |      | Sul                          | btotal Fringe | Benefits | \$604    | \$573    | \$1,177  |
|                             |      | τοτα                         | L Personnel S | Services | \$14,112 | \$13,713 | \$27,825 |
| Other                       |      |                              |               |          |          |          |          |
| Tuition                     |      |                              |               |          | \$15,120 | \$15,120 | \$30,240 |
|                             |      |                              | SUBTOTA       | L Other  | \$15,120 | \$15,120 | \$30,240 |
|                             |      | тоти                         | L Other Dire  | ct Costs | \$15,120 | \$15,120 | \$30,240 |
|                             |      |                              |               |          |          |          |          |
|                             |      | тс                           | TAL DIRECT    | COSTS    | \$29,232 | \$28,833 | \$58,065 |
| Indirect Costs              |      |                              |               |          |          |          |          |
| Cal Poly Recovered F&A Base |      |                              |               |          | \$14,112 | \$13,713 | \$27,825 |
| Cal Poly Recovered F&A      |      | 0.0% of Modified Total Di    | rect Costs    |          | \$0      | \$0      | \$0      |
|                             |      | elere el mounou rotal bi     |               |          | ψu       | 40       | ÷2       |
|                             |      | TOT                          | AL SPONSOR    | COSTS    | \$29,232 | \$28,833 | \$58,065 |
|                             |      | 1017                         | AL SPONSOR    | 00313    | \$29,23Z | ¥20,033  | \$30,005 |

#### **Budget Narrative:**

#### **PERSONNEL:**

- Lauren Garner, Cal Poly- Plant Sciences Professor; Pl overseeing project; no support requested
- Andrew Schaffner, Cal Poly- Statistics Professor; Statistical support; 20 and 15 hours per year in year 1 and 2, respectively
- Graduate Student, Cal Poly- Research technician to oversee data collection and analysis and undergraduate research assistants; 129 hours/year
- Undergraduate employees, Cal Poly- student research assistants 290 hours/year (data collection and entry) and student orchard employees 200 hours/year (assist in orchard management)

**SALARIES AND WAGES**: The salary and wage rates are based on the California Polytechnic State University (CPSU) and Cal Poly Corporation (CPC), jointly Cal Poly, established salary and wage rates paid during the 2024-2025 Fiscal year (July 1 – June 30). In general, faculty duties at CPSU consist of fifteen units in each of three Academic terms per eight-month Academic contract year, exclusive of academic breaks and summer sessions. Faculty 12-month appointments may include a combination of academic and administrative duties and encompass academic breaks and summers. Cal Poly will transition from three academic year terms to

two academic year semesters by Fall 2026, but this is not expected to affect institutional base salaries, and faculty duties will still consist of 15 units per semester term. The salary and wage rates for faculty and non-student staff generally include a projected 5% salary increase per year. The rates shown are for budgetary purposes; the rates in effect at the time the work is performed will be charged to the project.

# FRINGE BENEFITS & EMPLOYER PAYROLL TAXES:

Benefits for CPSU Faculty summer and overload work include FICA, SUI and Workers Compensation and are calculated at the proposed DHHS pooled rate of 8.5%.

CPC undergraduate student benefits include SUI and Worker's Compensation. The proposed DHHS pooled rate of 2.5% is used for budgetary purposes.

CPC graduate student fringe benefits include SUI and Worker's Compensation which would result in the proposed DHHS pooled rate of 2.5%. CPC graduate students convert to intermittent employees if the graduate student is not fully enrolled when the work is performed, resulting in the addition of FICA to fringe benefits and the current intermittent fringe benefit rate of 8.5%. Cal Poly elects to budget graduate student fringe benefits at the proposed DHHS pooled intermittent rate of 8.5%, assuming that the graduate students will not be fully enrolled. It is not feasible to assess enrollment status at the time of proposal submission.

The rates in effect at the time the work is performed will be charged to the sponsor.

**OTHER COSTS:** Tuition for a graduate student is requested at \$15,120/year.

# FACILITIES AND ADMINISTRATIVE (F&A) COSTS:

Per sponsor guidelines, "It is the policy of the California Avocado Commission to only pay direct project costs, indirect or overhead costs are not allowed."

#### Integrating Chemical and Cultural Practices for Bot Canker Control in Avocado

Project start date: 1 November 2025 Project end date: 31 October 2027 Project Leader: Fatemeh Khodadadi Position Title: Assistant Professor of Extension and Assistant Plant Pathologist Address: University of California, Riverside, 900 University Avenue, Riverside, CA 92521 Primary Telephone Contact Number: 951-827-4764 (mobile 845-901-3046) E-mail Address: fatemehk@ucr.edu

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**Executive Summary:** Avocado branch canker, a fungal disease caused by various species in Botryosphaeriaceae family, significantly threatens global avocado production. These fungal pathogens have been associated with branch canker and dieback in avocado trees worldwide, including Brazil, Chile, Greece, Italy, Mexico, New Zealand, and Spain<sup>1-6</sup>. Recent surveys indicate a dramatic increase in avocado branch canker prevalence across Southern California orchards. Botryosphaeriaceae incidence has surged in Ventura (48% to 73%), San Diego (20% to 65%), and San Luis Obispo (39% to 83.3%) counties, posing a serious threat to the avocado industry's sustainability<sup>7,8</sup>. Pre-harvest avocado branch canker is characterized by distinct cankers with necrotic bark, reddish-brown wood discoloration, and potential whitish exudate. Once established, the pathogen disrupts xylem and cambium, leading to reduced tree vigor, leaf scorch, and branch dieback. Severe infections result in yield loss and tree mortality<sup>7-10</sup>. These fungi, acting as latent endophytes or saprobes, exploit environmental stressors like drought, nutrient deficiency, or mechanical damage to become pathogenic. Wounds from pruning, mechanical damage, sunburn or insect infestations serve as entry points, facilitating spore production and spread.

Avocado branch canker management is challenging due to limited registered fungicides. While some fungicides show potential<sup>11</sup>, research is sparse compared to other crops. Current control relies on cultural practices, which are insufficient, highlighting the need for fungicide efficacy studies tailored to California's avocado industry. Water stress, both in terms of amount and timing of irrigation, is suspected to significantly influence tree susceptibility. Drought or inconsistent irrigation can weaken defenses, while over-irrigation or waterlogged conditions compromise root health, both potentially exacerbating canker development. Similarly, salinity stress weakens trees by disrupting nutrient and water uptake, creating entry points for the pathogen. Understanding the precise relationships between irrigation, salinity, and branch canker is crucial for developing effective management strategies.

Having identified and characterized the primary *Botryosphaeria* species causing branch canker in Southern California<sup>12</sup>, this project will develop and implement an IDM strategy to minimize disease impact and enhance long-term orchard health and productivity.

# **Project Objectives**

- 1. Evaluate the efficacy of various fungicides against *Botryosphaeria* species through *in vitro* **and field** trials, assessing both **curative and preventative** applications, and determine optimal application timing and frequency.
- 2. Investigate the impact of different irrigation levels on branch canker development in avocado trees, both in greenhouse and field settings.
- 3. Determine the salinity tolerance of *Botryosphaeria* species *in vitro* and to determine how salinity stress influences disease development and avocado tree health under controlled greenhouse conditions.
- 4. Integrate research findings into a practical IDM guide for avocado growers, disseminated through extension activities.

# **Project Deliverables:**

This project will deliver several key outcomes to combat Avocado Branch Canker in avocados. Firstly, a comprehensive report will detail the efficacy of various fungicides, determined through *in vitro* and field trials, including optimal application timing and frequency for both curative and preventative treatments. This report will be supported by detailed data tables and statistical analyses. Secondly, a research report will document the impact of varying irrigation regimes on canker development, presenting data on disease severity, soil moisture, and tree health, alongside corresponding analyses. Thirdly, the salinity experiment will deliver comprehensive data on Botryosphaeria spp. responses to salt stress. In vitro studies will yield EC<sub>50</sub> values for mycelial growth and spore germination across various salt concentrations, documented through tables, graphs, and microscopic assessments. Greenhouse experiments will provide detailed records of canker symptom development, disease incidence, and fungal growth in avocado trees subjected to varying salinity regimes, alongside tree health parameters and soil EC. Both phases will culminate in detailed reports with statistical analyses, elucidating the impact of salinity on fungal biology and avocado disease development. Finally, the project will culminate in the development of a practical IDM strategy, integrating fungicide and cultural practice optimizations. This strategy will be accompanied by a user-friendly guide for avocado growers, providing clear instructions, visual aids, and decision-making tools. Workshops will be conducted to disseminate the information, and both digital and physical copies of the guide will be made available to ensure effective implementation.

#### • Work Plan and Methods:

# 1. Efficacy of various fungicides against Botryosphaeria species through in vitro and field trials, assessing both curative and preventative applications, and determine optimal application timing and frequency.

*In Vitro* Screening: Isolates of the predominant *Botryosphaeria* species have been collected from symptomatic avocado trees in various California growing regions and identified using morphological and molecular methods<sup>12</sup>. To identify effective fungicides, we will conduct standard laboratory assays, including mycelial growth inhibition and spore germination inhibition, using fungicides representing diverse modes of action. Specifically, we will measure mycelial growth (colony diameter), spore germination rates, and calculate  $EC_{50}$  values for each fungicide. In vitro experiments will be conducted in our UC Riverside laboratory. Comprehensive fungicide screening will occur in Year 1.

**In Field Screening:** To assess the curative effect of fungicides on avocado branch canker in avocado pruning wounds, a field trial will be conducted using a randomized complete block design. Mature avocado trees will have three green shoots of similar thickness tip-pruned at approximately 12-15 cm from the basal ends and immediately inoculated with 20 µL of a *Botryosphaeria* isolate conidium suspension at a specific concentration. Following inoculation, each treated shoot will be covered with a transparent plastic bag for 24 hours to maintain humidity. To assess curative efficacy, designated pruning wounds will be sprayed with selected fungicides at label rates either 24 hours (day 1), 3 days, or 7 days post-pruning and inoculation, while positive and negative control wounds received no fungicide treatment. For each experiment we will use the most effective fungicides from *in vitro* tests, fungicide application combined with 1.15% NAA (Tre-Hold A-112), and NAA application alone. The trees will be maintained under standard field conditions, and lesion development will be assessed eight months post-inoculation by measuring lesion lengths and attempting fungal re-isolations from lesion margins to confirm Koch's postulates.

For preventative treatments in our avocado field trials, we will utilize the most effective fungicides identified from in vitro tests, alongside applications of 1.15% NAA (Tre-Hold A-112) alone, and a combination of fungicides with NAA. Selected branches will be pruned, and treatments will be immediately applied to the pruning wounds using a paintbrush. Subsequently, a 20 µL mixed Botryosphaeria isolate conidium suspension will be applied to each wound with a micropipette at days 1, 7, or 14 post 'pruning and treatment'. For the untreated control, branches will be treated with sterile distilled water immediately after pruning and then inoculated with the *Botryosphaeria* conidium suspension following the same procedure used for the other preventative treatments. After eight months disease incidence (number of cankers per tree), disease severity (canker size, branch dieback), and yield (fruit weight, number) data will be collected. Field trials will be conducted in cooperating commercial avocado orchards with a history of branch canker or will be done in research orchards in Pine Tree Ranch in Santa Paula (Ventura County). Field trial

preparation will commence in Year 1, along with fungicide applications, Year 2 will be dedicated to data collection, and initial data analysis and repeat field trials (contingent on Year 1 results), final data analysis, and report completion. We foresee minimal challenges for the *in vitro* fungicide assay. However, potential obstacles for the field trials include weather variability impacting results, the possible development of fungicide resistance, and difficulties in securing cooperating orchards.

# 2. Impact of different irrigation levels on Botryosphaeria canker development in avocado trees, both in greenhouse and field settings.

To comprehensively investigate the impact of water stress on branch canker development in avocado, a two-pronged approach will be employed. A controlled field experiment will begin by subjecting mature avocado trees to different irrigation regimes: optimal, water deficit, and overirrigation. Soil moisture sensors will continuously monitor water content. Trees will be inoculated with the pathogen and simultaneously treated with selected fungicides during varying irrigation regimes to assess the independent impact of water stress and the combined effect of irrigation and fungicide application. Second, a complementary pot experiment will be established, allowing for greater control over environmental variables. Young avocado trees will be grown in containers and subjected to the same irrigation treatments as the field experiment. Critically, in the pot experiment, trees will be inoculated with the dominant Botryosphaeria species. The pot experiment will also include fungicide treatment groups to isolate the effects of water stress and evaluate the combined impact of water stress and fungicide application on disease control/development. Both experiments will monitor canker lesion development, disease incidence, and tree health parameters. Statistical analysis will be used to determine the impact of irrigation treatments, and fungicide applications on disease development, providing insights into optimal management strategies. Selected branches on each tree will be inoculated with a standardized Botryosphaeria strain. Disease severity will be assessed by measuring canker lesion size and recording disease incidence at regular intervals. The greenhouse experiment will be performed during the first year of the project at the UCR campus greenhouse. The field experiment will be executed in the second year, utilizing the same orchard as the fungicide assay. This objective faces potential challenges, notably unpredictable rainfall that can disrupt irrigation regimes and extreme temperatures that may adversely impact tree health and pathogen development.

# 3. Determine the salinity tolerance of Botryosphaeria species in vitro and to determine how salinity stress influences disease development under controlled greenhouse conditions.

To investigate the effects of salinity on *Botryosphaeria* spp. *in vitro*, we will evaluate the impact of various salt concentrations on colony growth and spore germination of ten isolates from each identified *Botryosphaeria* species. Spore suspensions and mycelial plugs will be obtained from 7-day-old colonies grown on Potato Dextrose Agar (PDA) at 25°C. Mycelial plugs and standardized spore suspensions (quantified using a hemocytometer) will be inoculated into PDA and Potato Dextrose Broth (PDB) media amended with varying concentrations of NaCl, KCl, MgSO4, MgCl2, or CaCl2. Cultures will be incubated at 25°C in the dark, with liquid cultures agitated in a

shaker incubator. Colony growth (measured as colony diameter) and spore germination rates will be assessed microscopically at multiple time points (e.g., 24, 48, 72, 96 hours). Liquid cultures will be assessed for visible growth (mycelial development or turbidity) after 4 weeks. Sterile, salt-free media will serve as negative controls. All treatments will be performed in triplicate.

To examine the effects of salinity on *Botryosphaeria* species in a controlled environment, we will conduct greenhouse experiments using potted Hass avocado trees grafted onto Duke 7 or Toro Canyon rootstock. Prior to initiating salt treatments, trees will be acclimated to greenhouse conditions. Plants will be randomly assigned to one of three treatment groups: a non-saline control (NS) receiving irrigation at optimal electrical conductivity (EC) for avocado growth, a leached salt treatment (LS), and a continuous salt treatment (CS). For the LS and CS groups, irrigation solutions will be amended with a 1:1 equivalent ratio of NaCl and CaCl2. The EC of these solutions will be incrementally increased over eight days in four equal steps, reaching a maximum of 7 dS·m-1. On day nine, the LS group will undergo leaching with non-saline irrigation solution, while the CS group will continue to receive the 7 dS·m-1 solution. One week after the maximum salt levels are reached (day 15), select branches on each tree will be inoculated with a standardized Botryosphaeria conidial suspension. Throughout the experiment, we will monitor symptom development, disease incidence, and fungal growth in tree tissues across all three treatment groups. In vitro experiments will be conducted at the UC Riverside laboratory, while greenhouse experiments will be performed at the UCR campus greenhouse facility. The *in vitro* salinity data will be collected in year one, while the greenhouse experiment will be conducted in year one and two of the project. Challenges include maintaining precise salinity levels in irrigation, ensuring uniform salt distribution in potting media, and effectively leaching salts from the LS treatment, requiring determination of optimal leaching time and volume.

# 4. Integrate research findings into a practical IDM guide for avocado growers, disseminated through extension activities.

We will create a comprehensive IDM strategy for branch canker by analyzing fungicide and cultural practice data, including pot studies, using statistical methods. A risk assessment framework will guide the development of integrated protocols, combining optimized irrigation, salinity management, and fungicides. On-farm trials will validate the strategy, which will be translated into a user-friendly grower guide with practical tools and disseminated through workshops and ongoing support. To effectively reach California avocado growers, we will use a multi-pronged approach: creating accessible extension publications, conducting in-person and virtual grower meetings, and engaging industry partners like PCAs and Farm Advisors. We will develop clear, visual-based publications available in print and digital formats, hold interactive meetings with Q&A sessions, and provide training workshops and resource development.

# Milestone

The following Milestone Table outlines the activities associated with the project and scheduled completion dates.

| Year 1    | 11/1/2025-10/31/2026  |   |           |
|-----------|---|---|-----------|
| Milestone | Activities  | Scheduled Completion                        | Budget    |
| 1         | PhD student Salary (Valentina Valencia<br>Bernal)                                     | October, 2026                               | \$61,149  |
| 2         | In vitro fungicide sensitivity testing  | February, 2026                              | \$2,000   |
| 3         | In vitro salt sensitivity testing   | March, 2026                                 | \$2,000   |
| 4         | Irrigation impact greenhouse trial  | October, 2026                               | \$4,000   |
| 5         | Greenhouse salinity effects experiment  | October, 2026                               | \$4,000   |
|           |   | Year 1 Total                                | \$73,149  |
| Year 2    | 11/1/2026-10/31/2027  |   |           |
| 1         | PhD student Salary (Valentina Valencia<br>Bernal)                                     | October, 2027                               | \$63,970  |
| 2         | Continuation of greenhouse assay  | March, 2027                                 | \$2000    |
| 3         | Setting up the trials for the efficacy of fungicides in the field and collecting data | October 2027                                | \$4,500   |
| 4         | Field trial for irrigation impact test  | October, 2027                               | \$4,500   |
|           |   | Year 2 Total                                | \$74,970  |
|           |   | Total Project<br>Budget excluding<br>travel | \$148,119 |

#### References

- Arjona-Girona, I., Ruano-Rosa, D., and López-Herrera, C. J. 2019. Identification, pathogenicity and distribution of the causal agents of dieback in avocado orchards in Spain. Span. J. Agric. Res.17: e1003. https://doi.org/10.5424/sjar/2019171-13561
- Auger, J., Palma, F., Pérez, I., and Esterio, M. 2013. First report of Neofusicoccum australe (Botryosphaeria australis), as a branch dieback pathogen of avocado trees in Chile. Plant Dis. 97:842.
- Hernández, D., García-Pérez, O., Perera, S., González-Carracedo, M. A., RodríguezPérez, A., and Siverio, F. 2023. Fungal pathogens associated with aerial symptoms of avocado (Persea americana Mill.) in Tenerife (Canary Islands, Spain) focused on species of the family Botryosphaeriaceae. Microorganisms. 11:585
- Guarnaccia, V., Polizzi, G., Papadantonakis, N., and Gullino, M. L. 2020. Neofusicoccum species causing branch cankers on avocado in Crete (Greece). J. Plant Pathol. 102:1251–1255. https://doi.org/10.1007/s42161-020-00618-y

- Guarnaccia, V., Vitale, A., Cirvilleri, G., Aiello, D., Susca, A., Epifani, F., Perrone, G., and Polizzi, G. 2016. Characterisation and pathogenicity of fungal species associated with branch cankers and stem-end rot of avocado in Italy. Eur. J. Plant Pathol. 146: 963–976.
- Valencia, A. L., Gil, P. M., Latorre, B. A., and Rosales, I. M. 2019. Characterization and pathogenicity of Botryosphaeriaceae species obtained from avocado trees with branch canker and dieback and from avocado fruit with stem end rot in Chile. Plant Dis. 103:996–1005.
- Avenot, H. F., Vega, D., Arpaia, M. L., and Michailides, T. J. 2023. Prevalence, identity, pathogenicity, and infection dynamics of Botryosphaeriaceae causing avocado branch canker in California. Phytopathology 113:1034–1047.
- 8. McDonald, V., and Eskalen, A. 2011. Botryosphaeriaceae species associated with avocado branch cankers in California. Plant Dis. 95:1465–1473.
- Dann, E. K., Ploetz, R. C., Coates, L. M., and Pegg, K. G. 2013. Foliar, fruit, and soilborne diseases. Pages 380-422 in: The avocado: botany, production and uses. B. Schaffer, B. N. Wolstenholme, A. W. Whiley, eds. CABI, Wallingford, U. K
- 10. Mehl, J. W., Slippers, B., Roux, J., and Wingfield, M. J. 2013. Cankers and other diseases caused by the Botryosphaeriaceae. Infect. For. Dis. 298-317.
- Twizeyimana, M., McDonald, V., Mayorquin, J. S., Wang, D. H., Na, F., Akgül, D. S., and Eskalen, A. 2013b. Effect of fungicide application on the management of avocado branch canker (formerly Dothiorella canker) in California. Plant Dis. 97:897–902.
- Valencia Bernal, V., Pegahrad, Z., Kamali Dashtarzhaneh, M., Khodadadi, F. 2025. Identification, characterization, and fungicide sensitivity of Botryosphaeriaceae fungi associated with avocado branch canker disease in Southern California. Plant Dis. doi: 10.1094/PDIS-12-24-2674-RE. Epub ahead of print. PMID: 39946273.

#### Budget

| Year 1   | Budget    |
|--|-----------|
| Personnel (includes salary, benefits, fees etc.) (Salary: \$40,130+<br>Benefits: \$843 + Tuition and Fees: \$20,176) | \$61,149  |
| Supplies   | \$12,000  |
| Travel   | \$4,000   |
| Year 1 Total   | \$77,149  |
| Year 2   |           |
| Personnel (includes salary, benefits, fees etc.) (Salary: \$42,574+<br>Benefits: \$894 + Tuition and Fees: \$20,502) | \$63,970  |
| Supplies   | \$11,000  |
| Travel (weekly trips to field sites [car rental, gas], meetings etc.)  | \$5,000   |
| Year 2 Total   | \$79,970  |
| Total Budget   | \$157,119 |

#### **Budget Justification:**

**A. Senior Personnel – \$0 Fatemeh Khodadadi**, Lead Principal Investigator (\$0) Dr. Khodadadi will be overseeing the project.

#### B. Other Personnel - \$82,704

Graduate Student Researcher, Valentina Valencia Bernal/Dr. Khodadadi Lab (\$82,704): Dr. Khodadadi will supervise one graduate student researcher at 50% FTE for 12 months during years 1 and 2 of the project. Costs for wages in Year 1 are \$40,130 and \$42,574 in Year 2.

UC Riverside defines a year as the Fiscal Year from July 1<sup>st</sup> through June 30<sup>th</sup>. All salaries and wages are estimated using UC Riverside's academic and staff salary scales. Anticipated cost of living increases of 3% per year are included for the PI and Graduate Student Researcher . Where appropriate, merit increases are included in the calculations. Merit increases for academic personnel are estimated at 5%.

#### Fringe Benefits - \$1,737

Employee benefits are estimates, using the composite rates agreed upon by the University of California. Graduate Student Researcher fringe benefit rates are estimated at 2.1%.

#### C. Travel - \$9,000

Dr. Khodadadi's lab - \$9,000. PI Dr. Khodadadi requests a travel budget to cover travel expenses for grower meetings, workshops, and field trials in Ventura. This will include car rental from Enterprise at \$40 per day plus fuel, and overnight lodging and meals at per diem rates or actual expenses for survey location trips. Year 1: \$4000; Year 2: \$5000.

The travel destinations are tentative and are subject to change. Costs are based upon historical usage and include coach airfare on domestic U.S. flag carriers, ground transportation, lodging, registration fees, meals, and incidental expenses.

#### D. Other Direct Costs - \$63,678

#### 1. Materials & Supplies - \$23,000

Dr. Khodadadi Lab - \$23,000. Dr. Khodadadi is requesting \$23,000 to support the following project needs: rental of greenhouse space in Riverside, purchase of necessary chemicals and slats, acquisition of supplies for in vitro fungicide and salt assays, and the purchase of avocado trees for use in greenhouse experiments. Year 1: \$12,000; Year 2: \$11,000

#### 2. Tuition & Fees - \$40,678

University policy requires inclusion of partial fees and tuition remission and Graduate Student Health Insurance (GSHIP) for GSRs employed during each academic year with an appointment of 25% effort or more. GSR Valentina Bernal will be employed at 50% FTE which will result in tuition and fees costs of \$20,176 in Year 1 and \$20,502 in Year 2 for a total of \$40,678.

#### E. Total Direct Costs - \$157,119

#### F. Indirect Costs - \$0

No Indirect Costs are requested.

#### K. Total Cost - \$157,119

# Addressing the relationship between soil characteristics and soil salinity in California avocado orchards

Project lead: Jesse Landesman, UC Santa Barbara, (626) 240-9169 Project Cooperators:

- Jennifer King, UC Santa Barbara
- Maureen Cottingham, CamLam Farms
- Iris Holzer, UC Santa Barbara
- Anna Trugman, UC Santa Barbara
- Rick Shade, Shade Management

Executive Summary:

With increasing climate variability, soil salinization has significantly contributed to land degradation over the last century<sup>1</sup>. By 2050, it is predicted that 50% of arable land will be salinized because of decreasing precipitation, increasing surface evaporation, increasing weathering, and irrigation with poor quality water<sup>2</sup>. Since irrigation plays an important role in the salinization of soils, soil salinity is an especially pertinent problem in agriculture. Salinity is a major issue specifically for the avocado industry, a crop highly sensitive to increases in salinity. California produces 95% of domestically grown avocados, and salinity is an increasing issue due to the geographic distribution of avocado orchards along the California coast as well as increased pressure for farmers to irrigate with reclaimed water. Specifically, chloride ions cause the greatest harm in avocado trees<sup>3</sup>. While past research has documented the effects of salinity on avocado productivity, little is known about the mechanisms by which variation in soil properties affects accumulation of soil salinity and accompanying changes in soil health. Therefore investigating the effects of soil physical and chemical properties on soil salinity in avocado orchards is critical to the future of avocados. This research project is separated into three separate components. The first component is to highlight and understand the scope of the problem of soil salinization in California avocado orchards. The second component is to identify what soil physical and chemical properties are most correlated with soil salinity across hillslopes in contrasting parent materials. The third component is to understand how differing irrigation water chemistries interact with different soil types to retain and accumulate salts. Research question 1: What California avocado-producing areas are most at risk of soil salinization?

**Hypothesis:** Areas with poorer quality irrigation water and more severe drought and unpredictable rainfall events will experience the greatest risk of soil salinization.

# **Objectives:**

- Modify and run the HYDRUS (2D/3D) model to incorporate all necessary forcings for soil salinization risk mapping, including historical water quality data and climate change predictions
- Incorporate grower input in the form of a survey sent out via the GreenSheet to collect current and historical irrigation water data and assess growers' understanding of the chemistry and quality of their irrigation water

 <sup>&</sup>lt;sup>1</sup> Shrivastava, Pooja, and Rajesh Kumar. "Soil Salinity: A Serious Environmental Issue and Plant Growth Promoting Bacteria as One of the Tools for Its Alleviation." Saudi J Biol Sci., vol. 22, no. 2, Mar. 2015, pp. 123–31, https://doi.org/10.1016/j.sjbs.2014.12.001.
 <sup>2</sup> Jamil, A., Riaz, S., Ashraf, M., & Foolad, M. R. (2011). Gene Expression Profiling of Plants under Salt Stress. *Critical Reviews in Plant Sciences*, 30(5), 435–458. https://doi.org/10.1080/07352689.2011.605739.

<sup>&</sup>lt;sup>3</sup> Acosta-Rangel, A. M., Li, R., Celis, N., Suarez, D. L., Santiago, L. S., Arpaia, M. L., & Mauk, P. A. (2019). The physiological response of 'Hass' avocado to salinity as influenced by rootstock. *Scientia Horticulturae*, *256*, 108629. https://doi.org/10.1016/j.scienta.2019.108629

# Deliverables:

- A map of three categories of soil salinization risks (Table 1) across the areas where avocados are grown in the state of California
- A presentation at a CAC board meeting on secondary soil salinization risks across California avocados

#### Methods:

- 1. Create a biogeochemical model of secondary soil salinization to understand the most important forcings of salt accumulation in avocado soils.
- 2. Compile necessary data for model, including location of California avocado orchards, temperature and precipitation data, soil characteristics, irrigation water quality, and historical water quality data.
- 3. Send out a survey in CAC's Greensheet. Here are example survey questions:
  - What form of irrigation do you use (i.e. drip or microsprinkler)?
  - What are the sources of water to your orchard?
  - Do you know the chemistry/quality of your irrigation water? If not, would it be useful to have your irrigation water analyzed?
  - Does your irrigation water quality change seasonally?
  - Would you be willing to share your irrigation water quality data and/or any historical data you have on irrigation water quality?
- 4. Run the HYDRUS (2D/3D) model with collected survey data and compiled climatic and soil data to create soil salinization risk assessment map.
- 5. Randomly select 8 sites on the map to visit and collect ground-truth data to test the accuracy of the model in its current form, without the climate change projections.
- 6. Present results of the model to the CAC board and to the greater scientific community.

# Work plan timeline:

- May 2025: read literature on modeling soil salinization and compile data (steps 1 and 2)
- July 2025: send out grower survey in CAC GreenSheet (step 3)
- August 2025: run model and create risk assessment map (step 4)
- October 2025: visit field sites to collect samples to validate the model (step 5)
- November 2025: present model results at Soil Science Society of America meeting and to CAC board of directors (step 6)

Table 1: Three levels of soil salinization risk that would be used in the risk assessment map

| Color | Soil electrical conductivity | Avocado yield reduction |
|-------|------------------------------|-------------------------|
|       | 0 - 0.8 dS/m                 | None                    |
|       | 0.9-1.19 dS/m                | 10%                     |
|       | 1.2 and higher dS/m          | 25%                     |

(ANR Publication 8562, 2016)

**Research question 2:** What soil physical or chemical properties are most correlated with soil salinity across hillslopes in contrasting parent materials? How do differences in salinity affect tree health and soil health, measured by tree thermal stress and soil microbial respiration?

**Hypothesis:** Soil salinity will be higher at the bottom of the slope (toeslope) than in the backslope and summit. Soil salinity will be higher in soils with marine sedimentary alluvium parent material. Trees at the top of a slope will be more water stressed, with a higher tree water deficit (TWD) and microbial activity will be limited.

# **Objectives:**

- Identify how soil physical and chemical properties and soil salinity changes along a hillslope gradient across different parent materials in Hass avocados on Toro Canyon rootstock
- Identify how avocado tree thermal stress changes along a hillslope gradient and across two different soil parent materials, using thermal infrared (TIR) imaging from drone flights
- Quantify how soil microbial respiration and community composition changes across hillslopes in contrasting parent materials

# **Deliverables:**

- A map of tree water deficit for CamLam farms using drone imagery
- Principal component analysis (PCA) figures of various physical and chemical soil properties colored by hillslope location and soil parent material
- Figures showing continuously collected data over a two year time period of soil moisture and soil electrical conductivity (EC) across hillslopes in contrasting parent materials

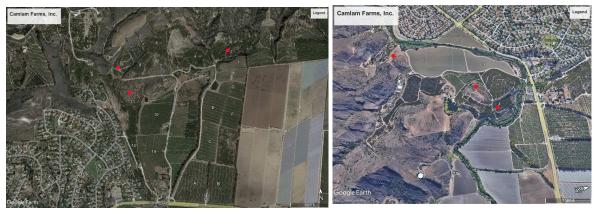


Figure 1: Red dots are site locations. At each site, there will be sensors and sampling occurring at 3 hillslope locations; the summit, the backslope, and the toeslope. The North side is dominated by marine and non-marine sedimentary materials (left panel) and the South side is dominated by volcanic materials (right panel)

# Methods:

- 1. Collect soil samples from three hillslopes in the north section of CamLam Farm and three hillslopes in the south section. The hillslope locations are identified as locations where Hass avocados are grown on Toro Canyon rootstock, to try and control for differences in salinity stress that may occur based on having differing amounts of tolerance to salinity in the rootstocks. At each hillslope location, there will be samples collected at the summit, backslope, and toeslope. Five depths will be sampled at each of these locations, resulting in a total of 90 soil samples. Soil physical properties will also be measured in the field, including:
  - Infiltration
  - Aggregate stability

- Depth to bedrock
- Penetration resistance

- Equivalent soil mass
- 2. A hillslope in the north side and south side will be selected that are the most similar to each other (i.e. same aspect, etc). Soil matric potential moisture sensors, soil EC sensors, and soil respiration flux bots will be placed at three different depths at three different locations at the two hillslopes, to collect continuous soil moisture, soil EC, and soil microbial respiration data.
- 3. Using a drone with a TIR imaging camera, we will conduct a flyover of the orchard to collect tree thermal stress data.
- 4. Soil samples collected in the field will be analyzed in the lab for specific various physical and chemical properties. Here are the measurements we are interested in:
  - Cation exchange capacity (CEC) and base saturation
  - Soil texture analysis
  - Specific concentrations of ions: Na+, Cl-, Ca2+, Mg2+
  - Phospholipid-derived fatty acids (PLFAs)
  - Soil mineralogy by XRD
  - Total organic carbon (TOC), Total inorganic carbon (TIC)
  - ∘ pH
- 5. In-field soil sensors will continue collecting data for two years, but researchers will return to the field to do seasonal drone flyovers for tree thermal stress data and collect season measurements of the following same soil chemical properties:
  - EC and specific concentration of Na, Cl, Ca, Mg
  - ∘ pH
  - PLFAs
- 6. Data will be analyzed to determine the relationship between soil physical and chemical properties and soil salinity, as well as soil salinity and tree thermal stress and soil microbial respiration and community composition.

#### Work plan timeline:

- July 2025: Begin field campaign and soil sampling and place in-field soil sensors (steps 1 and 2)
- August 2025: First drone flyover to assess tree thermal stress (step 3)
- September 2025: Begin laboratory soil analyses (step 4)
- February 2026: Return to the field for drone flyover and soil sampling (step 5)
- July 2026: Return to the field for drone flyover and soil sampling (step 5)
- February 2027: Return to the field for drone flyover and soil sampling (step 5)

**Research question 3:** How do different soils react differently to different irrigation water chemistries, specifically in regards to chloride ion retention? How does the addition of biochar alter the retention of chloride?

**Hypothesis:** Soils with higher clay content and more soil organic matter (SOM) will retain more chloride ions because clay accumulates water and SOM binds chloride. Soils amended with biochar will retain more chloride and less will be present in the leachate.

# **Objectives:**

- Identify a quantitative relationship between soil properties and chloride retention
- Identify a quantitative relationship between biochar applied and chloride retention

### **Deliverables:**

- Reports for farmers on what their soil characteristics mean for irrigation practices
- Quantitative information on the viability of biochar as a possible soil salinity solution
- Information packet on how soil characteristics interact with irrigation water chemistry and how growers should incorporate this into their irrigation management and leaching of salts
- Presentation at a CAC field day

#### Methods:

- Using the soils with the most distinctive differences in salinity that we observed from R2 at CamLam Farms, collect 40 cm PVC pipes of soil from six different locations, with replicates of four soil samples per location, leading to a total of 24 PVC pipes of soil. Collect additional soil samples to analyze for:
  - Soil texture
  - Soil mineralogy
  - TOC and TIC

- Aggregate stability
- Equivalent soil mass
- Penetration resistance

- Infiltration
- 2. Bring soil samples back to the lab and analyze soil for these characteristics. Amend one soil column per site with biochar. Apply high salinity water to a regular soil column and a biochar column, low salinity water to a soil column, and distilled water for a control column.
- 3. Collect leachate below the soil column and analyze the leachate chemistry. Continue applying respective irrigation water and collecting leachate for next 90 days.
- 4. After 90 days, analyze the chemical properties of the soil in the columns again to see how they have changed with their respective irrigation water treatment.
- 5. Analyze the data on leachate chemistry of the different irrigation water qualities and compile an information packet on how irrigation water interacts with different soils.
- 6. Present this information at a CAC field day.

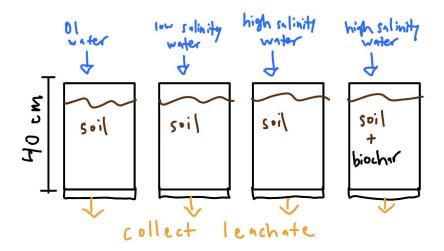


Figure 2: Soil column laboratory experiment set up with soils collected from six different sites **Work plan timeline:** 

• June 2026: Collect soil samples (step 1)

- July 2026: Analyze soil samples and begin irrigation water addition (step 2)
- September 2026: Finish analyzing collected leachate chemistry and analyze soil chemical properties (steps 3 and 4)
- October through November 2026: analyze data and present results at a CAC grower field day (steps 5 and 6)

Project outreach: Throughout the project, I will continually ask for grower feedback and input from CAC's Production Research Committee. I will utilize the GreenSheet to disperse information on grower surveys that will be used to assess the geography of soil and irrigation water chemistry. Once I have results, I will participate in on-site field days and grower meetings to communicate and create more opportunities for application of the findings.

#### Milestone table

| Task accomplished   | Research<br>objective | Date     | Estimated cost |
|---|-----------------------|----------|----------------|
| First draft of soil salinization risk assessment map  | R1                    | 09/01/25 | \$2,702        |
| Visit avocado field sites to collect water and soil samples to validate model output                                  | R1                    | 10/01/25 | \$300          |
| Present model results at SSSA in Salt Lake City   | R1                    | 11/10/25 | \$755          |
| Attend a California avocado grower meeting to present results and risks across the state                              | R1                    | 01/26    | \$0            |
| Deploy soil sensors in field site   | R2                    | 07/15/25 | \$11,868       |
| Collect first round of soil samples at field site and complete first drone flyover                                    | R2                    | 8/01/25  | \$4,460        |
| Analyze first round of soil samples   | R2                    | 10/01/25 | \$28,260       |
| Compile data into meaningful figures and publish the findings   | R2                    | 09/01/27 | \$0            |
| Collect soil samples for soil columns   | R3                    | 07/01/26 | \$3,960        |
| Run and collect data for soil column experiment   | R3                    | 10/01/26 | \$792          |
| Present data on relationship between soil type, irrigation water, and biochar at a CAC grower field day               | R3                    | 11/01/26 | \$0            |
| Compile information packet for farmers on soil characteristics and irrigation and advocate for on-farm biochar trials | R3                    | 06/01/27 | \$0            |
|   |                       | TOTAL    | \$53,097       |

| Project title<br>Time period                        | Addressing the relationship between soil characteristics and soil salinity in California avocado orchards 07/01/2025-10/31/2027 |               |               |             |             |
|---|---|---------------|---------------|-------------|-------------|
|   | Description   | Research obje | ctiv Quantity | Year 1 cost | Year 2 cost |
| <b>Salaries</b><br>Undergraduate research assistant | work 3 months/year, 10 hours/week, at \$16.50/hr  | R2 and R3     |               | 2 \$3,960   | \$3,960     |
| <b>Benefits</b><br>Undergraduate research assistant | benefits at 1.5%  | R2 and R3     |               | 2 \$59.40   | \$59.40     |
| Travel  |   |               |               |             |             |
| Travel to SSSA Conference 2025                      | plane ticket to SLC   | R1            |               | \$425       |             |
| SSSA Conference attendance                          | SSSA conference registration fee  | R1            |               | \$355       |             |
| Travel to field site                                | Vehicle miles traveled, \$0.70/mile   | R1, R2, R3    | 600 miles     | \$420       |             |
| Supplies  |   |               |               |             |             |
| Soil electrical conductivity sensor                 | METER TEROS 12 5 meter, \$258 each  | R2            |               | 18 \$4,644  |             |
| Soil moisture matric potential sensor               | METER TEROS 21 5 meter, \$274/each  | R2            |               | 18 \$4,932  |             |
| Soil respiration fluxbot                            | Do it yourself, at \$362/each   | R2            |               | 6 \$2,172   |             |
| iPad  | for in-field data collection  | R2            |               | 1 \$349     |             |
| Supplies for soil columns                           | PVC pipe, stands, miscallaneous laboratory supplies   | R3            |               | 18 \$300    |             |
| Contracted services                                 |   |               |               |             |             |
| soil salinization model software                    | HYDRUS (2D/3D)  | R1            |               | \$2,702     |             |
| Soil texture analysis                               | Laser diffraction particle size analysis, \$109/sample  | R2            |               | 90 \$9,810  |             |
| Soil microbial community composition                | PLFAs, at \$90/sample   | R2            |               | 90 \$8,100  |             |
| Soil mineralogy analysis                            | by X-Ray diffraction, \$25/sample   | R2            |               | 90 \$2,250  |             |
| Cation exchange capactiy                            | \$60/sample   | R2            |               | 90 \$5,400  |             |
| Exchangeable ions                                   | \$30/sample   | R2            |               | 90 \$2,700  |             |
| Drone consultant                                    | Collaboration with drone pilot with TIR imaging capabilities  | R2            |               | \$500       |             |
|   |   |               | TOTAL         | \$53,097.80 |             |

#### Salaries and benefits, \$8,038.80

The salaries and benefits section adds up to \$4,019.40 per year, over the span of two years, totaling \$8,038.80. This category will go directly toward paying undergraduate research assistants to help collect soil samples and process and analyze laboratory samples. Since the cost of living in Santa Barbara is quite high, being able to pay undergraduate research assistants is important to ensure their commitment and ability to do their best work. Since this is a large-scale soil sampling campaign, it is necessary to have multiple people involved to get the work done in a reasonable amount of time. It will also provide undergraduate students with important and unique opportunities to get involved in agricultural research and laboratory measurements.

# Travel, \$1,200

The money allocated in the travel category will mainly go towards the attendance of an annual conference. The specific conference, the tri-societies meeting, convenes the Agronomy Society of America, the Crop Science Society of America, and the Soil Science Society of America. This will be an important opportunity to present the work and gain exposure for the issue of soil salinization in California avocado orchards. The rest of the funds in the travel category will go towards vehicle miles traveled reimbursement for travel to the field site and to other orchards to collect samples to validate the model in R1.

#### Supplies, \$12,397

The supplies category consists mainly of the in-field sensors that will be deployed at the field site. These sensors are important because they will allow us to collect continuous data without having to disrupt day to day operations at the working farm. Since soil EC and soil water are quite closely coupled, it is important to have both of these sensors at different depths and locations. The soil respiration sensors will allow us to measure an important variable of soil biological health that is often linked with soil health. Having an iPad to collect data in the field is also important specifically for the collection of soil physical properties like penetration resistance and infiltration, since those measurements will be taken in the field. Lastly, it is important to have the supplies to create the soil columns in the laboratory so that we can carry out R3.

#### Contracted services, \$31,462

The contracted services category is the largest section of our budget. It consists of the purchase of the soil salinization model, HYDRUS (2D/3D) in order to model soil salinity risk throughout California avocado orchards. It is possible that we will be able to get this software at a discounted rate with collaborations with UC Riverside. The bulk of the contracted services come from sending soil samples for various laboratory analyses to measure various chemical and biological soil properties. These may also come at a cheaper rate as I continue to establish collaborations with UC Riverside and some soil processing capabilities that they have at their campuses. The final contracted service is to compensate my colleagues from the GROVE lab for their time and use of their drone to obtain thermal infrared imaging of the field site.

**1. Project Title:** Development and Demonstration of a Cost-effective Electrodialysis Reversal (EDR) Process for Chloride Removal from Avocado Irrigation Water

**2. Project Lead**: Haizhou Liu, PhD, PE; Department of Chemical and Environmental Engineering, 900 University Ave, University of California, Riverside, CA 92521. Email: haizhou@engr.ucr.edu; Phone: 951-827-2076. (UCR contracting point of contact: Victoria Sissac, Principal Contract and Grant Officer, Email: victoria.sissac@ucr.edu; T: 951-827-3377)

**3. Project Cooperator**: Lindsey Pedroncelli, PhD; Interim Director, UC Agricultural South Coast Research and Extension Center, Irvine, CA, Email: lrpedroncelli@ucanr.edu

# 4. Executive Summary:

This project aims to address the priority topic to pursue promising desalination technologies to help mitigate chloride in groves. Elevated chloride in irrigation water is one of the greatest threats to avocado productivity for many growers in California. The development of efficient, costeffective on-site desalination technologies to selectively remove chloride from the irrigation water at Californian avocado groves will significantly increase the yield of avocado trees, provide reliably high-quality irrigation water, and consequently increase the profits and competitiveness of Californian avocado groves. Based on a previously funded phase-one feasibility study to develop chloride mitigation technologies from irrigation water at Californian avocado groves, the project team at UC Riverside has identified electrodialysis reversal (EDR) as the most promising chloride removal technology uniquely fitted for avocado groves on-farm applications. This selection is based on a comprehensive selection criteria including chloride removal efficiency, economics and operational easiness. EDR process is estimated to incur the lowest total cost among all candidate technologies (60-80% lower cost than membrane-based and ion exchange technologies), and saves more than 70% cost than directly purchasing treated water from municipal water districts. To further pursue this promising technology platform, this phase-two project aims to develop and optimized a prototype EDR apparatus to removal chloride from California grove irrigation water, and demonstrate and validate the pilot-scale EDR treatment process to produce fresh irrigation water via chloride removal from irrigation water onsite at a California grove.

# 5. List of specific project objectives

This 3-year project has the following three main objectives:

- 1. Develop a prototype EDR apparatus and conduct chloride removal studies at lab scale using salinity-elevated irrigation water collected from an avocado grove. Optimize the EDR process by evaluating different options including ion selective membranes, applied voltage and water recovery to maximize chloride removal selectivity, minimize emerging consumption and capital/operational cost.
- 2. Demonstrate a pilot-scale electrodialysis reversal (EDR) operation on site on a California avocado grove to remove chloride, produce low-salinity irrigation water. to generate accurate data on chloride removal efficiency, water production rate, energy consumption rate and capital/maintenance cost.
- 3. Quantify the operational and capital cost of the pilot-scale demonstration and estimate the total cost for future full-scale operation in comparison to other chloride removal technologies.

# 6. List of specific project deliverables

| Performance Objectives   | Data Requirements  | Deliverables  |
|--|--|---|
| Construct a prototype EDR system at laboratory scale   | Design schematics, images,<br>and videos of the prototype.   | Fully functional prototype<br>EDR system with<br>interchangeable membrane<br>and electrode configurations.  |
| Test chloride removal efficiency using<br>different ion-selective membranes and<br>electrode materials with salinity-<br>elevated irrigation water from an<br>avocado grove. | Analyze chloride<br>concentration before and<br>after treatment for each<br>prototype configuration.   | Achieve chloride<br>concentration reduction to <<br><b>100 mg/L</b> .   |
| Evaluate energy consumption and<br>operational cost for each prototype<br>configuration.   | Conduct cost analysis based<br>on each prototype<br>configuration.   | <ul> <li>a. Determine cost per gallon<br/>to reduce chloride to &lt; 100<br/>mg/L.</li> <li>b. Select optimal prototype<br/>configuration for on-site<br/>demonstration.</li> </ul> |
| Assess chloride removal efficiency<br>through an on-site demonstration at a<br>California avocado grove.   | Conduct chloride<br>concentration analysis<br>before and after on-site<br>treatment.   | Reduce chloride concentration<br>to < <b>100 mg/L</b> for real<br>irrigation water.   |
| Evaluate energy footprint and cost for<br>both pilot-scale and full-scale<br>operations.   | <ul> <li>a. Analyze operating and<br/>maintenance (O&amp;M) costs.</li> <li>b. Assess capital costs based<br/>on pilot-scale EDR<br/>demonstration.</li> </ul> | Determine <b>cost per gallon</b> to<br>reduce chloride to < 100<br>mg/L.  |
| Operational consistency  | Maintain complete<br>recordkeeping of system<br>uptime.  | Achieve <b>80% uptime</b> during planned operations.  |
| System robustness and ease of maintenance  | Document system<br>operations and<br>troubleshooting procedures.   | Ensure the treatment process<br>is easy to implement and<br>maintain.   |

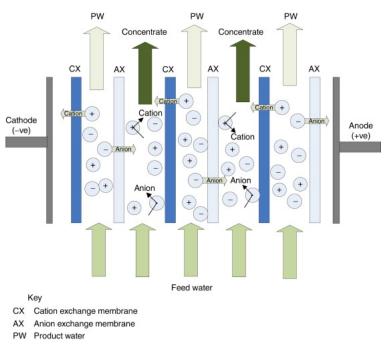
The project has the following performance objectives and deliverables:

# 7. Technology Description

Electrodialysis (ED) and electrodialysis reversal (EDR) are advanced desalination technologies that use an electric field and ion-selective membranes to remove chloride and other charged ions from water. In ED/EDR, chloride ions (anions) migrate toward the anode, while sodium ions (cations) move toward the cathode. These ions are blocked by alternating anion- and cation-selective membranes, resulting in two separate streams: purified water with reduced ion concentrations and a concentrated brine waste stream (**Figure 1**). However, a major drawback of ED is the buildup of charged particles on the membrane surface, which reduces efficiency over time.

EDR improves upon traditional ED by periodically reversing the electrical polarity, which helps prevent membrane fouling and ensures more consistent performance. This self-cleaning feature makes EDR particularly well-suited for agricultural irrigation, especially for water with low-to-

moderate total dissolved solids (TDS). EDR offers several key advantages: 1. Selective Chloride Removal – EDR removes over 95% chloride while of preserving beneficial minerals such as sulfate and other divalent ions, which are essential for crop health. 2. Higher Water Recovery - EDR achieves a significantly higher water recovery rate (90-95%) compared to reverse osmosis (RO) and nanofiltration (NF), which typically discard a larger portion of water as brine waste. Additionally, EDR requires minimal pretreatment and does not need anti-scalants, unlike RO/NF. 3. Reduced Brine Waste - EDR generates much less brine, only 5-10% of the feedwater volume, making it more environmentally friendly and cost-effective for disposal.



**Figure 1**. A schematic diagram to illustrate the working principle of the electricity-driven EDR membrane process.

For agricultural applications, EDR stands out compared to RO and NF. While RO/NF remove nearly all dissolved salts, including essential nutrients, and require expensive pretreatment chemicals, EDR selectively removes unwanted chloride without depleting beneficial minerals. Its ability to operate efficiently on water with low-to-moderate TDS makes it an ideal choice for irrigation. Although EDR does not remove uncharged contaminants like boron, this is generally not a concern for freshwater sources used in agriculture, particularly in California. Given its high efficiency, lower operating costs, and targeted desalination approach, EDR is a superior choice for agricultural irrigation water treatment.

#### 8. Work plan and methods

As part of the bench-scale work and field demonstration, the team will collect sufficient data to properly develop, demonstrate and validate the *electrodialysis* system for irrigation water chloride removal. Chloride concentration in untreated and treated water samples will be quantified by an ion chromatography coupled with a conductivity detector. Conductivity of the water samples will be measured using a conductivity meter. Sample analysis will follow strict Quality Assurance/Quality Control (QA/QC) requirements.

# Task 1. Pre-field bench-scale testing and prototype buildup – Year 1

To baseline the operational parameters of the pilot-scale system and properly select the type of EDR unit and operational parameters required for the treatment of the irrigation water samples that will be used in the field demonstration, we will conduct a series of bench-scale tests by assembling a bench-scale EDR system that will operate in a recirculation mode in the lab at UC Riverside. Real salinity-elevated irrigation water will be collected from the University of California South Coast Research and Extension Center (SCREC) in Irvine, California and used as the feedwater for

treatment (*see attached letter of support from Dr. Pedroncelli, Director of SCREC*). SCREC has 200 acres of fields in an arid/semi-arid region used for growing avocados, various fruit trees and agronomic crops. The irrigation water at SCREC is recycled water produced by Irvine Ranch Water District as a municipal wastewater effluent. This irrigation water is elevated in salinity, with a chloride concentration in the range of 150 to 250 mg/L. This provides an ideal sample of real-world feedwater to evaluate and demonstrate the EDR treatment efficiency.

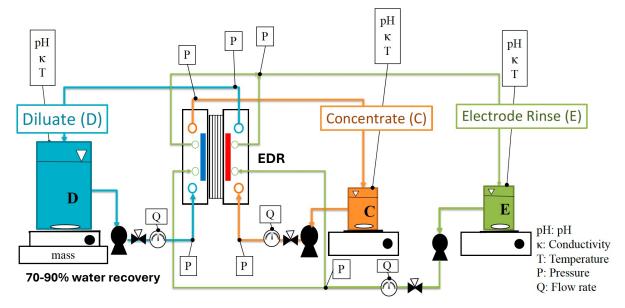


Figure 2 Schematics of the bench-scale electrodialysis (ED) experimental apparatus in recirculation mode.

We will assemble the EDR system and it will mainly consists of the electrodialyzer and three streams: diluate (D), concentrate (C), and electrode rinse (E) (**Figure 2**). The electrodialyzer includes the anode, the cathode, and two end-plates. Between the anode and the cathode, multiple pairs of cation and anion exchange membranes (CEMs and AEMs, respectively), separated by thick plastic woven screen spacers to allow solution flow, are aligned in a repeatable manner (e.g., CEM – spacer – AEM – spacer - ... - spacer - CEM). The anode and cathode will consist of expanded titanium with platinum/iridium coating and are secured to polypropylene end-plates. A small voltage per cell pair will be applied to the electrodialyzer throughout the EDR experiments, and the EDR system will be operated under constant voltage mode.

In this task, each of the three streams will be circulated by laboratory-scale gear pumps. both dilute and concentrate solutions begin with the same feed water. As the system operates, their concentrations change. The water is recirculating throughout the experiment, causing the dilute concentration to decrease and the concentrate concentration to increase. The rinse solution will be made of sodium sulfate with an ionic strength similar to that of the feed water. The flow rates (Q) for the concentrate and diluate will be controlled by digital liquid flow controllers (McMillan Liquid Flo-Controller Model 400-6-A4).

The goal of the treatment is to achieve 70%-90% of the water recovery as diluate treated water, and chloride removal to achieve a final treated water with less than 100 mg/L chloride. To optimize the EDR system to achieve these treatment goals, several EDR operational parameters will be investigated to achieve the best EDR treatment performance. First, three different ion exchange

membranes will be evaluated for the EDR system to achieve the best chloride removal efficiency, including two conventional ion exchange membrane with different surface functional groups, and a third monovalent ion selective membrane that targets chloride removal. Second, we will evaluate the tuning of voltage of applied to the EDR system. The range of voltage applied to each cell pair will be from 0.5 to 5 V. Third, we will optimize the water recovery percentage and match it with the chloride removal goal. It is expected that a higher water recovery combined with a lower voltage applied can achieve the desirable chloride removal.

# Task 2. Field Demonstration of Pilot-Scale EDR System – Years 2-3

In this task, a pilot-scale electrodialysis reversal (EDR) system will be deployed and demonstrated over a 3 to 6-month period at a selected avocado grove in Southern California. The system will be designed to treat chloride-impacted irrigation water at a significantly larger scale, processing approximately 2,000 gallons at a flow rate of 1–2 gallons per minute (gpm). The EDR system will operate in recirculation mode, ensuring optimal chloride removal. If a single pass through the system does not achieve the desired chloride reduction, the treated water will be recirculated back to the start of the block flow diagram for additional treatment. The treated water will be used for irrigation of avocado trees, and its impact on tree growth and productivity will be evaluated. The production rate of the trees irrigated with low-chloride treated water will be compared to a control group of trees irrigated with high-chloride irrigation water to assess the benefits of chloride reduction on crop health and yield. To monitor chloride removal efficiency, daily grab samples will be collected and analyzed for chloride concentration throughout the demonstration period, enabling continuous process evaluation and optimization.

# Task 3. Estimate the energy and total cost of the pilot-scale and future full-scale operation – Year 3.

An economic analysis of the EDR chloride removal technology will be developed to predict cost of future scale operations based on the results from the field demonstration and chloride removal kinetics. Capital, operating and maintenance (O&M) costs will be included in the economic analysis. Capital costs of treatment components will be estimated using "Cost Build-up Approach" which is based on vendor quotations, cost estimating guides, and best professional judgment. The annual capital cost will be estimated from an appropriate capital recovery factor using the net present value (NPV) method. The O&M costs will be calculated based on experimental results in this study that considers the electric energy and chemical consumption costs. In addition, the limited volume of brine concentrate disposal options will be evaluated and incorporated into the overall cost.

# 9. Project Outreach

Considering the urgency, relevance, importance and promise of chloride removal from irrigation water, the development of efficient water treatment technologies to selectively remove chloride can become a game-changer for the Californian avocado industry to increase its profit and enhance its global competitiveness. Outreach methods will include extension publications with SCREC websites, article publication and progress update via the in the California Avocado Commission's quarterly magazine *From the Grove*, on-site field days at SCREC, in-person or virtual grower meetings, communications with CAC committees and other industry partners as appropriate. The PI has conducted these proposed outreach activities during the Phase-one chloride technology review CAC project.

# 10. Milestone Table

The research work plan of individual tasks and significant milestones is developed as below.

| Task 1: Preliminary Testing at UCR                                      |          |
|---|----------|
| Subtask 1: Construct the EDR system in recirculation mode               | Year 1   |
| Subtask 2: Collect feedwater from Extension SCREC Partner               | Year 1   |
| Subtask 3: Evaluate EDR lab-scale optimization for chloride removal     | Year 1   |
| Subtask 4: Collect data and Quantify the total energy dosage requirment | Year 1   |
| Task 2: Field demonstration and testing at SCREC                        | Year 2   |
| Subtask 1: Design and construct the field demonstration pilot           | Year 2   |
| Subtask 2: Update site readiness  | Year 2   |
| Subtask 3: Transport and install the pilot system                       | Year 2   |
| Subtask 4: Conduct EDR pilot demonstration at Extraction Point          | Year 2-3 |
| Subtask 5: Perform analytical pause and validate performance            | Year 2-3 |
| Subtask 6: Decommission the pilot system                                | Year 2-3 |
| Task 3: Data energy cost calculation and final report                   | Year 3   |
| Subtask 1: Anlayse data   | Year 3   |
| Subtask 2: cost calculation   | Year 3   |
| Subtask 3: final report   | Year 3   |

# **Budget Table**

|                              | CAC FY 1           | CAC FY 2           | CAC FY 3           | Total     |
|------------------------------|--------------------|--------------------|--------------------|-----------|
|                              | 11/01/25 to        | 11/01/26 to        | 11/01/27 to        |           |
|                              | 10/31/26           | 10/31/27           | 10/31/28           |           |
| Principal                    | \$19,945           | \$20,743           | \$21,572           | \$62,260  |
| Investigator (PI)<br>salary  |                    |                    |                    |           |
| PI benefits (7.9% of salary) | \$1,576            | \$1,639            | \$1,704            | \$4,919   |
| Graduate Student             | \$40,174           | \$43,288           | \$46,643           | \$130,105 |
| Researcher (GSR)             |                    |                    |                    |           |
| salary                       |                    |                    |                    |           |
| GSR benefits                 | \$22,282           | \$23,223           | \$24,211           | \$69,716  |
| (2.1% of salary +            |                    |                    |                    |           |
| tuition fee                  |                    |                    |                    |           |
| remission)                   |                    |                    |                    |           |
| Travel                       | \$1,000            | \$1,000            | \$1,000            | \$3,000   |
|                              | (Car rental \$400, | (Car rental \$400, | (Car rental \$400, |           |
|                              | and                | and                | and                |           |
|                              | lodging \$600)     | lodging \$600)     | lodging \$600)     |           |
| Materials and                | \$10,000           | \$10,000           | \$10,000           | \$30,000  |
| Supplies                     |                    |                    |                    |           |
| Total                        | \$94,977           | \$99,892           | \$105,131          | \$300,000 |

# **Budget Narrative**

This budget requests \$300,000 for three years beginning November 1, 2025. Details of this request are provided below.

#### Personnel

Haizhou Liu, Professor of Chemical and Environmental Engineering, (1.0 summer months in each project year) will serve as the PI of this grant/project and will assume its administrative responsibility. In addition, he will oversee the design and implementation of the whole project, and supervise the graduate student researcher (GSR) who will work on this project. The salary requested is based on actual rates, and escalated by 4% annually, as per institutional policy.

One TBN Graduate Student Researcher (GSR), starting at increment 1, is requested at 4.5 academic months and 1.92 summer months for each project year. This GSR, under the supervision of Prof. Liu, will work on all proposed research tasks. The salaries requested are based on the University's published salary scale for GSRs.

#### Benefits

The University's Federally approved composite benefit rates (CBR) are for the period July 1, 2024 through June 30, 2025, and provisional thereafter per Department of Health and Human

Services (DHHS) agreement dated April 9, 2024. The CBR for faculty summer is 7.90% and that for students is 2.10%. The University includes graduate student tuition/fee remission in benefits. These costs are as follows.

| Student fee remission 2025-26 AY | \$21,439 |
|----------------------------------|----------|
| Student fee remission 2026-27 AY | \$22,314 |
| Student fee remission 2027-28 AY | \$23,232 |

# Travel

This budget requests \$1,000 for each project year for domestic travel by the PI and GSR to attend the California Avocado Society Annual Meeting and another agriculture-themed national conference, as well as and periodical visits of partner avocado groves to collect salinity-elevated irrigation water for technology testing and demonstration. For each year, \$600 is requested for lodging and \$400 for transportation. This estimate is based on the PI's experience from previous travel.

# **Materials and Supplies**

\$10,000 is requested for each project year for the purchase of lab consumables that are critical to the operation of the chloride desalination system and analytical consumables that measures chloride, including tubing, ion exchange membranes, water chambers and containers, peristatic pumps, holding tanks, metal beams, timers and pressure valve for pilot-system setup, electrodes for the electrodialysis units, ion chromatography sample vials, analytical columns that measure chloride, conductivity probe, beakers, volumetric flasks needed to carry out the proposed work. This estimates is based on the PI's experience from previous similar purchases.

#### **UNIVERSITY OF CALIFORNIA** Agriculture and Natural Resources

South Coast Research and Extension Center

March 14, 2025

Production Research Committee California Avocado Commission

#### Re: Letter of Support from UC ANR South Coast Research and Extension Center

Dear California Avocado Commission Production Research Committee:

I am writing this letter to enthusiastically support Dr. Liu's proposal titled "Development and Demonstration of a Cost-effective Electrodialysis Reversal (EDR) Process for chloride removal from Avocado Irrigation Water". As the director of the South Coast Research and Extension Center (South Coast REC), I will collaborate with Dr. Liu to provide recycled wastewater effluent as irrigation feedwater and the site for his team to demonstrate the treatment of recycled wastewater effluent to remove chloride from irrigation water.

As part of the University of California (UC) division of Agriculture and Natural Resources (ANR), South Coast REC was established in 1956 as a representative site for agricultural and horticultural research in California's south coastal plain-temperate climatic zone. South Coast REC serves as a regional field laboratory for UC scientists to conduct agricultural and natural resources management research and extend research-based information to a wide spectrum of audiences. The Center provides land, irrigation water, labor, equipment, and other facilities, and it serves as a repository for germplasm collections of many subtropical plants. Intensive research efforts are focused on fruits and vegetables. The Center is also complemented by supporting work in entomology, plant pathology, biological control, and integrated pest management. Staffing at South Coast REC consists of multiple full-time equivalent employees engaged in administration, education outreach, and agricultural field and physical plant operation. South Coast REC is also home to the UC Cooperative Extension Orange County office, with multiple full-time programmatic and research academics and staff.

I am excited about this opportunity to collaborate with Dr. Liu on this project and look forward to new collaborations with Dr. Liu at South Coast Research and Extension Center.

Sincerely,

Lindsey Pedroncelli

Lindsey Pedroncelli, Ph.D. Interim Director, South Coast Research and Extension Center

#### Title: Impact of Natural Vegetation on Insect Pollinators in Agroecosystems

Principal Investigator: Carson Loudermelt, graduate student, Cal Poly Pomona Co-Principal Investigator: Dr. Hamutahl Cohen, Assistant Entomology Advisor, Ventura, UC ANR Co-Principal Investigator: Dr. Adam Lambert, Associate Researcher, UC Santa Barbara Co-Principal Investigator: Dr. Elizabeth Scordato, Associate Professor, Cal Poly Pomona

#### **Research Problem & Project Synopsis**

The demand for pollination services in agriculture frequently exceeds the supply (Mashilingi et al. 2022). This is a particular problem for the avocado industry. Avocado growers typically rely on managed honeybee populations for pollination of avocados, but the most effective pollinators of this crop are likely solitary bees, wasps, and flies. In fact, when wild pollinators are present, avocado crops can have a more than 25% increase in production (Lara-Pulido et al 2021). Furthermore, declining wild pollinator populations have been shown to adversely impact avocado yields (Biesmeijer et al., 2006). However, it is unclear which species are the most common avocado visitors and how growers can support these wild pollinator populations through management practices (Lara-Pulido et al 2021), especially in Ventura County. While avocado visitors have been identified in Mexico and Central America (Can-Alonzo et al. 2005), the pollinators of avocados have never been described in California. We know that crop visitation by pollinators and pollinator diversity increases with the surrounding natural habitat, which improves crop yield (Eeraerts et al 2021). However, there is no consensus on the optimal distance from orchards or the size of natural vegetation patches required to achieve these benefits. While many growers already take steps to protect wild bees, we still have a limited understanding of how land management practices at different spatial scales affect bees and other insects that are potentially pollinating avocado flowers. This gap in knowledge leaves avocado growers without relevant guidelines for using non-crop vegetation to support pollinators, even though many show interest in enhancing natural habitats for improved ecosystem services (Esquivel et al 2021). Avocados are likely dependent upon a unique community of pollinator species, so it is important to address how these pollinators respond to natural vegetation at different spatial scales (Sagwe et al 2022). The goal of this project is to provide clear, applicable recommendations to help growers establish natural vegetation on orchard margins to enhance pollinator visitation and diversity, ultimately supporting avocado yields. We will share the results of our work through at least one field day, a minimum of two blog posts through the UC ANR Topics in Subtropics blog, and communication with the California Avocado Society.

#### **Objectives**

The <u>first objective</u> of this project is to identify the species of pollinator insects that are responsible for pollination in avocado crops. We hypothesize that certain species of bees, flies, wasps, and other insects may play a key role in the transfer of pollen between avocado flowers. To achieve this objective, we will conduct visitor observations along our transects during the blooming period of avocado trees in our orchards. This will provide information on what species may be contributing to the pollination of avocados, possibly providing evidence of any flies, solitary bee species, or other types of insects pollinating avocados.

The <u>second objective</u> of this project is to evaluate how different features of orchards, both at local and landscape scales, influence pollinator diversity and abundance. To achieve this objective, we will be

sampling pollinators within our orchards that have varying quality and diversity of natural habitats surrounding the orchards, at local and landscape scales.

### **Study Design**

This study will be conducted in eight avocado orchards and four riparian sites throughout Ventura County. At each orchard site, we will establish a transect that is 150 meters long, running from the edge of an orchard block to the center of the block. Half of the orchard research sites will have bare margins and half will have vegetated margins (either planted hedgerows or naturally-occurring native vegetation). Additionally, the sites vary in distance to natural riparian habitat on the landscape scale. We will use sites in the riparian channel to catalog pollinator species that could be found in orchards, therefore using them as a control for pollinator diversity (Figure 1).

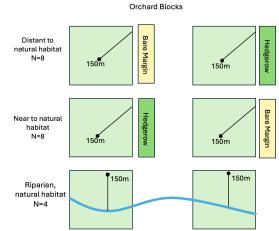


Figure 1. Study design in the SCRV

To accomplish objective 1, we will conduct pollinator visitation surveys along our transects. Observers will implement three-minute visual observations within one meter-squared quadrant at eight trees along the transect, followed by three minutes of vouchering to collect insects observed in the visual survey. Visual observations will include all specimens seen touching parts of an open flower. The quadrants will be flagged and we will return 5 months later to count fruits and measure height and width.

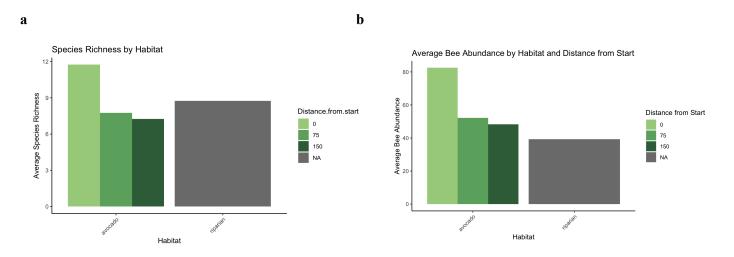
To accomplish objective 2, we will survey pollinators using pan traps and blue vane traps at each site. These traps will be set in openings next to trees at the 0m, 75m, and 150m points along the transect, and insect pollinators will then be transferred to the lab for identification to the lowest taxonomic unit possible. We will characterize variations in pollinator abundance, diversity, and community structure among riparian transects, orchards adjacent to the riparian corridor, and orchards distant from the riparian corridor. To assess how hedgerow (small-scale plantings along orchard margins) and larger riparian landscape composition and structure impact pollinator communities, we will collect and incorporate data on non-crop vegetation and flower abundance and diversity. Information on the composition and structures of the hedgerows, located along the margins of some of the orchards, will be used to understand how local-scale vegetation features affect pollinator communities within different landscapes. Additionally, flower abundance and diversity will be measured along the transects at the 0m, 75m, and 150m points, to assess floral resource availability at different orchards and riparian sites. To assess the impacts of landscape composition on pollinator communities, we will evaluate the percent of non-crop vegetation within 100, 250, 500, and 1000-meter buffers around transect points using ArcGIS. This data will provide insight into the broader landscape vegetation structure that could potentially serve as habitats or resources for pollinator communities. By examining the combination of these local and landscape features along with pollinator communities at each site, we aim to determine what characteristics of these heterogeneous landscapes support more diverse and abundant communities of pollinators.

#### **Data Analysis**

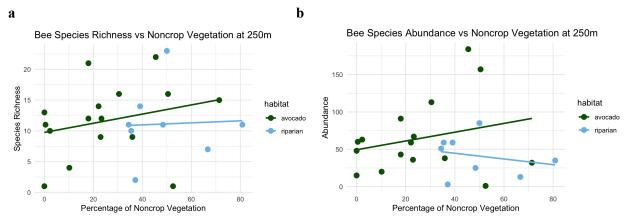
With the collected data, we aim to explore the relationship between pollinator diversity, abundance, and various environmental variables at local and landscape scales. We will use generalized linear mixed models (GLMMs) to explore how vegetation/floral composition and structure at the local and landscape scales influence pollinator diversity and abundance. Predictor variables will include transect flower cover and vegetation composition and the percentage of non-crop vegetation at the landscape scale, with site included as a random effect to account for site variation. Additionally, we will use Non-Metric Dimensional Scaling (NMDS) with Bray-Curtis dissimilarity to examine the overall community structure of pollinators to visualize patterns of how community composition relates to our environmental variables. This approach will allow us to better understand the local and landscape features that impact pollinator communities and affect agricultural production.

#### **Preliminary Data**

Preliminary analysis shows that average bee species richness and abundance are similar in both avocado and riparian sites. Riparian sites and points on the margins of our avocado orchards (0 meters) have higher species richness than points within the interior of the avocado orchards (fig. 2a). We also found that the average bee abundance is higher in avocado orchards than riparian, with the trees adjacent margins, (at 0 meters along our transect), harboring the highest abundance (fig. 2b). From preliminary analyses, we are also seeing that as non-crop vegetation increases within all of our buffers (100, 250, 500, and 1000 meters) bee species richness within avocado orchards increase as well, shown in figure 3 in the 250-meter buffer. Here, we propose to expand this work by collecting more insect pollinator data at more transects. More visual observations and pollinator samples at more transects will help us be more sure that our data captures the true pollinator communities and how they respond to the natural landscape.



**Figure 2.** (a) Average bee species richness in avocado and riparian transects, with color corresponding to distance from the orchard margin with 0 being exterior and 150 being 150 meters into the interior. (b) Average bee abundance between avocado and riparian transects, with color corresponding to distance from the margin of the avocado orchard.



**Figure 3:** (a) Bee species richness across percent noncrop vegetation within a 250-meter buffer, colored by habitat type (avocado and riparian) (b) Bee abundance across percentage of noncrop vegetation within a 250-meter buffer.

With this project, we hope to enhance our understanding of the relationship between pollinator diversity, abundance, avocado yields, and local and landscape vegetation features to provide tractable and actionable recommendations to help support sustainable avocado farming and preserve essential pollinator communities within these agroecosystems.

#### **Support from CAC**

Support from CAC is critical for the success of this project and supports the training of PI Carson Louderment, a graduate student interested in pursuing entomology and agricultural research. Furthermore, this project will support the training of one undergraduate assistant in field methods in Ventura County, which faces a lack of trained agricultural sciences personnel.

| Budget  | Description  | Year 1<br>(July 1, 2025 - Oct<br>31, 2025) | Year 2<br>(Nov 1, 2025 - Oct<br>31, 2026) |
|---|--|--|---|
| Travel to the field and<br>outreach events from<br>Pomona | Gas & mileage: 67 cents/mile<br>~ 180 miles round trip<br>~40 miles between sites<br>~ 7 trips | \$516                                      | \$516                                     |
| Accommodations  | Hotel 2 nights/trip<br>~7 trips<br>~\$200/night  | \$1,400                                    | \$1,400                                   |
| Food per diem   | \$25/day<br>One assistant<br>~3 days per trip<br>~7 trips                                      | \$525                                      | \$525                                     |
| Collection equipment                                      | nets, pans, vials, coolers, vane<br>traps  | \$300                                      |   |
| Identification costs                                      | Insect pins, Cornell drawers,  | \$250                                      | \$250                                     |

|  | shipping samples to experts                  |       |                |
|--|--|-------|----------------|
| Undergraduate Insect<br>Identification Assistant | \$17/hour<br>~100 hours                      | \$850 | \$850          |
| Undergraduate field assistant                    | \$16.50/hour<br>~21 field days<br>~120 hours | \$990 | \$990          |
|  |  |       | Total: \$9,362 |

# Milestone table

| Milestone                                   | Estimated<br>Completion Date | Estimated budget<br>amount |
|---|------------------------------|----------------------------|
| Complete surveys in SVRC                    | July 2026                    | \$6,130                    |
| Complete identification of surveyed insects | September 2026               | \$2,200                    |
| Complete data analysis                      | September 2026               | -                          |
| Outreach events                             | July 2026                    | \$1,032                    |
| Submit research for publication             | October 2026                 | -                          |

# References

- Biesmeijer, J. C., et al. "Parallel Declines in Pollinators and Insect-Pollinated Plants in Britain and the Netherlands." Science (New York, N.Y.), vol. 313, no. 5785, July 2006, pp. 351–54. PubMed, https://doi.org/10.1126/science.1127863.
- Can-Alonzo, Cesar, et al. "Pollination of 'Criollo' Avocados (Persea Americana) and the Behaviour of Associated Bees in Subtropical Mexico." Journal of Apicultural Research, vol. 44, no. 1, Jan. 2005, pp. 3–8. DOI.org (Crossref), https://doi.org/10.1080/00218839. 2005.11101138.
- Eeraerts, Maxime. "A Minimum of 15% Semi-Natural Habitat Facilitates Adequate Wild Pollinator Visitation to a Pollinator-Dependent Crop." Biological Conservation, vol. 278, Feb. 2023, p. 109887. DOI.org (Crossref), https://doi.org/10.1016/j.biocon.2022. 109887.
- Esquivel, Kenzo Emiliano, et al. "The 'Sweet Spot' in the Middle: Why Do Mid-Scale Farms Adopt Diversification Practices at Higher Rates?" Frontiers in Sustainable Food Systems, vol. 5, Oct. 2021, p. 734088. DOI.org (Crossref), https://doi.org/10.3389/fsuf s.2021.734088.
- Lara-Pulido, José Alberto, et al. "A Meta-Analysis of Economic Valuation of Ecosystem Services in Mexico." Ecosystem Services, vol. 31, June 2018, pp. 126–41. DOI.org (Crossref), https://doi.org/10.1016/j.ecoser.2018.02.018.
- Mashilingi, Shibonage K., et al. "Honeybees Are Far Too Insufficient to Supply Optimum Pollination Services in Agricultural Systems Worldwide." Agriculture, Ecosystems & Environment, vol. 335, Sept. 2022, p. 108003. DOI.org (Crossref), https://doi.org/10.1016/j.agee.2022.108003.
- Sagwe, Rose Nyakemiso, et al. "Pollinator Efficiency of Avocado (Persea Americana) Flower Insect Visitors." Ecological Solutions and Evidence, vol. 3, no. 4, Oct. 2022, p. e12178. DOI.org (Crossref), https://doi.org/10.1002/2688-8319.12178.

# Improve *Phytophthora cinnamomi* management by monitoring field populations for changes in fungicide sensitivity and conducting efficacy field trials

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**Project Co-PI:** Dr. James Adaskaveg, Microbiology and Plant Pathology Department, The Regents of The University of California, 245 University Office Building, Riverside, CA 92521, jim.adaskaveg@ucr.edu, (951)827-7577.

## **EXECUTIVE SUMMARY**

Phytophthora root rot (PRR), caused by *Phytophthora cinnamomi (Pc)*, is one of the most devastating avocado diseases worldwide. PRR severity and incidence are exacerbated under flooding and hypoxic conditions caused by inappropriate irrigation practices and soil waterlogging conditions, which are common conditions in California (CA). This oomycete root pathogen causes trunk cankers, leaf chlorosis, leaf defoliation, and kills feeder roots reducing fruit yield. This invasive pathogen spreads rapidly and is prevalent in many agricultural systems, attributable to its adaptability to new environments, broad host range, saprophytic capabilities, host resistance, and production of resilient structures for survival and dispersal<sup>1,2,3</sup>. PRR control methods include cultural practices including the use of resistant rootstocks like 'Dusa' and fungicidal treatments such as potassium phosphite (PP), mefenoxam (Ridomil Gold), and oxathiapiprolin (Orondis). Orondis was recently registered to manage avocado PRR based on greenhouse and field efficacy trial results conducted by the Manosalva and Adaskaveg teams<sup>2,4</sup>. Growers have been relying on the combination of using 'Dusa' and field treatments of PP for managing PRR, however, *Pc* isolates, are overcoming these practices by becoming more virulent and developing PP resistance in CA<sup>1-4</sup>.

# Phytophthora cinnamomi populations in California exhibit large variability in fungicide sensitivity.

Manosalva and Adaskaveg's teams have been reporting a shift towards PP insensitivity in Pc populations collected from CA avocado orchards. We reported that isolates obtained between 2004 and 2017 from Riverside and San Diego counties exhibiting EC<sub>50</sub> values (the concentration to inhibit Pc mycelial growth by 50%) of as high as 382.5  $\mu$ g/ml as compared to <25  $\mu$ g/ml for sensitive isolates<sup>1,2</sup> (Fig. 1). We also reported that the more PP insensitive Pc isolates (Riverside and San Diego counties) were also more virulent on avocado rootstocks. We have detected Pc isolates with high EC<sub>50</sub> values for PP (up to 763 ug/ml) also in Santa Barbara and Ventura counties in 2020 and 2022 (Fig. 1). This insensitivity likely reflects the continued overuse of PP applications in orchards and

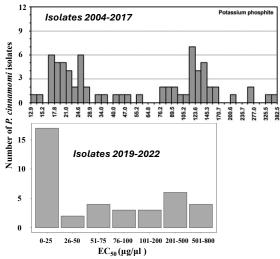


Figure 1. Frequency histograms of effective concentrations to inhibit *P. cinnamomi* mycelial growth by 50% ( $EC_{50}$ ) for potassium phosphite. Bar height = number of isolates within each bin. Bin widths were calculated using Scott method.

subsequent spread of PP insensitive isolates from southern CA areas to Ventura and Santa Barbara Co. Thus, it is critical that we continue surveying and monitoring the pathogen population to develop more effective protocols for disease chemical control based on fungicide rotations (i.e., PP + Ridomil Gold/ Ridomil Gold + Orondis/ PP + Orondis). Manosalva and Adaskaveg evaluated the *in vitro Pc* sensitivity to additional chemistries including ethaboxam (Elumin), fluopicolide (Presidio), mandipropamid (Revus), oxathiapiprolin (Orondis), and mefenoxam (Ridomil Gold) and found that all these Oomycota-targeting fungicides exhibited high *in vitro* toxicity with relatively low effective concentrations to inhibit *Pc* mycelial growth and found significant variability among isolates<sup>1,2</sup>. This range in sensitivities probably reflects natural variation within the pathogen populations since with the exception of oxathiapiprolin and mefenoxam, these fungicides have not been registered or approved for use on avocado but are registered on other crops. Our recent studies with isolates obtained from 2019 to 2022 also indicated that the sensitivities to these fungicides with the exception on PP have not changed. Orondis was registered on avocado in 2022 and since then, more growers are applying Orondis to control PRR in their orchards. The majority of these Oomycota fungicides have a single target gene which increases the risk for resistance development. Moreover, resistance to these chemistries has been found in Oomycota pathogens including *Phytophthora* spp.<sup>4-8</sup>. Thus, it is critical to continue the survey of CA orchards and gather information regarding frequency of Orondis applications and if rotations were used of Orondis with PP/Ridomil Gold. More importantly, isolates from these orchards needs to be collected to determine their EC<sub>50</sub> to Orondis and to the other chemistries to monitor for any shift in the current CA pathogen populations. *Note that these chemistries have been registered on other crops including citrus so there is still a risk for exposure in orchards applying these on citrus in the proximity of avocado orchards.* 

## A combination of fungicides and new Pc UCR resistant rootstocks results in a better PRR protection under

greenhouse and field conditions. The University of California Riverside (UCR) rootstock breeding program has developed and evaluated under greenhouse (GH) & field conditions, five UCR advanced Pc resistant rootstocks (PP40, PP35, PP42, PP45, and PP80) which also exhibit salinity tolerance (PP40, PP35, and PP80), another major production challenge. These UCR rootstocks grafted to 'Hass' in combination with these new fungicides were tested for their efficacy in controlling PRR under GH conditions. All fungicides

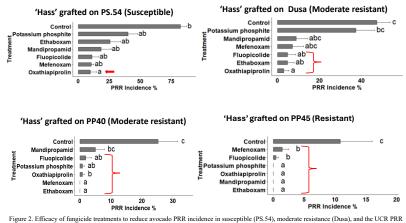


Figure 2. Efficacy of fungicide treatments to reduce avocado PRR incidence in susceptible (PS.54), moderate resistance (Dusa), and the UCR PRR resistant rootstocks (PP40 & PP40 & PP45) grafted to 'Hass' under greenhouse conditions. Statistics were done using generalized linear mixed models (GLMMs) and LSMeans tests at P<0.05 using R. Different letters above the bars indicate significant differences

reduced the PRR incidence caused by a mixture of the most virulent isolates when compared to untreated inoculated control plants. Oxathiapiprolin, mefenoxam, and fluopicolide outperformed ethaboxam, mandipropamid, and PP. Some fungicides paired with the most resistant rootstocks had a synergistic effect, enhancing PRR control (**Fig. 2**). These new UCR rootstocks will be released in 2025-2027, and the new Oomycota fungicides described above will be registered by 2026. This integrated PRR management strategy holds promise for growers by adopting new resistant rootstocks in combination with appropriate fungicide treatments, however, the effectiveness and durability of these new control methods still deserves extensive evaluation due to the great genome plasticity and adaptative capacity of *Pc* populations<sup>3</sup>. The combination of resistant rootstocks and fungicide rotations or mixture rotations will be desirable to reduce the pathogen selection pressure for breaking the rootstock resistance and developing chemical insensitivity.

In 2018, Adaskaveg, established two fungicide field trials with Duke 7 and Dusa® rootstocks under heavy PRR disease pressure (natural infection). Soil applications of oxathiapiprolin, ethaboxam, fluopicolide, and mefenoxam alone and in combinations were compared to untreated controls and to tree injection with PP (standard grower treatment). Oxathiapiprolin and fluopicolide alone and in combinations with other fungicides were the best treatments for reducing PRR incidence<sup>4</sup> (Fig. 3). These studies are important to determine the best rotation protocols and the different combinations that growers can use for PRR control in their orchards and reduce the risk of *Pc* resistance to

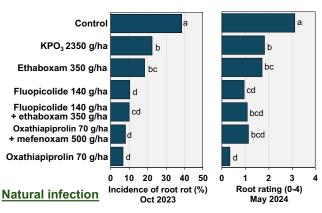


Figure 3. Efficacy of fungicides for managing avocado PRR of 'Hass' trees grafted on Dusa rootstocks in a commercial field trial in Riverside Co., CA established in 2022.

recently registered fungicides or in the pipeline for federal registration through IR-4.

Our overall goal is to ensure the long-term sustainability and competitiveness of the CA avocado industry by reducing production inputs and yield losses due to avocado PRR and by decreasing the risk for the emergence of fungicide resistant pathogen populations threatening the durability and efficacy of current and future chemical PRR control. Thus, in this project, we will continue monitoring Pc populations in CA by conducting extensive surveys in orchards throughout CA especially where Orondis, Ridomil Gold, and PP are used to: i) determine their current fungicide sensitivity; ii) assess if the sensitivities to mefenoxam and oxathiapiprolin have been changing; and iii) determine if more isolates are acquiring PP insensitivity and if they continue to spread through CA growing regions. Resistance assessments for fungicides (except for PP) will be conducted alone and in mixtures to: i) determine the risk for shifting baseline sensitivities and acquiring resistance from over usage (i.e., multiple & sequential applications); ii) assess how fungicide mixtures will affect the risk for emergence of resistance; and iii) determine if the use of resistant rootstocks can further reduce the emergence of Pc fungicide resistance. Finally, we will continue collecting data including vield from our current fungicide efficacy field trial and will establish a replicated trial in Ventura. These efficacy trials will allow us to: i) test different timings of application to reduce the negative effects of PRR in tree health and productivity, ii) determine the anti-resistance rotation and mixture programs to set sustainable and durable protocols for PRR control in CA, and iii) compare results between two environmental distinct growing areas in terms of pathogen population, climate, irrigation water quality, and soil conditions.

## **PROJECT OBJECTIVES.**

**Objective 1.** Survey *Pc* populations across major CA avocado growing regions and assess their *in vitro* sensitivities to registered and new Oomycota fungicides to compare those with established baseline sensitivities. Information regarding cultural practices and fungicide history applications at each orchard surveyed will be recorded. Resistance assessments of fungicides alone and in mixtures will be conducted using a genetically and phenotypically representative *Pc* populations under laboratory and greenhouse conditions.

#### Deliverables

- Collection of current *Pc* populations (2025-2028) for which their *in vitro* sensitivities for potassium phosphite (PP), oxathiapiprolin (Orondis), mefenoxam (Ridomil Gold), ethaboxam (Elumin), fluopicolide (Presidio), and mandipropamid (Revus) will be determined.
- Information regarding the continue increase of *Pc* EC<sub>50</sub> values for PP (> 763 μg/ml) in the same orchards surveyed before or new orchards. We will continue getting insights for emergence of PP insensitive isolates by gathering information regarding PP application rates and frequency, rootstocks, raining events, production data, etc. We will provide recommendations to reduce the emergence and spread of this PP insensitivity *Pc* populations.
- Ridomil Gold and Orondis baseline sensitivities for the current *Pc* populations especially for isolates collected from orchards where these products have been used. The presence/risk of isolates exhibiting a shift towards fungicide insensitivity will be determined and correlated with cultural practices. Thus, we can provide recommendations on how to delay/avoid the emergence of Orondis and Ridomil Gold resistant isolates early in the process.
- Expanded baseline sensitivities for ethaboxam, fluopicolide, and mandipropamid that are currently not registered on avocado to confirm the previously published baselines.
- Fungicide resistance assessment experiments will provide critical information to assess: i) how many single applications of fungicides will be required to gain insensitivity/resistance, ii) how to best rotate registered products to avoid/delay the emergence of fungicide resistance, and iii) provide an integrated management for PRR control and *Pc* fungicide resistance management by combining the more effective fungicide rotation protocols with host resistance to increase the durability of current controls methods.

**Objective 2.** Conduct fungicide efficacy trials under commercial conditions to determine the best protocol to maximize chemical protection and reduce the emergence of *Pc* resistant isolates. We will continue the evaluation of several fungicides alone (potassium phosphite, oxathiapiprolin, mefenoxam, ethaboxam, and fluopicolide), in combination, and in mixture rotations of different modes of action (e.g., FC49+FC4, i.e., Orondis Gold sold by Syngenta Crop Protection, rotated with FC22+FC43 [both sold by Valent USA]). In the absence of PP resistance, PP can be mixed with any of the other modes of action to reduce PRR incidence and

damage in plant health and productivity in our current trial of 'Hass' trees grafted to Dusa rootstocks in Riverside Co. We will also establish a similar fungicide efficacy trial in a commercial orchard in Ventura Co.

## Deliverables

- Provide different alternatives of effective fungicide mixtures and rotation protocols that growers can use in their orchards to manage PRR and reduce the risk of emergence of fungicide resistant isolates.
- Share the data and results with extension agents and farm advisors so these protocols and recommendations can be disseminated to all CA growers.

### WORK PLAN AND METHODS

**Objective 1.** To accomplish this, we have divided this Obj. 1 in several activities:

**1.1.** Survey avocado orchards and isolate Pc (April-May 2026, 2027, and 2028). We will select avocado orchards to survey and visit them to collect samples by several approaches. We will visit orchards based on previous collections conducted by the Manosalva and Adaskaveg labs and through advertisements of the objectives of this project requesting information and participation of avocado stakeholders willing to have their orchards surveyed and tested. Surveys will also be conducted in collaboration with farm advisors, the California Avocado Society (CAS), California Avocado Commission (CAS), and Avocado Growers of California (AGC) members which always support the UCR avocado rootstock breeding program and the Manosalva Lab research activities. At each visit we will gather as much information from the growers regarding their grove establishment (i.e., year, rootstocks and scions, size of grove, etc.), and management practices (i.e., fertilization, chemical applications, etc.). Root & soil plating and baiting will be performed as described previously<sup>1,2</sup>. Suspected colonies matching the *Pc* morphological characteristics will be subjected to molecular identification using Internal Transcribed Spacer Region (ITS) sequence analyses and using a TaqMan qPCR assay *Pc*-specific test<sup>1,2</sup>. Single zoospore cultures will be obtained for each confirmed isolate and used in fungicide sensitivity assays.

**1.2.** Fungicide in vitro sensitivity (June-July 2026, 2027, and 2028). The *in vitro* toxicities of oxathiapiprolin (Syngenta Crop Protection), mefenoxam (Syngenta Crop Protection), mandipropamid (Syngenta Crop Protection), ethaboxam (Valent USA), and fluopicolide (Valent USA) to Pc mycelial growth will be determined using the spiral gradient dilution method as described in Förster *et al.* (2004)<sup>9</sup>. For PP sensitivity assays, we will use the traditional agar dilution method<sup>1,2</sup>. Pathogen reference isolates with known EC<sub>50</sub> values will be used as controls in every experiment conducted. Each isolate will be assayed in duplicate, and the experiment will be conducted twice for publication purposes. Data analyses will be conducted as described in Belisle *et al.* (2019b)<sup>2</sup>.

1.3. Assessment of the resistance potential of Pc to fungicides under laboratory and greenhouse conditions (Dec 2026 – Aug 2028). To estimate the *in vitro* potential of resistance development of Pc populations to oxathiapiprolin, fluocopilide, ethaboxam, mandipropamid, and mefenoxam, we will select 20 Pc isolates that are genetically and phenotypically diverse and represent the current CA pathogen population<sup>1-4</sup>. We will select isolates based on geographical location, population structure, sensitivity to PP fungicide (low, mid, and highly resistant), sensitivity based on EC<sub>50</sub> for all other fungicides, virulence phenotypes, etc. We will conduct this

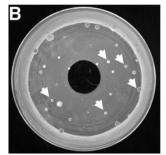


Figure 4. Spiral gradient dilution plates with exponential concentration gradients of fludioxonil ( $EC_{95}$  concentrations were positioned at the edge of the plate). Several putative fludioxonil-resistant colonies (arrows) of *P. digitatum* are found in the clear area of the agar plate treated with fludioxonil.

experiment as described in Chen *et al.*  $(2021)^{10}$ . Briefly, we will calculate the EC<sub>95</sub> (effective concentration to inhibit mycelial growth by 95%) for each selected isolate and use this value in spiral gradient dilution assays where the EC<sub>95</sub> concentrations will be positioned 20 mm from the edge of the Petri dish with exponential dilutions of the fungicides towards the center of the plate (**Fig. 4**). A zoospore suspension with equal parts of the 20 selected isolates described above will be prepared and applied uniformly to each of 10 spiral plates and will be incubated at 25°C in the dark for 3-4 days. Plates will be evaluated for the presence of colonies growing at concentrations above EC<sub>95</sub> values (**Fig. 4**). Data analyses and resistance frequency will be calculated as described in Chen *et al.*  $(2021)^{10}$ . This experiment will be repeated twice. If resistant colonies develop for these fungicides, we will recover them and determine their corresponding EC<sub>50</sub> values. We will conduct similar experiments with the original parental sensitive populations used above and

repeat the experiment but using another fungicide. For example, if we detected resistant colonies to oxathiapiprolin in one batch of 20 parental sensitive isolates, then we will repeat the experiment with oxathiapiprolin and in combination with another fungicide like mefenoxam/PP to determine if the mixture avoids the risk for resistance development.

To estimate the *in vivo* potential for fungicide resistance, we will replicate results from the laboratory assays described above for those fungicides and parental *Pc* populations where resistant populations were obtained. We will inoculate avocado seedlings of the susceptible rootstock Zutano and the soon to be released moderate (PP40), and highly resistant (PP45) UCR advanced rootstocks with the parental sensitive populations and expose the inoculated seedlings to repeated applications with increasing concentrations of the fungicides alone and in mixtures. Fungicide applications and recovery of isolates after each application will be conducted as described in Belisle *et al.* (2019a)<sup>1,2</sup>. We will re-isolate and re-assess the *in vitro* sensitivity of the pathogen populations each cycle of fungicide exposure to detect changes in EC<sub>50</sub> as described above by comparing them with the EC<sub>50</sub> baseline values of the parental sensitive populations. In addition, the emergence of resistant populations will be detected based on the evaluation of virulence that will be calculated as PRR incidence, pathogen propagules per gram of soil (ppg), and root health scorings and comparing them with the untreated inoculated controls and one-time single application treatments.

Caveats and pitfalls. We do not foresee major difficulties in the methods and approaches described in Obi.1.2. & 3 since all protocols described and similar experiments have been successfully conducted at Manosalva and Adaskaveg laboratories. There is a possibility that our in vitro or in vivo resistance assessment assays do not generate Pc resistant populations which might indicated either that methods need to be adjusted or the low risk of Pc to acquire resistance to these chemistries. In this case, we will test new methods to conduct the resistance assessment only for one of the fungicides (e.g., fluopicolide or oxathiapiprolin). For in vitro, we will subculture the isolates and exposing sensitive isolates for several generations to one of these fungicides until resistance arises as described by Miao et al. (2016)<sup>8</sup> and Childers *et al.*  $(2015)^{11}$  (Fig. 5). For *in vivo*, we will use a detached leaf inoculated assay developed by the Manosalva lab to expose and test the parental sensitive population used in the in vitro and conduct resistance assessments as described by Massi et al.  $(2021)^{12}$  (Fig. 6). Based on the combined resistance risk assessment published by FRAC<sup>13</sup>, soil-borne pathogens have a low-risk potential, the risk of FC 49 is low to medium & the agronomic risk is also low with less susceptible rootstocks resulting in a maximum combined risk of 4 to 6 of a possible total of 18. In contrast, a foliar Oomycota disease like grape downy mildew has a combined risk of 12-18 for a FRAC 49 fungicide.

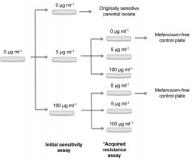


Figure 5. Fungicide exposure method to determine the resistant potential of *P. infestans* isolates to McFenoxam described in Childers *et al.*, (2015) using mycelial plugs plating method of increasing fungicide concentrations.



Figure 6. Leaf disc assay developed to determine the resistant potential of the oomycete *Plasmopara viticola* to fungicides described in Massi *et al.*, (2021).

*Objective 2. Fungicide efficacy field trials (Nov 2025- Sept 2028).* For the continued evaluation of new Oomycota fungicides against avocado PRR, a 50-tree orchard of 'Hass' trees grafted on 'Dusa' located in the Temecula area of Riverside Co. will be used for treatment applications and data collections. PRR incidence and fungicide sensitivity for isolates before and after treatments have been monitored since 2022 and will be continued in this project after each treatment. Seven treatments will be applied twice/year (May & Sept): control (untreated), Orondis 4.8 fl oz/A, Presidio 4 fl oz/A, Elumin 10 fl oz/A, Presidio 4 fl oz/A + Elumin 10 fl oz/A, Orondis 4.8 fl oz/A + Ridomil Gold 14.4 fl oz/A, and Prophyt 64 fl oz/A using 7 trees per treatment in a complete randomized design. Fungicides will be applied to the soil dripline around each tree at the concentration recommended by the chemical companies. The grower will treat trees with PP as a control treatment, and several trees will remain untreated. We will make sure that each treatment contains trees with low-, medium-, and high populations of the pathogen. A second similar fungicide trial will be established in Ventura Co. by adding Orondis + ProPhyt combinations. Before establishing the trial, *Pc* soil populations will be determined. We will locate putative grower collaborators by communication with growers associated or surveyed before by the UCR rootstock breeding program and through advertisements that will be done with the

assistance of farm advisors, CAS, and CAC. We will apply fungicides as described above. At two field sites with a very high incidence and level of PP resistance, we will evaluate the persistence of PP resistance. For this, we will apply rotations of non-PP fungicides for each of the three years of the project and determine if PP resistance levels are stable in the pathogen population or decline over time. We will calculate the efficacy of each treatment 6 months after each application as the reduction of PRR incidence, soil populations, tree health, and production data. Root health will be evaluated visually using a 0 to 4 rating scale with 0 = healthy and 1-4 increasing levels of discolored roots. Data analyses will be done by ANOVA followed by Tukey-Kramer HSD.

*Caveats and pitfalls*. We do not foresee major difficulties in the methods and approaches described in this objective since Dr. Adaskaveg has extensive experience, is an expert on these experiments, and he is already obtained important data at this trial site in Temecula (Fig. 3).

**PROJECT OUTREACH.** We will ensure that our project results, outcomes, and recommendations are delivered and translated into actionable recommendations for growers and other stakeholders with a robust and multi-faceted outreach plan. Manosalva and Adaskaveg research teams are in constant communication with avocado stakeholders including growers, nursery men, industry representatives including chemical companies, and IR-4 staff which will ensure the dissemination of our outcomes and recommendations. By being active collaborators, growers will test and will be direct observants of the results of the projects regarding the best chemical, mixture, and rotation protocols to better control PRR in their orchards decreasing the risk for emergence of fungicide resistance. Outcomes will be also outreached to stakeholders through presentations at CAC, CAS, Avocado Growers of CA (AGC), and UCANR- meetings, workshops. Stakeholders from these groups include conventional and organic growers. Our team will also participate in Avocado Café. We report our progress and outcomes in grower journals, newsletters, and social media.

#### **MILESTONE TABLE**

| Obj. |     | Objective/Sub-task Description   | Year | 1 (No  | v 25  | - Oct 26) | Year | 2 (No  | v 26 - | Oct 27) | Year | 3 (No | v 27 - | Oct 28) |
|------|-----|--|------|--------|-------|-----------|------|--------|--------|---------|------|-------|--------|---------|
| 1    |     | Survey orchards and determine current fungicide in vitro sensitivities       |      |        |       |           |      |        |        |         |      |       |        |         |
|      | 1.1 | Project advertisement and gather orchard & grower information                |      |        |       |           |      |        |        |         |      |       |        |         |
|      | 1.2 | Visit orchards in CA and collect samples & information on cultural practices |      |        |       |           |      |        |        |         |      |       |        |         |
|      | 1.3 | Pathogen isolation, identification, and storage                              |      |        |       |           |      |        |        |         |      |       |        |         |
|      | 1.4 | Conduct in vitro fungicide sensitivity assays                                |      |        |       |           |      |        |        |         |      |       |        |         |
|      | 1.5 | Assessment of resistant P. cinnamomi potential In vitro (laboratory)         |      |        |       |           |      |        |        |         |      |       |        |         |
|      | 1.6 | Seed collection (Zutano, PP40, and PP45) for in vivo greenhouse studies      |      |        |       |           |      |        |        |         |      |       |        |         |
|      | 1.7 | Assessment of resistant P. cinnamomi potential In vivo (in planta, GH)       |      |        |       |           |      |        |        |         |      |       |        |         |
|      | 1.8 | Data analyses and Integration  |      |        |       |           |      |        |        |         |      |       |        |         |
|      | 1.9 | Outreach and publications  |      |        |       |           |      |        |        |         |      |       |        |         |
|      |     | ESTIMATE BUDGET FOR THIS MILESTONES ACTIVITIES                               |      | \$47,  | 843.0 | 0         |      | \$48,9 | 42.00  |         |      | 59,7  | 24.00  |         |
| 2    |     | Fungicide efficacy field trials  |      |        |       |           |      |        |        |         |      |       |        |         |
|      | 2.1 | Continue fungicide treatments alone and mixtures in Temecula trial           |      |        |       |           |      |        |        |         |      |       |        |         |
|      | 2.2 | Continue data collection (Temecula):PRR incidence & tree health              |      |        |       |           |      |        |        |         |      |       |        |         |
|      | 2.3 | Production data collection (Temecula). Depending on 'Hass' price market      |      |        |       |           |      |        |        |         |      |       |        |         |
|      | 2.4 | Identify growers cooperators in Ventura and survey orchards                  |      |        |       |           |      |        |        |         |      |       |        |         |
|      | 2.5 | Design trial and conduct initial PRR assessments at the orchard (Ventura)    |      |        |       |           |      |        |        |         |      |       |        |         |
|      | 2.6 | Start treatments (fungicide alone and mixtures)                              |      |        |       |           |      |        |        |         |      |       |        |         |
|      | 2.7 | Data collection (Ventura): PRR incidence & tree health                       |      |        |       |           |      |        |        |         |      |       |        |         |
|      | 2.8 | Production data collection (Ventura). Depending on 'Hass' price market       |      |        |       |           |      |        |        |         |      |       |        |         |
|      | 2.9 | Data analyses and Integration  |      |        |       |           |      |        |        |         |      |       |        |         |
|      | 2.1 | Outreach and publications  |      |        |       |           |      |        |        |         |      |       |        |         |
|      |     | ESTIMATE BUDGET FOR THIS MILESTONES ACTIVITIES                               |      | \$53.4 | 423.0 | 0         |      | \$56,7 | 54.00  |         |      | 58,2  | 15.00  |         |

#### REFERENCES

<sup>1</sup>Belisle et al. 2019a. Phytopathology 109(3):384-394. DOI: 10.1094/PHYTO-09-17-0326-R.

<sup>2</sup> Belisle et al. 2019b. Plant Disease 103(8):2024-2032. https://doi.org/10.1094/PDIS-09-18-1698-RE.

<sup>3</sup> Shands et al. 2024. Frontiers in Microbiology 15:1341893. DOI 10.3389/fmicb.2024.1341803.

<sup>4</sup>Adaskaveg et al. 2024. Journal of Plant Diseases and Protection 131:1203-1209. https://doi.org/10.1007/s41348-024-00873-6

<sup>5</sup> Matson *et al.* 2015. *Phytopathology* 57:728-736. https://doi.org/10.1111/j.1365-3059.2008.01831.x.

<sup>6</sup>Peng et al. 2019. Phytopathology 109:2096-2106. https://doi.org/10.1094/PHYTO-01-19-0032-R.

<sup>7</sup>Siegenthaler 2021. *Plant Disease* 105:3000-3007. https://doi.org/10.1094/PDIS-08-20-1805-RE.

<sup>8</sup>Miao et al. 2016. Front. Microbiol. 7:615. doi: 10.3389/fmicb.2016.00615.

<sup>9</sup>Föster et al. 2004. Phytopathology 94:163-170.

<sup>10</sup>Chen et al. 2021. Plant Disease 105:2114-2121. https://doi.org/10.1094/PDIS-07-20-1421-RE.

<sup>11</sup>Childers et al. 2015. Phytopathology 105(3):342-349. http://dx.doi.org/10.1094/PHYTO-05-14-0148-R

<sup>12</sup>Massi et al. 2021. Microorganisms 9(1):119. https://doi.org/10.3390/microorganisms9010119.

<sup>13</sup>https://www.frac.info/docs/default-source/publications/pathogen-risk/frac-pathogen-list-2019.pdf

### **PROJECT BUDGET**

| Table 1. Manosalva & Adaskaveg budget description   | 11/01/2025-10/31/2026 | 11/01/2026-10/31/2027 | 11/01/2027-10/31/2028 |
|---|-----------------------|-----------------------|-----------------------|
| Personnel Salary  |                       |                       |                       |
| Ph. D Graduate Student Researcher (GSR), Two academic quarters/year & Summer Quarter in Y3                                | 17,801                | 18,335                | 28,327                |
| Postdoc level I (Jim Adaskaveg)   | 37,141                | 39,864                | 41,059                |
| Personnel Benefits  |                       |                       |                       |
| Ph. D Graduate Student Researcher (GSR), Two academic quarters/year & Summer Quarter in Y3                                | 374                   | 385                   | 595                   |
| Postdoc level I (Jim Adaskaveg)   | 8,282                 | 8,890                 | 9,156                 |
| Tuition & Fees  |                       |                       |                       |
| Ph. D Graduate Student Researcher (GSR), Two academic quarters/year & Summer Quarter in Y3                                | 13,668                | 14,222                | 14,802                |
| Obj. 1 Phytophthora cinnamomi survey, fungicide sensitivities, and resistance assessments                                 |                       |                       |                       |
| Rental Car to travel to Aprox. 10 orchards in Riverside & San Diego areas & 10 in Ventura & Santa Barbara areas           |                       |                       |                       |
| UCR fleet Rental: Sedan car/Cargo Van@ 55.21/day and long rental 552.1/month.   |                       |                       |                       |
| Field Pc isolate collection 2x/year   |                       |                       |                       |
| 5 days to collect data @ Southern CA  |                       |                       |                       |
| 7 days to collect data @ more Northern CA areas   |                       |                       |                       |
| Total 12 days/year (2x)   | \$1,104               | \$1,104               | \$1,104               |
| Hotel for field data collection/ 2 people/2x per year (@180/night/person)   |                       |                       |                       |
| 7 days to collect data @ Northern Trials  | \$4,320               | \$4,320               | \$4,320               |
| Meals for field data collection/ 2 people/2x per year (@79/day/person)  |                       |                       |                       |
| 7 days to collect data @ Northern Trials  | \$2,212               | \$2,212               | \$2,212               |
| Gas/mileage and incidentals   | \$364                 | \$364                 | \$364                 |
| UC Mix soil   | 1000                  | 1000                  | 1000                  |
| Germination pots  | 300                   | 300                   | 300                   |
| Lab general supplies, chemical, and consumables for pathogen isolation, identification, and fungicide in vito sensitivity | 4500                  | 4500                  | 4500                  |
| Rent of 2 benches at GH11C at 130Sq/ft per bench at \$100/month   | 1200                  | 1200                  | 1200                  |
| Sanger Sequencing service at UCR core for ITS sequencing @\$10/sample   | 1000                  | 1000                  | 1000                  |
| Obj. 2 Fungicide field testing in Riverside and Ventura (Jim Adaskaveg)   |                       |                       |                       |
| Hotel for field data collection/ 2 people/2x per year (@180/night/person)   |                       |                       |                       |
| 2 days to collect data @ Northern Trials  | \$2,836               | \$2,836               | \$2,836               |
| Meals for field data collection/ 2 people/2x per year (@79/day/person)  |                       |                       |                       |
| 7 days to collect data @ Northern Trials  | \$800                 | \$800                 | \$800                 |
| Gas/mileage and incidentals   | \$364                 | \$364                 | \$364                 |
| Lab general supplies, chemical, and consumables for pathogen isolation, identification, and fungicide in vito sensitivity | 4000                  | 4000                  | 4000                  |
| SUBTOTAL  | 101,266               | 105,696               | 117,939               |
|   |                       | TOTAL                 | 324,901               |

## BUDGET JUSTIFICATION Total UCR budget requesting for three years: \$324,901

#### Personnel Salary (\$182,527).

Funds are requested to cover the salary for: i) one Graduate Student Researcher (GSR) for two academic quarters each year of the project and one summer quarter in year 3 and ii) one Postdoctoral Researcher Level I at 50% EFT for every year of the project. The GSR will be working under the supervision of Manosalva and will be responsible to conduct all the field, greenhouse, and laboratory research activities described under Obj. 1. In addition, the GSR will be assisting Dr. Manosalva in all the grant reporting activities as well outreaching events to disseminate the results including the writing of publications describing our findings. Pathogen field surveys and collection at all fields will be conducted with the assistance of Manosalva. The GSR I will be working with the Postdoctoral Researcher I in the activities for Obj. 1.3 regarding the resistance assessments of registered products alone and in mixtures under laboratory and greenhouse conditions to estimate the risk of resistance/the potential of resistance and the development of fungicide rotation schemes to prevent emergence of resistant *Pc* populations and optimize efficacy in disease management protocols. The postdoctoral Research I will be working under the supervision of Co-PI Adaskaveg and will conduct the research activities described in Obj. 1.3 and 2. The postdoc will lead & conduct field trials in two locations (Temecula and Ventura Co.) and will be assisting Dr. Adaskaveg in grant reporting activities, outreach events, and writing of publications describing our findings.

**Fringe Benefits (\$27,682).** Employee benefits are estimates, using the composite benefit rates agreed upon by the University of California. The composite benefit rate for the GSR I is 2.1% and for the Postdoc Level I is 22.2%. Subsequent years include increases based on recommendations by our campus administrative office.

**Tuition Fees (\$42,692).** In addition to fringe benefits for the GSR, university policy requires inclusion of partial fees and tuition remission and Graduate Student Health Insurance (GSHIP) for GSR employed during each academic year with an appointment of 25% time or more.

**Domestic travel (\$36,000).** Funds are requested to cover travel of the GSR and postdoc to cover all field activities for the project to conduct pathogen & sample collections, field treatments, and field data collection. **For Manosalva,** travel cost is estimated based on historical data of surveying and collecting samples for *Pc* isolations. We will survey orchards in major avocado growing regions including Riverside, San Diego,

Ventura, and Santa Barbara Co. We will also obtain samples from central California areas such as Fresno and Visalia in the Central Valley with the assistance of rootstock breeding program collaborators (samples will be mailed to the Manosalva lab). Funds requested include the cost of a cargo van rental from UCR fleet services at a monthly rate of \$552.10. *It is cheaper to rent by month than by day (\$55.21/day) if we need to do more than 10 trips*. For Ventura and more northern orchards, travel cost includes lodging with an average rate of \$180/night and meals at a per diem rate of \$79/day. In addition, we have budgeted money to cover fuel that will be used to travel to collect samples @ \$4.50/gallon and 20 miles/gallon. For Adaskaveg, travel costs will be based on traveling four times a year to two locations, one in Temecula and one in Ventura Co., for a total of 8 trips per year. Trips to Temecula will be day trips while trips to Ventura will be overnight using the hotel, meals, and fuel estimations as indicated above.

**Supplies (\$29,400)**. Funds are requested to cover greenhouse and laboratory supplies and consumables including UC Mix soil, pots, plant labels, chemicals to prepare solutions for fungicide treatments, fertilizers, tree sticks, ziploc bags for sample collection, media to prepare pathogen inoculum and for pathogen isolation, pipette tips, tubes, petri dishes, gloves, and PPE. In addition, we are budgeting money to cover molecular supplies and consumables to conduct *Pc* identification using ITS region Sanger Sequencing and *Pc*-Specific TaqMan qPCR assays. These supplies were estimated based on historical amounts and cost of similar research projects and activities in the Manosalva and Adaskaveg laboratories.

**Services and others (\$6,600).** Funds are also requested to cover UCR greenhouse fees at a rate of \$100 month for two benches each year of the project. This space will be used to conduct the greenhouse activities described in Obj. 1. We are budgeting funds to conduct Sanger Sequencing at the UCR sequencing core for pathogen identification in samples collected at different orchards in CA at a rate of \$10/sample. Diversity of pathogen isolates will be critical for assessment of resistance potential studies and will be shared between the two labs in addition to sourcing isolates from the *Phytophthora* collection at UCR.

# Leveraging and establishing rootstock/scion trials to determine the effect of rootstocks on improving scion's abiotic and biotic stress resilience

**Project Lead**: Dr. Patricia Manosalva, Microbiology and Plant Pathology Department, The Regents of The University of California, 245 University Office Building, Riverside, CA 92521, patricia.manosalva@ucr.edu, (951)827-3773.

**Project Cooperators**: Dr. Mary Lu Arpaia, Botany and Plant Sciences, The Regents of The University of California, mlarpaia@ucanr.edu; Dr. Ben Faber, Farm Advisor, Agricultural & Natural Resources University of California, bafaber@ucdavis.edu; and Grower Cooperators at field sites.

#### **EXECUTIVE SUMMARY**

Avocado (*Persea americana*, Mill), a major fruit tree crop produced worldwide, is highly susceptible to water stress. Avocado production and sustainability are under threat due to the increase of production challenges related to climate change (i.e., heat, salinity, drought, and flooding) which also exacerbates major avocado diseases such as Phytophthora Root Rot (PRR), caused by *Phytophthora cinnamomi* and Avocado Branch Canker (ABC), caused by several Botryosphaeriaceae species. These stressors usually occur together and can destroy avocado orchards if not managed properly. This is particularly true for California (CA) which produces ~90% of the USA avocado crop.

Climate change events will continue to increase PRR severity and incidence. PRR is one of the most devastating avocado diseases worldwide and affects 75% of CA growers causing annual losses of \$40 Millions of dollars. PRR severity and incidence are exacerbated under flooding and hypoxic conditions caused by inappropriate irrigation practices and soil waterlogging conditions, which are common conditions in CA. Growers heavily rely on moderate resistant rootstocks and phosphonate-based fungicides for management, however *P. cinnamomi* isolates overcoming these both practices, have been reported in CA<sup>1,2,3</sup>. The recent registration of Orondis® for avocados, as well as existing chemistries and new UCR resistant rootstocks to be released in 2025, hold promise for growers, however, the durability of these new control methods still deserves intensive evaluation since the great genome plasticity and adaptative capacity of *P. cinnamomi* populations to local environments and controls methods, will determine their effectiveness<sup>3</sup>.

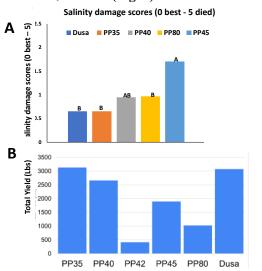
*Water stress including flooding, drought, and salinity will continue to increase due to climate change events and will exacerbate stress-related disease such as Avocado Branch Canker (ABC).* Salinity is an everincreasing problem due in part to climate change events (drought and heat). There have been numerous studies demonstrating that abiotic stressors cause significant yield losses in major crops due to reduction of growth and photosynthesis. Avocado, especially the dominant variety 'Hass', is extremely sensitive to salinity with yields declining when irrigation water has values of electrical conductivity (EC) above 0.75 dS/m and chloride (Cl-) concentrations over 100 ppm<sup>4</sup>. A decade of extreme heat and drought in CA has resulted in a significant increase in California's soil and water salinity. Soil salinity levels >1,000 ppm have been reported in Los Angeles, Ventura, and Santa Barbara Counties. The salinity of the Colorado River which provides irrigation water for Riverside and San Diego is also increasing (>700 ppm). Due to increasing scarcity and high cost of freshwater, growers are adopting efficient irrigation systems such as drip and microsprinklers, however, these strategies will take long time in order to supply enough water to satisfy leaching requirements to reduce salinity. Extreme heat waves and salinity have increased the severity and incidence of ABC, a stress-related disease. Currently, commercially available avocado varieties and rootstocks are susceptible to ABC<sup>5</sup>. Avocado branch canker and dieback represent a threat to avocado production, causing significant economic losses to the avocado industry.

In CA, each production area/orchard has their own microclimate, production challenges, and cultural practices; Thus, it is crucial to choose the right rootstock which will best support the chosen scion. High pH and alkalinity due to high levels of CaCO<sub>3</sub> in CA water and soil is becoming more prominent and growers use different practices to lower pH and alkalinity. The knowledge regarding the performance of 'Hass' and other scions to ABC and high pH & alkalinity as influence by rootstocks is largely unknown. Genetically diverse biotic and abiotic resistant rootstocks are needed to mitigate a wide range of production challenges in CA. Growers have acknowledged that genetic improvement and diversification combined with effective disease management approaches are key long-term solutions to mitigate these major avocado production challenges; thus, growers are eager to diversify their orchards with commercial and new UCR improved rootstocks (RS) and appropriate RS/Scion (SC) combinations to sustain their orchard productivity and industry competitiveness. The California

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Avocado Commission (CAC) is currently funding the field evaluation of five UCR advanced PRR resistant rootstocks (PP35, PP40, PP80, PP42, and PP45) in CA. These fields represent diverse environmental conditions and cultural practices: i) disease problems, ii) high salinity and chloride toxicity, iii) high pH and alkalinity (as CaCO<sub>3</sub>), iv) waterlogging conditions and clay soils, and v) different cultural practices. For this project, we selected 10 field trials in production stage (3- to 8-years old trees) in Northern and Southern CA where our UCR rootstocks are being tested (Table 1). Multiyear data collection at these 10 field trials shown that 'Hass' grafted on PP45, is more salinity susceptible compared with 'Hass' when is grafted to PP40, PP35, and PP80 rootstocks (Fig. 1). Data collected at these fields showed that PP35 and PP40 shown a consistent good performance and production across all field trials supporting their commercial released in FALL 2025/early 2026 (Table 2). In a recent greenhouse experiment, we demonstrated that ungrafted PP40 can tolerate irrigation water with EC = 3.2 dS/m, Cl- levels of 640 ppm, and Sodium (Na+) levels of 195 ppm by excluding Na+ and Cl- ions in the leaves when compared to the more susceptible rootstock, Thomas (Fig. 2).



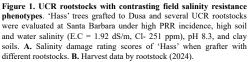


Table 1. Description of the rootstock field trials established in California to be used in this project. Evaluation is currently funded by the California Avocado Commission (CAC) & USDA-SCRI Grant.

| rrently funded by the California Avocado Commission (CAC) & USDA-SCRI Grant. |                                       |  |  |  |  |  |  |
|--|---------------------------------------|--|--|--|--|--|--|
| Grower   | County                                | Rootstock  | Field conditions   |  |  |  |  |
| .eo<br>AcGuire   | Temecula/<br>Riverside<br>(2 fields)  | Scion: Hass; Rootstock: Field 1 (2019):<br>PP35 (T), PP40 (T); Design: planted by<br>rows/rootstock<br>Field 2 (2021): Dusa (T), PP42 (MT), and<br>PP80(U); Design: planted by<br>blocks/rootstock   | EC= 0.82 dS/m, chloride= 102 mg/L,<br>pH = 7.9, CaCO <sub>3</sub> =130 mg/L, and high<br>PRR incidence (Orondis application).<br>Loamy sand to sandy loam.                           |  |  |  |  |
| Agua Tibia<br>LC, Ranch  | Pala/San<br>Diego<br>(2022)           | Scion: Hass, Gem, Lamb-Hass, and Reed;<br>Rootstock: Dusa (T), PP35 (T) PP40 (T),<br>Toro Canyon (T), Steddom (T), PP80 (U);<br>Design: planted by blocks of scions<br>where rootstocks are completely<br>randomized.  | EC =2.48 dSm, chloride =259.6 mg/L,<br>pH =8.7, CaCO <sub>3</sub> =110 mg/L, and PRR.<br>Sandy loam. No fungicides.  |  |  |  |  |
| ohn Lamb   | Camarillo/<br>Ventura<br>(2 fields)   | Scion: Hass; Rootstock: Field 1 (2019):<br>PP35 (T), PP40 (T); Design: planted by<br>rows/rootstock<br>Field 2 (2021): Dusa (T), PP42 (MT), and<br>PP80(U); Design: planted by<br>blocks/rootstock   | EC = 1.16 dS/m, chloride = 148 mg/L,<br>pH = 8.7, CaCO <sub>3</sub> = 160 mg/L, No PRR.<br>Loamy sand to silt loam. No<br>fungicides.  |  |  |  |  |
| ete Miller   | Goleta/<br>Santa<br>Barbara<br>(2020) | Scion: Hass; Rootstock: Dusa (T), PP35<br>(T), PP40 (T), PP45 (S), PP80 (U); Design:<br>planted by blocks/rootstocks in 5<br>sections of the orchard.  | EC = 3.65 dS/m, chloride = 251 mg/L,<br>CaCO <sub>3</sub> = 220 mg/L, and high PRR.<br>Loam to clay soil. No fungicides.   |  |  |  |  |
| ete Miller   | Goleta/<br>Santa<br>Barbara<br>(2022) | Scion: Hass, Gem, and Lamb-Hass.<br>Rootstock: Dusa (T), PP35 (T), PP40 (T),<br>PP80 (U); Design: planted by blocks of<br>scions where rootstocks are completely<br>randomized.  | EC =1.92 dS/m, chloride = 236 mg/L,<br>pH = 8.3, CaCO <sub>3</sub> = 220 mg/L, and PRR<br>incidence. Loam. No fungicides.  |  |  |  |  |
| Chris Sayer,<br>Petty<br>Ranch)  | Ventura<br>(2020)                     | Scion: Hass; Rootstock: Dusa (T), PP35<br>(T), PP40 (T), PP45 (S); Design: planted by<br>blocks/rootstock  | EC =2.3 dS/m, chloride = 92 mg/L, pH<br>= 7.4, CaCO <sub>3</sub> = 320 mg/L (severe). No<br>PRR incidence. Loamy sand to silt<br>Ioam. High limestone. No fungicides                 |  |  |  |  |
| Adna<br>Farms  | Temecula/<br>Riverside<br>(2020)      | Scion: Hass; Rootstock: Dusa (T), PP35<br>(T), PP40 (T), PP45 (S); Design: planted by<br>blocks/rootstock  | EC= 0.82 to 1.1 dS/m, chloride= 102<br>mg/L, pH = 7.9, CaCO <sub>3</sub> = 130 mg/L, and<br>high PRR incidence (Orondis). High<br>clay composition. Treated with<br>Orondis.         |  |  |  |  |
| Pine Tree<br>Ranch<br>Salinity Tole  | Ventura<br>(2017)<br>erant. MT = Sal  | Scion: Hass; 30 Rootstocks: Dusa, Leola,<br>Zerala, Steddom, Toro Canyon, Uzi,<br>Zentmyer, Topara, PP35, PP40, PP45,<br>PP42, PP80, 11 new UCR selections, 4<br>selections from SA, and two Israeli<br>rootstocks; Design: planted in a complete<br>randomized block design.<br>inity Moderate Tolerant, S = Salinity Suscept | EC= 0.74 to 1.1 dS/m, chloride= 35<br>mg/L, pH = 7.56, CaC0 <sub>3</sub> =130 mg/L.<br>No PRR. 2.7% of limestone. Alkalinity<br>problems. Loamy sand to Loam soil.<br>No fungicides. |  |  |  |  |

T = Salinity Tolerant, MT = Salinity Moderate Tolerant, S = Salinity Susceptible, U = unknown, PRR = Phytophthora root rot. SA = South Africa.

| Tier 3<br>Advanced<br>Rootstock  | Characteristics   |
|----------------------------------|---|
| UCR-R01 (PP35)<br>UCR-R02 (PP40) | <ul> <li>Resistant to different <i>Phytophthora <u>cinnamomi</u></i> clonal populations.</li> <li>Salinity tolerance (E.C values 0.75 - 3.7 <u>dS</u>/m, Chloride 100 - 350 ppm).</li> <li>UCR-R02 exhibited tolerance at 600 ppm chloride levels (under GH conditions).</li> <li>Good productivity: pH 7 - 9; from Sandy loam to clay soils, high alkalinity as CaCO<sub>3</sub> &gt; 200 ppm.</li> <li>Medium (R02) and small (R01) vigorous productive trees.</li> <li>Moderate heat tolerance.</li> <li>Compatible with 'Hass', GEM, Lamb, Reed, and Carmen.</li> </ul> |
| UCR-R04 (PP42)<br>UCR-R05 (PP45) | <ul> <li>Highly resistant to different <i>Phytophthora <u>cinnamomi</u></i> clonal populations. Good for replanting problems due to PRR.</li> <li>Medium vigorous productive trees.</li> <li>Salinity susceptible</li> <li>Good productivity: pH 7 - 9; from Sandy loam to clay soils, high alkalinity as CaCO<sub>3</sub> &gt; 200 ppm.</li> <li>Compatible with 'Hass', GEM, Lamb, Reed, and Carmen.</li> </ul>   |
| UCR-R03 (PP80)                   | <ul> <li>Resistant to different <i>Phytophthora <u>cinnamomi</u></i> clonal populations.</li> <li>Heat tolerance</li> <li>Moderate tolerant to salinity (E.C values 0.75 - 3.7 dS/m, Chloride 100 - 350 ppm).</li> <li>Needs more field data evaluation to support its commercial release.</li> </ul>   |

 Table 2. UCR advanced rootstock characteristics based on greenhouse and field data

 Tier 3
 Characteristics

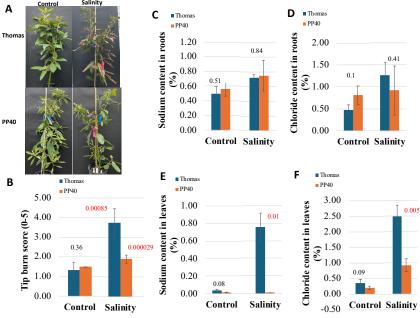


Figure 2. PP40 can tolerate high salinity by preventing Na+ and Cl- ion accumulation in leaves. Ungrafted Thomas and PP40 rootstocks were treated by a control solution plus fertilizer (E.C = 1.3 dS/m, Cl- 34 ppm, Na+ 39 ppm) and high salinity solution (E.C = 3.2 dS/m, Cl- 640 ppm, Na+ 195 ppm) for six months. A. Pictures of plants at the end of experiment. B. Tip burn phenotype using score 0-5. C-D. Sodium and Chloride content in roots at the end of the experiment. C-D. Sodium and Chloride content in leaves at the end of the experiment. Error bars indicate standard deviation using 5 plants per treatment. T-test was conducted and P values are indicated.

Field data regarding the performance of these five UCR PRR resistant rootstocks grafted with scions other than 'Hass' under major production challenges in CA is limited. We will leverage these established field trials to assess rootstock productivity and identify the best rootstock/scion combinations that growers can use to mitigate major production challenges and sustain productivity and competitivenes. By integrating phenotypic, physiological, ionomic, and production data from several rootstock grafted to 'Hass' and other scions at these field trials, we will gain insights regarding the salinity tolerance mechanisms of these scions as influenced by several rootstocks under field conditions. Furthermore, we will assess the salinity responses of ungrafted and 'Hass' grafted commercial rootstocks to gain knowledge regarding key salinity traits and mechanism(s) explaining their resistance/vulnerabilities to high saline conditions. Our overall goal is to ensure the long-term sustainability and competitiveness of the CA avocado industry by reducing production inputs and yield losses due to major production challenges by identifying the best rootstocks & rootstock/scion combination(s) to mitigate salinity, alkalinity, PRR, and ABC conditions. Our results will also aid the development of new superior salinity resistant cultivars by identifying key salinity traits resistance mechanisms.

**PROJECT OBJECTIVES.** To accomplish this goal, we are proposing the following objectives: **Objective 1.** Determine the best rootstocks or rootstock/scion combinations to mitigate major production challenges by integrating phenotypic, physiological, ionomic, and production data from rootstocks evaluated under several abiotic and biotic stressors at trials established across major CA growing regions (**Table 1**).

#### Deliverables

- Best rootstock and rootstock/scion combinations x environment (tree performance and productivity) to mitigate production challenges in CA and recommendations to growers of which combination to use based on their orchard conditions.
- Salinity (phenotypic, physiological, and ionomic datasets) and pathogen responses (phenotypic, & physiological datasets) of 'Hass' and other scions as influence by several rootstocks.
- Key salinity traits and mechanisms explaining the observed tolerance/susceptibility of the rootstock/scion combinations under field conditions.
- Commercial released of the UCR rootstocks (PP35 & PP40 [2025-2026], PP45 & PP42 [2026-2027], and PP80 [2028-2029]) with a comprehensive protocol for the conditions where these should be planted and field data available from CA.

**Objective 2.** Assess the salinity, PRR, & ABC responses of commercial rootstocks and gain insights regarding the mechanism(s) of their salinity responses observed under greenhouse conditions. *Deliverables* 

- Phenotypic, physiological, and ionomic salinity responses of commercial ungrafted rootstocks.
- Phenotypic, physiological, and ionomic salinity responses of 'Hass' as influenced by commercial rootstocks.
- Key salinity traits & mechanisms explaining the salinity responses of commercial rootstocks (i.e., leaf necrosis, Na+/CL- root/leaf exclusion) and how they influence 'Hass' responses.
- An integrative approach for screening rootstocks for salinity resistance without the obvious bias involved in using empirical/subjective screening approaches such as visual scoring of leaf tip burn, leaf discoloration, and poor root growth.
- Phenotypic and physiological PRR and ABC responses of commercial rootstocks when they are ungrafted or grafted to 'Hass'.
- Rank commercial rootstocks by their salinity, PRR, and ABC resistance responses from the best to the worse performers and provide recommendations to growers of what rootstocks to plant in their orchards depending on the disease incidence and severity of these production challenges at their orchards.

## WORK PLAN AND METHODS

**Objective 1.** We will continue the evaluation of all rootstock/scion combinations planted at the 10 fields described in **Table 1**. We will collect a wide range of datasets to identify interactions of rootstock/scion genotypes x environment, abiotic, and biotic stressors. We will also gain insights regarding the mechanisms of salinity tolerance/susceptibility observed. We will collect:

<u>Tree phenotypic (November 2025-October 2028).</u> We will conduct tree measurements (height and width) to calculate canopy volume. Field tree performance data regarding overall tree health, salinity and heat damage scores, flushing, blooming, and fruit set scores will be recorded. Disease incidence and severity scores will be also collected for ABC and *Persea mites* when present in the field. Once per year, we will calculate PRR incidence. We will use our well-established scoring system developed by the UCR rootstock breeding program. Data will be collected twice a year (Spring and Fall).

**Production data (April 2026-July 2028).** Yield data will be collected depending on the grower collaborators timeline which is based on: i) price market, ii) crew availability, and iii) variety seasonality. 'Hass' and 'GEM' fruits will be harvested from January-May and 'Lamb Hass' and 'Reed' harvest will occur from July to November. At Pine Tree and Agua Tibia trials, we will collect yield data: total fruits, total weight, weight/fruit, and yield efficiency/tree and per rootstock. In addition to the data described above, we will get packing size data per rootstock at all the fields with the exception of Pine Tree and Agua Tibia trials.

*Physiological parameters (November 2025-October 2028).* Leaf chlorophyll content will be measured using a portable Chlorophyll Meter. One of the toxic effects of salt exposure is oxidative stress-induced chlorophyll degradation and leaf necrosis. Loss of chlorophyll serves as an early indicator of salinity damage. Leaf stomatal conductance will be measured using a leaf porometer. During salinity exposure, high leaf concentrations of Sodium (Na+) are associated with reduced leaf stomatal conductance which together with chlorophyll degradation reduces photosynthetic carbon assimilation, and ultimately leads to yield reduction. We will collect these parameters twice/year with phenotypic data in every tree. Stem Water Potential (SWP) will be measured using a pressure chamber unit (Model 615 manufactured by PMS Instruments) as indicator and predictor of salinity, PRR, and ABC responses. During water stress triggered by abiotic (i.e., salinity, drought, heat) and biotic (i.e., PRR & ABC) stressors, the tree availability/abortion of water decreases and avocado experience reduction in SWP. Since SWP is time consuming, we will measure SWP in 30 trees corresponding to the best and worst combinations at each field trial (n=600 trees). We will measure SWP in two mature leaves/tree following the protocols used by the rootstock breeding program during our PRR and salinity screenings. We will measure SWP once a year (August/September).

*Water and soil comprehensive analyses (August every year).* We will collect soil and water samples from all fields for irrigation water and comprehensive soil analyses which will be done by Fruit Grower Labs (FGL, Santa Paula). These analyses include primary (i.e., nitrogen [N], phosphorous [P], and potassium [K+]), secondary (Ca2+, Mg2+, Na2+), toxic (Boron [B] and Cl-), and micronutrients (i.e., Mn2+) as well as other soil properties including EC, pH, and soil texture. Irrigation water analyses provide similar information. These

analyses are important since soil and water compositions are subjective to change by weather, irrigation water quality & regiment, type of soils, and oxygen availability. It is critical conduct these analyses to correlate field salinity conditions that can explain the salinity phenotypes, yield, tree performance, tree growth, physiological parameters, and ionomic profiles.

**Lonomic profiles (November 2025-October 2028).** A comprehensive leaf and root ionomic profiles will be conducted at each field in August/September tentatively in 2027 based on our records. *Note that we will be also guided by our phenotypic data and we will select the time when large variability and severity on salinity, heat, and disease phenotypes will be observed.* These analyses will be conducted in year 2. Leaf and roots will be collected from 6-10 trees (selected based on similar phenotypic and physiological ratings) per combination and pooled. A total of three replicates (each 6-10 trees) will be submitted to FGL to quantify below- and above-ground concentrations of nitrogen (N), phosphorous (P), potassium (K), calcium (Ca), magnesium (Mg), zinc (Zn), manganese (Mn), iron (Fe), cooper (Cu), boron (B), sodium (Na+), chloride (Cl-), and sulfur (S) to identify potential mechanisms of transport, exclusion, and/or sequestration of Na+ and Cl-. Macro- and micro- nutrients quantifications will allow us to understand the impact of salinity and nitrogen levels on plant physiology and nutrient levels particularly on nitrogen uptake/utilization and carbon allocation which will have effects on CO<sup>2</sup> assimilation, photosynthesis, and ultimate in growth and yield. This field ionomic data will be correlated with the phenotypic, physiological, and yield data to elucidate the mechanism(s) explaining these salinity/pathogen responses under different field conditions and cultural practices.

**Data analyses (November 2025-October 2028).** Data analysis, correlation, and integration will be conducted to identify: i) the best salinity and pathogen tolerant rootstock/scion combinations and ii) salinity traits and mechanisms conferring salinity tolerance of high-performing rootstock/scion combinations under field conditions. Datasets will be assessed for normality using the Shapiro-Wilk test and examined with Generalized Lineal Mixed Models (GLMMs). Least squares mean will be assessed with a Tukey-Kramer HSD adjustment. GLMM and mean separation analyses will be performed in R v.4.3.2 (R Core Team 2023). Results will be considered significant at  $P \le 0.05$ . Unpaired two tailed Student's t-test will be used to compare two samples. *Caveats and pitfalls.* We do not foresee major problems with the collection of phenotypic and yield data since we have been collected these datasets since the field trials were established. We have been collected the physiological and ionomic data described above under greenhouse conditions during our rootstock salinity resistance screenings; thus, we do not foresee major complications in recording these data under field conditions. The timing of SWP data collection & ionomic profiles might vary depending on environmental conditions every year so we will adjust our collection times based on our phenotypic & physiological datasets and water & soil analyses. Moreover, we will conduct only Na+ and Cl- root and leaf content of the best and worst combination observed in the fields (Year 1) guided by our phenotypic & Physiological data as proof of concept.

**Objective 2.** To assess and confirm the salinity and pathogen responses of commercial rootstocks, a comprehensive salinity and pathogen greenhouse (GH) experiments will be conducted. Phenotypic and physiological responses in a nondestructive way as well as ionomic responses in a destructive way at the end of the experiments will be conducted. Approximately, one-year-old clonal experimental trees ungrafted and 'Hass' grafted corresponding to Steddom (Mexican [M] x Guatemalan [G]), Tami (VC801, West Indian [W] x M), Miriam (VC218, W x M), Leola<sup>TM</sup>, Zerala<sup>TM</sup>, and Thomas (salinity and PRR susceptible control) will be purchased from Brokaw Nursery LLC. (Santa Paula, CA). Trees will be transplanted into pots, arranged in a completely randomized design. Trees will be subjected to four treatments namely: i) uninoculated and no saline control (EC = 0.8 dS/m, Na+ 39 ppm, Cl- 34 ppm), ii) salinity (EC = 3.0 dS/m, Na+ = 100 ppm, Cl- = 400 ppm) for 5 to 6 months, iii) PRR inoculated and watered with no saline control solution, and iv) ABC inoculated and watered with no saline conditions. The salinity level we will use is close to those found in CA avocado orchards especially in Southern CA. Water analyses will be conducted in these solutions to ensure the desire levels of salinity and ion concentrations. A total of 6 replicates will be done per treatment/rootstock accessions. PRR and ABC inoculations will be conducted as described in Belisle et al. (2019)<sup>1</sup> and Avenot et al. (2023)<sup>5</sup>. A suite of phenotypic (tree measurements, tree health, trunk diameter, salinity and disease scores) and physiological datasets (leaf chlorophyll content, leaf conductance, and SWP) will be conducted before and during treatments (every two weeks and weekly after salinity damage phenotypes are showing). At the end of the experiment, root and leaf samples corresponding to two trees per treatment will be collected and pooled for Na+ and Cl- content analyses in triplicate (each 2 trees/treatment). The time point for the ionomic analyses will be based and adjusted based on our preliminary results conducting similar studies with two opposite salinity

resistant accessions (Fig. 2). We will record fresh weight of shoots and roots for each plant to correlate their salinity responses with plant growth. This greenhouse experiment will be conducted twice (Years 2, 3) for publication purposes. Data analyses will be done as described above (Obj. 1). *Caveats and pitfalls*. We do not foresee major difficulties in the methods and approaches described above since all protocols described are published and have been successfully used by at the UCR avocado rootstock breeding program and the Manosalva laboratory. One possible problem is that the conditions of salinity used in the GH experiments are over the threshold that these accessions tested can withstand; thus, we will add another level of salinity based on the average of salinity and chloride levels in our rootstock trials in CA in the second experiment. Since this is the first time that these commercial rootstocks are being tested together for salinity responses, the first experiment will serve as 'proof of concept' to assess the best time points for ionomic and SWP analyses. We do not oversee any difficulty regarding testing the accessions for PRR resistance using the current pathogen population in the USA. For ABC resistance screening, since ABC is a stress related disease, it is possible that our whole plant inoculations does not work or does not progress under current GH conditions (no stress). If this is the case, we will impose salinity and drought treatments in ABC inoculated trees to trigger/accelerate the disease progression.

**PROJECT OUTREACH.** We will ensure that our project results, outcomes, and recommendations are delivered and translated into actionable recommendations for growers and other avocado stakeholders with a robust and multi-faceted extension and outreach plan. Project members are UCANR extension faculty (Arpaia and Faber) and will ensure the dissemination of our outcomes and recommendations to growers and other stakeholders. By being active collaborators, growers will test our rootstocks grafted with 'Hass' and other scions at their orchards. Outcomes will be also outreached to stakeholders through presentations at CAC, CAS, Avocado Growers of CA (AGC), and UCANR meetings, workshops (i.e., Avocado Irrigation Workshop, UCANR), and field days (i.e., Pine Tree and Agua Tibia). Stakeholders from these groups include conventional and organic growers. Our team will also participate in Avocado Café, a virtual presentation and discussion forum that regularly brings together over 100 growers and stakeholders to discuss the scientific basis of solutions to emerging industry challenges. We will also report our progress and outcomes in grower journals such as *From the Grove*, the CAC's quarterly magazine), newsletters, and social media.

|      |      | I UNE TABLE  |      |        |        |        |      |         |        |        |           |        |        |         |
|------|------|--|------|--------|--------|--------|------|---------|--------|--------|-----------|--------|--------|---------|
| Obj. |      | Objective/Sub-task Description                                     | Year | 1 (Nov | 25 - 0 | ct 26) | Year | 2 (Nov  | 26 - 0 | ct 27) | Year      | 3 (Nov | 27 - 0 | Oct 28) |
| 1    |      | Determine the best rootstock/scion combination in the field        |      |        |        |        |      |         |        |        |           |        |        |         |
|      | 1.1  | Collect phenotypic data and physiological data (no SWP)            |      |        |        |        |      |         |        |        |           |        |        |         |
|      | 1.2  | Collect yield data   |      |        |        |        |      |         |        |        |           |        |        |         |
|      | 1.3  | Collect SWP data   |      |        |        |        |      |         |        |        |           |        |        |         |
|      | 1.4  | Collect water and soil samples for analyses                        |      |        |        |        |      |         |        |        |           |        |        |         |
|      | 1.5  | Collect root samples for PRR assessment                            |      |        |        |        |      |         |        |        |           |        |        |         |
|      | 1.6  | Collect samples for ABC pathogen isolation                         |      |        |        |        |      |         |        |        |           |        |        |         |
|      | 1.7  | Collect root and leaf samples for Ionomic analyses                 |      |        |        |        |      |         |        |        |           |        |        |         |
|      | 1.8  | Data analyses and Integration                                      |      |        |        |        |      |         |        |        |           |        |        |         |
|      | 1.9  | Commercial release for PP35 & PP40                                 |      |        |        |        |      |         |        |        |           |        |        |         |
|      | 1.10 | Collect fruit characteristics for PP45, PP80, & PP42               |      |        |        |        |      |         |        |        |           |        |        |         |
|      | 1.11 | Submit PVP forms and commercial release of PP42 & PP45             |      |        |        |        |      |         |        |        |           |        |        |         |
|      | 1.12 | Project outreach and publications                                  |      |        |        |        |      |         |        |        |           |        |        |         |
|      |      | ESTIMATE BUDGET FOR THIS MILESTONES ACTIVITIES                     |      | \$67,9 | 28.30  |        |      | \$112,3 | 64.30  |        | 71,241.30 |        |        |         |
| 2    |      | Screen commercial rootstocks for salinity. PRR, and ABC resistance |      |        |        |        |      |         |        |        |           |        |        |         |
|      | 2.1  | Order/Purchase trees from Brokaw Nursery LLC                       |      |        |        |        |      |         |        |        |           |        |        |         |
|      | 2.2  | Initiate treatments in the greenhouse                              |      |        |        |        |      |         |        |        |           |        |        |         |
|      | 2.3  | Collect phenotypic data and physiological data (no SWP)            |      |        |        |        |      |         |        |        |           |        |        |         |
|      | 2.4  | Collect SWP data   |      |        |        |        |      |         |        |        |           |        |        |         |
|      | 2.5  | Collect Ionomic data   |      |        |        |        |      |         |        |        |           |        |        |         |
|      | 2.6  | Assess PRR resisistance  |      |        |        |        |      |         |        |        |           |        |        |         |
|      | 2.7  | Assess ABC resistance  |      |        |        |        |      |         |        |        |           |        |        |         |
|      | 2.8  | Data analyses and Integration                                      |      |        |        |        |      |         |        |        |           |        |        |         |
|      | 2.9  | Project outreach and publications                                  |      |        |        |        |      |         |        |        |           |        |        |         |
|      |      | ESTIMATE BUDGET FOR THIS MILESTONES ACTIVITIES                     |      | \$9,0  | 36.70  |        |      | \$24,0  | 66.00  |        |           | 14,7   | 05.70  |         |

# MILESTONE TABLE

## REFERENCES

<sup>1</sup> Belisle R. et al. (2019a). *Phytopathology* 109(3):384-394.

<sup>2</sup>Belisle R. et al. (2019b). *Plant Disease* 103(8):2024-2032.

<sup>3</sup>Shands AC. et al (2024). *Front Microbiol*. 15:1341803.

<sup>4</sup>Acosta-Rangel, A. et al. (2019). *Scientia Horticulturae* Vol. 256:108629.

<sup>5</sup>Avenot et al. (2023). *Phytopathology* 113: 1034-1047.

# PROJECT BUDGET

| Personal Salar (Sec. 1997) Sec. 1997 (Sec. 1   | PROJECT BUDGET   | 1                     |                       |                       |
|--|--|-----------------------|-----------------------|-----------------------|
| Assuant Speciality II (i) 50% EFT (K) Craduals Solution Researcher (CSR), Summer Quarters (K) Craduals Solution Researcher (CSR), Summer Quarters (K) Craduals Solution Researcher (CSR), Summer Quarters (K) Constraints Speciality II (ii) 50% EFT (K) Craduals Solution Researcher (CSR), Summer Quarters (K) L Data Constraints Constr   | Table 3. Manosalva et al. budget description   | 11/01/2025-10/31/2026 | 11/01/2026-10/31/2027 | 11/01/2027-10/31/2028 |
| Ph. D Craduals Subdem Reservelver (GSR), Summer Quarters         \$5,000         \$9,167           Statistati Vocenskut RG (SVR), Summer Quarters         \$14,043         \$14,455         \$14,899           Assistati Vocenskut Reservelve (GSR), Summer Quarters         \$187         \$193         \$193           Obj. I. Data collection: phenotypic, innomic, physiological, & yield         \$187         \$193         \$170           Salt comprohensive analyses by FGL         \$570         \$570         \$570         \$570           Kata Comprohensive analyses by FGL         \$590         \$590         \$590         \$570           Kata Comprohensive analyses by FGL         \$590         \$570         \$570         \$570           Kata Comprohensive analyses by FGL         \$590         \$520         \$570         \$570           Kata Cont Favel (Manosalva)         Kata Cont Favel (Nanosalva)         \$100         \$11,04         \$11,04           Kata Cont Contract to 10 policy rate Kitking and Bayember)         \$510         \$11,04         \$11,04         \$11,04           Gald and Waye Wig (Rot one month wicely ear-\$52,1 x 2)         \$11,04         \$11,04         \$11,04         \$11,04           Yays to avel State Collect data (Workherther Trials         \$12,05         \$52,2         \$52,2         \$52,2         \$52,2         \$52,2 <t< td=""><td>Personnel Salary</td><td></td><td></td><td></td></t<>  | Personnel Salary   |                       |                       |                       |
| Personal Benefis Product States Use Control 16 30% EFT S14,405 S14,40 S14,405 S14,40 S14,4   | Assistant Specialist II @ 50% EFT  | \$32,136              | \$33,100              | \$34,093              |
| stakam tyseinht 10, 30% EFT<br>h. Grahum Submit Reserverk (SB), Summer Quarters<br>S187 S187 S193<br>S187 S193<br>S193<br>S193<br>S193<br>S193<br>S193<br>S193<br>S193<br>S193<br>S193<br>S193<br>S193<br>S193<br>S193<br>S193<br>S193<br>S193<br>S193<br>S193<br>S193<br>S193<br>S193<br>S193<br>S193<br>S193<br>S193<br>S193<br>S193<br>S193<br>S193<br>S193<br>S193<br>S193<br>S193<br>S193<br>S193<br>S193<br>S193<br>S193<br>S193<br>S193<br>S193<br>S193<br>S193<br>S193<br>S193<br>S193<br>S193<br>S193<br>S193<br>S193<br>S193<br>S193<br>S193<br>S193<br>S193<br>S193<br>S193<br>S193<br>S193<br>S193<br>S193<br>S193<br>S193<br>S193<br>S193<br>S193<br>S193<br>S193<br>S193<br>S193<br>S193<br>S193<br>S193<br>S193<br>S193<br>S193<br>S193<br>S193<br>S193<br>S193<br>S193<br>S193<br>S193<br>S193<br>S193<br>S193<br>S193<br>S193<br>S193<br>S193<br>S193<br>S193<br>S193<br>S193<br>S193<br>S193<br>S193<br>S193<br>S193<br>S193<br>S193<br>S193<br>S193<br>S193<br>S193<br>S193<br>S193<br>S193<br>S193<br>S193<br>S193<br>S193<br>S193<br>S193<br>S193<br>S193<br>S193<br>S193<br>S193<br>S193<br>S193<br>S193<br>S193<br>S193<br>S193<br>S193<br>S193<br>S193<br>S193<br>S193<br>S193<br>S193<br>S193<br>S193<br>S193<br>S193<br>S193<br>S193<br>S193<br>S193<br>S193<br>S193<br>S193<br>S193<br>S193<br>S193<br>S193<br>S193<br>S193<br>S193<br>S193<br>S193<br>S193<br>S193<br>S193<br>S193<br>S193<br>S193<br>S193<br>S193<br>S193<br>S193<br>S193<br>S193<br>S193<br>S193<br>S193<br>S193<br>S193<br>S193<br>S193<br>S193<br>S193<br>S193<br>S193<br>S193<br>S193<br>S193<br>S193<br>S193<br>S193<br>S193<br>S193<br>S193<br>S193<br>S193<br>S193<br>S193<br>S193<br>S193<br>S193<br>S193<br>S193<br>S193<br>S193<br>S193<br>S193<br>S193<br>S193<br>S193<br>S193<br>S193<br>S193<br>S193<br>S193<br>S193<br>S193<br>S193<br>S193<br>S193<br>S193<br>S193<br>S193<br>S193<br>S193<br>S193<br>S193<br>S193<br>S193<br>S193<br>S193<br>S193<br>S193<br>S193<br>S193<br>S193<br>S193<br>S193<br>S193<br>S193<br>S193<br>S193<br>S193<br>S193<br>S193<br>S193<br>S193<br>S193<br>S193<br>S193<br>S193<br>S193<br>S193<br>S193<br>S193<br>S193<br>S193<br>S193<br>S193<br>S193<br>S193<br>S193<br>S193<br>S193<br>S193<br>S193<br>S193<br>S193<br>S193<br>S193<br>S193<br>S193<br>S193<br>S193<br>S193<br>S193<br>S193<br>S193<br>S193<br>S193<br>S193<br>S193<br>S193<br>S193<br>S193<br>S193<br>S193<br>S193<br>S193<br>S193<br>S193<br>S193<br>S193<br>S193<br>S193<br>S193<br>S193<br>S193<br>S193<br>S193<br>S193<br>S193<br>S | Ph. D Graduate Student Researcher (GSR), Summer Quarters                             | \$8,900               | \$9,167               |                       |
| Ph. D Cruding: Sublem Researcher (GR): Summe Charter     \$187     \$193       Obj. I. Data collections: phenotypic, ionomic, physiological, & yield   | Personnel Benefits   |                       |                       |                       |
| Obj. 1. Data collection: phenotypic, ionomic, physiological, & yield           Soli comprehensive analyses for 10 field traik (\$572mmple) by FGL         \$570         \$570         \$570           Soli comprehensive analyses for 10 field traik (\$552mmple)         \$520         \$950         \$950           Leaf and more homoic analyses (FGL) \$553mmple         \$520         \$950         \$950           Travel ( Manosalva)         \$42,750         \$570         \$570           Rental Car to travel to 10 plats rate twice as year and harvest one/year         \$510         \$510         \$510           VCR field Rental, & Southern Traisbime         \$51,104         \$1,104         \$1,104           7 days to collect data (@ Northern Traisbime         \$51,104         \$1,104         \$1,104           7 days to collect data (@ Northern Traisbime         \$52,152         \$552         \$552           10 abayset (@ Ret on moth once/year-\$52,1)         \$51,52         \$552         \$552           10 abayset (@ Ret on moth once/year-\$52,1)         \$51,52         \$552         \$552           10 abayset (@ Ret on moth once/year-\$52,1)         \$52,160         \$2,160         \$2,160           2 days to collect data (@ Northern Traisbime         \$52,160         \$2,160         \$2,160           2 days to collect data (@ Northern'Traisbime         \$52,00         \$52  | Assistant Specialist II @, 50% EFT   |                       |                       | \$14,899              |
| Analyses by EGL         STO           Solic compredience analyses for 10 field triak (\$57/anample) by FGL         \$570         \$570           By Compredience analyses for 10 field triak (\$57/anample)         \$570         \$570           By Compredience analyses for 10 field triak (\$57/anample)         \$570         \$570           By Compredience analyses (FGL 12 Statum)         \$521         \$520         \$520           By Compredience analyses (FGL 12 Statum)         \$521/day and house compredience and by Compredience and Compred  | Ph. D Graduate Student Researcher (GSR), Summer Quarters                             | \$187                 | \$193                 |                       |
| Solid comprehensive analyses for 10 field train (S73/cample) bp FGL         \$570         \$570         \$570         \$570           Frequent waters studibly FGL servery (S75/cample)         \$950         \$950         \$950           Leaf and roos lonomic analyses (FGL) \$75/cample         \$950         \$950         \$950           Rental Car to travel to 10 plots rate twice ay sen and harvest once/year         \$100  | Obj. 1. Data collection: phenotypic, ionomic, physiological, & yield                 |                       |                       |                       |
| Irregular on water stability FCI, yardy. (2855ample)<br>Eq. and road stability FCI. Systemple<br>(294 of road information analysis (FCI. Systemple<br>(295 of 196  | Analyses by FGL  |                       |                       |                       |
| Leaf and rook Incomic analyses (FGL) \$755ample     \$42,750       Travel (Manosalva)     \$42,750       Rental Car to travel to 10 polis rate twice a year and harvest once/year     \$42,750       UCR Red Rental Cargo Vang 552 Libry and long rental 552. Imouth.     \$52,000       Field data collection 2x/year (Bring and September)     \$1,104       Tays to collect data (B) contem Trails/imas     \$1,004       Tays to collect data (B) contem Trails     \$2,100       Tays to collect data (B) contem Trails     \$1,106       Tays to collect data (B) contem Trails     \$1,106       Travel (Arpaia)     \$2,100     \$2,160       Tays to collect data (B) contem T   | Soil comprenhensive analyses for 10 field trials (\$57/sample) by FGL                | \$570                 | \$570                 | \$570                 |
| Travel (Manosalva)       Kental Car to travel to 10 plots rate twice a year and harvest once/year       VCR feet Rental Cargo Van (i) 55:21/day and long renal 55:21/month.         Field and collection Zx/our (i) 55:21/day and long renal 55:21/month.       Statistical Cargo Van (ii) 55:21/day and long renal 55:21/month.         Field and collection ance per year       Statistical Cargo Van (ii) 55:21/day and long renal 55:21/month.       Statistical Cargo Van (ii) 55:21/month.         Field and collection ance per year       Statistical Cargo Van (ii) 55:22/monther)       Statistical Cargo Van (ii) 55:22/monther)       Statistical Cargo Van (iii) 55:22/monther)         Field and collection ance per year       Statistical Cargo Van (iii) 54:320       Statistical Cargo Van (iii) 54:320       Statistical Cargo Van (iii) 54:320         Field and collection from people/2x per year (iii) 12 (aby)year (kiii) 12 (aby)year (iiii) 12 (aby) 12 (ab) 12  | Irrigation water suitability FGL yearly (\$95/sample)                                | \$950                 | \$950                 | \$950                 |
| Rental Car to travel to 0 polor rate twice a year and harvest once/year       Image: Constraint of the constraint of   | Leaf and roots Ionomic analyses (FGL) \$75/sample                                    |                       | \$42,750              |                       |
| Rental Car to travel to 0 polor rate twice a year and harvest once/year       Image: Constraint of the constraint of   | Travel (Manosalva)   |                       |                       |                       |
| UCR Bear Benul: Cargo Van @ 55.21/day and long renul 552.1/month.<br>Field data collect data @ Southem Trails/time<br>5 days to collect data @ Northem Trails/time<br>7 days to collect data @ Northem Trails/time<br>7 days to southen once per year (Rent one month twice/year=552.1 x.2)<br>Yeld data collect on none per year<br>8 days to harvest Southem Trails/<br>Trails 2 days/query (Rent one month once/year=552.1)<br>Trails 2 days/query (Rent one per gel/year (RENthight/person)<br>Trails 2 days/query (Rent one per gel/year (RENthight/person)<br>Trails 2 days days (Rent one days days average<br>S 2.160<br>S 2.160  |  |                       |                       |                       |
| Field data collection Zeyear (Spring and September)<br>3 days to collect data (@ Nothem Trails/time<br>7 days to sources to southem Trails<br>8 days to havest Nothem Trails (% 1,104<br>12 days/to collect data (@ Nothem Trails (% 1,104<br>12 days to collect data (@ Nothem Trails (% 1,104<br>12 days to collect data (@ Nothem Trails (% 1,104<br>13 days to collect data (@ Nothem Trails (% 1,104<br>14 days for flat data collection to popole/2x per year (@ 7,9/day/person)<br>15 days to collect data (@ Nothem Trails (% 1,104<br>14 days for flat data collection to popole/2x per year (@ 7,9/day/person)<br>15 days to collect data (@ Nothem Trails (% 1,106<br>14 days to collect data (@ Nothem Trails (% 1,106<br>15 days to collect data (@ Nothem Trails (% 1,204<br>14 days to collect data (@ Nothem Trails (% 1,204<br>15 days to collect data (@ Nothem Trails (% 1,204<br>15 days to collect data (@ Nothem Trails (% 1,204<br>15 days to collect data (@ Nothem Trails (% 1,204<br>16 days to collect data (@ Nothem Trails (% 1,204<br>17 days to collect data (@ Nothem Trails (% 1,204<br>18 days to collect data (@ Nothem Trails (% 1,204<br>19 days to collect data (@ Nothem Trails (% 1,204<br>19 days to collect data (@ Nothem Trails (% 1,204<br>19 days to collect data (@ Nothem Trails (% 1,204<br>19 days to collect data (@ Nothem Trails (% 1,204<br>19 days to collect data (@ Nothem Trails (% 1,204<br>19 days to collect data (@ Nothem Trails (% 1,204   |  |                       |                       |                       |
| 5 days to collect data @ Southen Trailstime<br>Total 12 days (brivelyear (Rent one month twicelyear=552.1 x 2)<br>Field data collection more per year<br>5 days to harvest Southen Trials<br>5 days to harvest Southen Trials<br>1 days to sole data (Rent one month once/year=552.1)<br>1 days to collect data (Rent one month once/year=552.1)<br>1 days to collect data (@ northen Trials<br>1 days to collect data (@ northen trials   |  |                       |                       |                       |
| 7 days to collect data @ Northem Trials/ine month twice/year=552.1 x 2) \$1,104 \$1,106  |  |                       |                       |                       |
| Total 12 days @ wice/ver (Rent one month twice/year~552.1 x.2)     \$1,104     \$1,104     \$1,104       Sdays to harvest Southern Trails         Total 12 days/ear (Rent one month once/year~552.1)     \$552     \$552       Total 12 days/ear (Rent one month once/year~552.1)     \$555     \$552       Total 12 days/ear (Rent one month once/year field data collection/ two people/year (@180/hight/person)     \$4,320     \$4,320       Total 12 days to collect data @ Northern Trails (on hights)     \$2,160     \$2,160       Ways to collect data @ Northern Trails (on hights)     \$2,210     \$2,212       Ways to collect data @ Northern Trails (on hights)     \$2,212     \$2,212       Ways to collect data @ Northern Trails (on hights)     \$2,210     \$2,212       Ways to collect data @ Northern Trails (on hights)     \$2,210     \$2,212       Ways to collect data @ Northern Trails (on hights)     \$2,210     \$2,212       Ways to collect data @ Northern Trails (on hights)     \$2,210     \$2,212       Ways to collect data @ Northern Trails (on hights)     \$2,210     \$2,212       Ways to collect data @ Northern Trails (on hights)     \$1,006     \$1,006       Tays to collect data @ Northern Trails (on hights)     \$2,160     \$2,160       Assume hold (Strop)     \$2,160     \$2,160     \$2,160       Assume hold (Strop)     \$2,160     \$2,160  |  |                       |                       |                       |
| 5 days to harvest Southern Trails Total 12 days/year (Rent one month once/year=552.1) Total 12 days/year (Rent and (Ronthern Trails (O inghts) S2.160  | Total 12 days @ twice/year (Rent one month twice/year=552.1 x 2)                     | \$1,104               | \$1,104               | \$1,104               |
| 5 days to harvest Southern Trails Total 12 days/year (Rent one month once/year=552.1) Total 12 days/year (Rent and (Ronthern Trails (O inghts) S2.160  | Yield data collection once per year  |                       |                       |                       |
| 7 days to harvest Northern Trials [1000 and the set of  | 5 days to harvest Southern Trials  |                       |                       |                       |
| Total 12 days/year (Rent one month once/year-552.1)     5552     \$552       Medle for field data collection/ two people/2x per year (@2180/injt/person)     \$4,320     \$4,320       7 days to collect data @ Northern Trials (6 injths)     \$2,160     \$2,160       7 days to collect data @ Northern Trials (6 injths)     \$2,160     \$2,160       8 days to collect data @ Northern Trials (6 injths)     \$2,160     \$2,160       9 days to collect data @ Northern Trials (6 injths)     \$2,212     \$2,212       9 days to collect data @ Northern Trials (6 injths)     \$2,160     \$2,160       9 days to collect data @ Northern Trials (6 injths)     \$2,212     \$2,212       9 days to collect data @ Northern Trials (6 injths)     \$2,212     \$2,212       9 days to collect data @ Northern Trials     \$2,212     \$2,212       9 days to collect data @ Northern Trials     \$2,212     \$2,212       9 days to collect data @ Northern Trials     \$2,160     \$1,066       10 ares conversent data collection twice/year (8 days/year)     \$1,840     \$1,840       11 days on collect data @ Northern Trials     \$2,160     \$2,160       12 days collect data @ Northern Trials     \$2,160     \$2,160       13 days collect data @ Northern Trials     \$2,160     \$2,160       14 days collect data @ Northern Trials     \$2,160     \$2,160       14 days collect data @ Northern Trials  | 7 days to harvest Northern Trials  |                       |                       |                       |
| Intel for field data collection/ non people/32 per year (6.18/bright/person)     \$4,320     \$4,320       7 days to collect data (0) Northem Trials (6 nights)     \$2,160     \$2,160       7 days to collect data (0) Northem Trials (6 nights)     \$2,160     \$2,160       8.08 for field data collection/ no people/32 per year (6.79/day/person)     \$2,160     \$2,160       7 days to collect data (0) Northem Trials (0) nights)     \$2,212     \$2,212       8.08 for field data collection/ no people/32 per year (6.79/day/person)     \$1,06     \$1,106       7 days to collect data (0) Northem Trials     \$1,106     \$1,106       8.08 for field data collection/ no people/32 per year (6.79/day/person)     \$1,06     \$1,106       7 days to collect data (0) Northem Trials     \$1,106     \$1,106       8.10 for field data collection vice over (8 days/pear)     \$1,800     \$1,840       14aves to nuce/year (1 night)     \$1,840     \$1,840       Supplice Stor measure SWP nuce/year     \$1,840     \$1,840       Supplies to measure SWP nuce/year     \$1,840     \$1,840       SWP 600 trees x 2 leaves = 1200 readings total '1 x year     \$1,840     \$1,840       SWP 600 trees x 2 leaves = 1200 readings total '1 x year     \$430     \$420       Swin rogen partable tank (20 cc) mestre SWP weill get 1 bit tank     \$355     \$355       System water potentiablags (5 hogs/pack) each pack 540. Total 12 packs  | Total 12 days/year (Rent one month once/year=552.1)                                  | \$552                 | \$552                 | \$552                 |
| 2 days to collect data @ Northern Trials (6 nights)     \$4,320     \$4,320     \$4,320       7 days to collect data @ Northern Trials (6 nights)     \$2,160     \$2,160     \$2,160       7 days to collect data @ Northern Trials (6 nights)     \$2,210     \$2,212     \$2,212       7 days to collect data @ Northern Trials     \$2,212     \$2,212     \$2,212       7 days to collect data @ Northern Trials     \$2,212     \$2,212     \$2,212       7 days to collect data @ Northern Trials     \$2,160     \$1,106     \$1,106       8 days to collect data @ Northern Trials     \$2,160     \$1,106     \$1,106       1 days to collect data @ Northern Trials     \$2,160     \$2,160     \$2,160       A sum crimburschern Trials (5 nights)     \$2,160     \$2,160     \$2,160       A sume reinburschern Trials (5 nights)     \$2,160     \$2,160     \$2,160       A sume reinburschern Trials (5 nights)     \$2,160     \$2,160     \$2,160       A sume reinburschern Trials (5 nights)     \$2,160     \$2,160     \$2,160       Supplies to measure SWP oto trock     \$1,840     \$1,840     \$1,840       Supplies to measure SWP apot trock     \$200 readits of \$1,840     \$1,840     \$1,840       Supplies to measure SWP apot trock applies A road 12 packs     \$480     \$480     \$480       Sup apot collect apperimental Trees     \$200 r   | Hotel for field data collection/ two people/2x per year (@180/night/person)          |                       |                       |                       |
| 7 days to collect data (@ Northern Trials (6 nights)       \$2,160       \$2,160       \$2,160         Meak for field data collection/ two people/2x per year (@ 79/day/person)       \$2,212       \$2,212       \$2,212         Meak for field data (@ Northern Trials       \$2,212       \$2,212       \$2,212         Meak for field data (@ Northern Trials       \$1,106       \$1,106       \$1,106         Tarvel (Arpaia)       \$1,106       \$1,106       \$1,106         Travel (Arpaia)       \$2,160       \$2,160       \$2,160         Assume collection twice/year (1 night)       \$2,160       \$2,160       \$2,160         Assume robustement rute as RS approved       \$1,840       \$1,840       \$1,840         Supplies to measure SWP once/year       \$2       \$20       \$20       \$20         Best and worst combination at each field = 600 trees to measure       \$1       \$1       \$1         SWP = 600 trees x 2 leaves = 1200 readings total / 1x year       \$520       \$520       \$520       \$520         Big Nitrogen tank Ariga 2 Each refit two small ones (\$355/1). We will get 1 big tank \$355       \$355       \$355       \$355         Strogen gas to refit big tank \$35 each tank refit=570 per two = 100 trees total 6 refits       \$420       \$420       \$420         Supplies fo measure soft physiological rates       \$40  | 7 days to collect data @ Northern Trials (6 nights)                                  | \$4,320               | \$4,320               | \$4,320               |
| Meak for field data collection/ two people/2x per year (@79/day/person)       52,212       \$2,212         7 days to collect data @) Northern Trials       \$2,212       \$2,212         8 dash for harvest data collection/ two people/year (@79/day/person)       51,106       \$1,106         7 days to collect data @) Northern Trials       \$1,106       \$1,106         Travel (Arpaia)       51,106       \$1,106         Field data collection twice/year (8 days/year)       52,160       \$2,160         Assume 200 miles one-way as average       \$2,160       \$2,160         Assume reimbursement rate as IRS approved       \$1,840       \$1,840         Supplics to measure SWP once/year       \$1,840       \$1,840         Best and worst combination at each field = 600 trees to measure       \$2,520       \$520         SWP = 600 trees x 1 leaves = 1200 readings total/ 1x year       \$355       \$355         Suiftriggen tank Airgas 2. Each raft How small ones (\$355/1). We will get 1 big tank       \$355       \$355         Strongen gas to refill big tank \$35 each tank refill=570 per two = 100 trees total of refills       \$420       \$420         Supplies to measure SWP       \$480       \$480       \$480         Strongen gas to refill big tank \$355       \$355       \$355       \$355         Strongen gas to refill big tank \$35 each tank refill=570 per two   | Hotel for harvest data collection/ two people/year (@180/night/person)               |                       | ·                     |                       |
| 7 days to collect data @ Northern Trials \$2,212 \$2,210 \$2,  | 7 days to collect data @ Northern Trials (6 nights)                                  | \$2,160               | \$2,160               | \$2,160               |
| Meab for harvest data collection/ two people/year (@?9/day/person)       S1,106       S1,106         / days to collect data (@ Northern Traits)       S1,106       S1,106         Fravel (Arppaia)       S1,106       S1,106         Field data collection twice/year (8 days/year)       S2,160       S2,160         Harvest once/year (1 night)       S2,160       S2,160         Assume 200 miles one-way as average       S2,160       S2,160         Assume to meables one-way as average       S2,160       S2,160         Supplies to measure SWP once/year       S       S2,00       S2,00         Best and worst combination at each field = 600 trees to measure       SVP = 600 trees x 2 leaves = 1200 readings total 1 x year       S2,00       S520       S520 <td< td=""><td>Meals for field data collection/ two people/2x per year (@79/day/person)</td><td></td><td></td><td></td></td<>  | Meals for field data collection/ two people/2x per year (@79/day/person)             |                       |                       |                       |
| 7 days to collect data @ Northem Trials       \$1,106       \$1,106       \$1,106         Travel (Arpaia)            Field data collection twice/year (8 days/year)            Harvest once/year (1 night)             Assume 200 miles one-way as average       \$2,160       \$2,160       \$2,160       \$2,160         Assume coll miles one-way as average       \$2,160       \$1,840       \$1,840       \$1,840         Supplies to measure SWP once/year             Best and worst combination at each field              Swup = 600 trees x 2 leaves = 1200 readings total '1 xy ear  | 7 days to collect data @ Northern Trials   | \$2,212               | \$2,212               | \$2,212               |
| Travel (Arpaia)       Field data collection twice/year (8 days/year)         Field data collection twice/year (1 high)       S2,160         Assume 200 miles one-way as average       \$2,160         Assume brief (160) and meals (570) = 5230day       \$1,840         Supplies to measure SWP once/year       S1,840         Best and worst combination at each field       S1,840         Stress of the best and worst combination at each field = 600 trees to measure       S1,840         SWP = 600 trees x 2 leaves = 1200 readings total/ 1x year       S120         SMall Nitrogen portable tank (20 cc) mesure 50 -plants. Each tank (a)250 we will get 2       S520         Sind Nitrogen nortable tank (20 cc) mesure 50 -plants. Each tank (a)250 we will get 2       S520         Sind Nitrogen so tarefill big tank \$35 each tank refill=570 per two = 100 trees total 6 refills       \$420         Stem water potential bags/pack) each pack \$40. Total 12 packs       \$480         Supplies Avocado Experimental Trees       S6,336         Supplies for SWP       S6,336         Supplies for SWP       S210         Supplies for SWP       S210         Supplies for SWP       S210         Supplies for SWP       S210         Stem water potential bags (Ob bags/pack) each pack \$40. Total 12 packs       S480         SWP = 288 trees 10 physiological rates       S210   | Meals for harvest data collection/ two people/year (@79/day/person)                  |                       |                       |                       |
| Field data collection twice/year ( 8 days/year)       Imight         Harvest once/year (1 night)       S2,160         Assume 200 miles one-way as average       \$2,160         Assume 200 miles one-way as average       \$2,160         Assume 200 miles one-way as average       \$1,840         Stupplies to measure SWP once/year       Imight         Best and worst combination at each field       Imight         30 tress of the best and worst combination at each field       Imight         50 tress of the best and worst combination at each field       Imight         SwP = 600 tress x 2 leaves = 1200 readings total/ 1x year       Imiget and thirding as the field ones (\$3557). We will get 1 big tank         Sintrogen portable tank (20 cc) mesure 50 - plants, Each tank (@\$260 we will get 2       \$520       \$520         Sintrogen portable tank (20 cc) mesure 50 - plants, Each tank (@\$260 we will get 2       \$520       \$520         Sintrogen portable tank (20 cc) mesure 50 - plants, Each tank (@\$260 we will get 2       \$520       \$520         Sintrogen portable tank (20 cc) mesure 50 - plants, Each tank (@\$260 we will get 2       \$520       \$520         Sintrogen gara the field 50 to ess to tal (2 packs)       \$480       \$440         Obj. 2 GH experiments       \$420       \$420       \$420         Supplies for Messing & Sis each tank refifles 70 per two = 100 trees. Total 3 refills   | 7 days to collect data @ Northern Trials   | \$1,106               | \$1,106               | \$1,106               |
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| Assume 200 miles one-way as average       \$2,160       \$2,160       \$2,160         Assume neimbursement rate as IRS approved       \$1,840       \$1,840       \$1,840         Assume hotel (\$160) and meaks (\$70) = \$230/day       \$1,840       \$1,840       \$1,840         Supplies to measure SWP once/year       \$1,840       \$1,840       \$1,840         Best and worst combination at each field       \$1,840       \$1,840       \$1,840         SWP = 600 trees x 2 leaves = 1200 readings total/ 1x year       \$520       \$520       \$520         Small Nitrogen portable tank (\$20 c) mesure 50 - plants. Each tank (\$260 we will get 2       \$550       \$355       \$355         Silrogen gas to refill big tank \$35 each tank refill=570 per two = 100 trees total 6 refills       \$420       \$420       \$420         Sterw water potential bags (50 bags/pack) each pack \$40. Total 12 packs       \$480       \$480       \$480         Supplies Avocado Experimental Trees       Trees = 6 RS x 2 (ungrafted vs Hass) x 4 treatments x 6 reps = 288       \$6,336       \$6,336         Supplies to measure SWP       \$10       \$210       \$210       \$210         Strees or SWP       \$220       \$420       \$240       \$220         Supplies for SWP       \$6,336       \$6,336       \$6,336       \$6,336         Supplies for SWP       <   | Field data collection twice/year (8 days/year)                                       |                       |                       |                       |
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| Supplies to measure SWP once/year  | Asume reimbursement rate as IRS approved   |                       |                       |                       |
| Best and worst combination at each field 30 trees of the best and worst combination at each field = 600 trees to measure SWP = 600 trees x 2 leaves = 1200 readings total/ 1x year SWP = 600 trees x 2 leaves = 1200 readings total/ 1x year SWP = 600 trees x 2 leaves = 1200 readings total/ 1x year SWP = 600 trees x 2 leaves = 1200 readings total/ 1x year Singul Nitrogen portable tank Airgas 2 Each refilt S00 per two = 100 trees total 6 refilts S420 S420 S420 S420 S420 S420 S420 S420  | Assume hotel ( $\$160$ ) and meals ( $\$70$ ) = $\$230/day$                          | \$1,840               | \$1,840               | \$1,840               |
| 30 trees of the best and worst combination at each field = 600 trees to measure          SWP = 600 trees x 2 leaves = 1200 readings total/ 1x year          Small Nitrogen partable tank (20 cc) mesure 50 -plants. Each tank @260 we will get 2       \$520       \$520         Big Nitrogen tank Airgas 2 Each refill two small ones (\$355/1). We will get 1 big tank       \$355       \$355         Nitrogen gas to refill big tank \$35 each tank refill=570 per two = 100 trees total 6 refills       \$4420       \$4420         Stem water potential bags (50 bags/pack) each pack \$40. Total 12 packs       \$480       \$480       \$480         Obj. 2 GH experiments             Supplies Avocado Experimental Trees              Supplies to measure SWP        \$6,336       \$6,336       \$6,336       \$6,336       \$6,336       \$6,336       \$6,336       \$6,336       \$6,336       \$6,336       \$6,336       \$6,336       \$500       \$6,336       \$6,5336       \$500       \$6,336<   | Supplies to measure SWP once/year  |                       |                       |                       |
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| SWP = 600 trees x 2 leaves = 1200 readings total/ 1x year       \$520         Small Nirogen partable tank (20 cc) mesure 50 -plants. Each tank (\$260 we will get 2       \$520       \$520         Big Nitrogen tank Airgas 2 Each refill two small ones (\$355/1). We will get 1 big tank       \$355       \$355         Sind Nirogen gas to refill big tank \$35 each tank refill=\$70 per two = 100 trees total 6 refills       \$420       \$420         Stem water potential bags (50 bags/pack) each pack \$40. Total 12 packs       \$480       \$480       \$480         Obj. 2 GH experiments       \$420       \$420       \$420         Supplies Avocado Experimental Trees       \$6,336       \$6,336         Trees= 6 RS x 2 (ungrafted vs Hass) x 4 treatments x 6 reps = 288       \$6,336       \$6,336         Supplies for SWP       \$6 rees per combination = 288 trees for physiological rates       \$600       \$6,336         SWP = 288 trees x 2 leaves = 576 readings total/ 1x year       \$210       \$210       \$210         Nitrogen gas to refill big tank \$35 goach tank refill=\$70 per two = 100 trees. Total 3 refills       \$480       \$440         SWP = 288 trees for physiological rates       \$210       \$210       \$210         Stem water potential bags (50 bags/pack) each pack \$40. Total 12 packs       \$480       \$4480         Ionomic analyses (Only Na + and Cl-) by FGL       \$6,480       \$6,480 <td< td=""><td>30 trees of the best and worst combination at each field <math>= 600</math> trees to measure</td><td></td><td></td><td></td></td<>  | 30 trees of the best and worst combination at each field $= 600$ trees to measure    |                       |                       |                       |
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| Obj. 2 GH experiments         Supplies Avocado Experimental Trees          Trees- 6 R5 x 2 (ungrafted vs Hass) x 4 treatments x 6 reps = 288          Liners @ \$22/liner       \$6,336         Supplies to measure SWP          Supplies for SWP          6 trees per combination = 288 trees for physiological rates          SWP = 288 trees x 2 leaves = 576 readings total/ 1x year          Nitrogen gas to refill big tank \$35 each tank refill=\$70 per two = 100 trees. Total 3 refills       \$210         Stem water potential bags (50 bags/pack) each pack \$40. Total 12 packs       \$4480         Ionomic analyses (Only Na+ and Cl-) by FGL          6 R5x 2 condx 2 treat x 3 reps x 2 sample type (root/leaf). @\$45/sample Na/Cl, FGL)       \$6,480         General lab supplies       \$3,000       \$3,300       \$3,500         GH fees PR1 300/month (6 month each exp)       \$12,00       \$1,200       \$1,200  |  | \$480                 | \$480                 | \$480                 |
| Supplies Avocado Experimental Trees       Image: Supplies Avocado Experimental Trees       Image: Supplies Avocado Experimental Trees         Trees= 6 RS x 2 (ungrafted vs Hass) x 4 treatments x 6 reps = 288       Image: Supplies for SWP       Supplies to measure SWP         Supplies for SWP       Image: Supplies for SWP       Image: Supplies for SWP       Image: Supplies for Formation and the supplies for Formation and the supplies for Formation and the supplies for two and the supplies for two and the supplies for two and the supplies       Supplies for SWP       Image: Supplies for Formation and the supplies for Formation and the supplies for two and the supplies for two and the supplies for two and the supplies for two a   |  |                       |                       |                       |
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| Liners @ \$22/liner       \$6,336       \$6,336         Supplies to measure SWP  | •••  |                       |                       |                       |
| Supplies to measure SWP       Image: Swp of the system of th   |  |                       | \$6.226               | \$6.226               |
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| 6 RSx 2 condx 2 treat x 3 reps x 2 sample type (root/leaf). @\$45/sample Na/Cl, FGL)         \$6,480         \$6,480           General lab supplies         \$3,000         \$3,300         \$3,500           GH fees PR1 300/month (6 month each exp)         \$1,200         \$1,200         \$1,200           SUBTOTAL         77,015         136,430         85,94   |  |                       | \$480                 | \$480                 |
| General lab supplies         \$3,000         \$3,300         \$3,500           GH fees PR1 300/month (6 month each exp)         \$1,200         \$1,200         \$1,200           SUBTOTAL         77,015         136,430         85,94  |  |                       |                       |                       |
| GH fees PR1 300/month (6 month each exp)         \$1,200         \$1,200           SUBTOTAL         77,015         136,430         85,94   | 6 RSx 2 condx 2 treat x 3 reps x 2 sample type (root/leaf). @\$45/sample Na/Cl, FGL) |                       | \$6,480               | \$6,480               |
| GH fees PR1 300/month (6 month each exp)         \$1,200         \$1,200           SUBTOTAL         77,015         136,430         85,94   | General lab supplies   | \$3,000               | \$3,300               | \$3,500               |
| SUBTOTAL         77,015         136,430         85,94  | GH fees PR1 300/month (6 month each exp)   |                       | \$1,200               |                       |
|  |  | 77.015                |                       |                       |
| TOTAL 299,392.0  | SUBIUTAL   | //,015                |                       |                       |
|  |  |                       | TOTAL                 | 299,392.00            |

#### **BUDGET JUSTIFICATION** Total UCR budget requesting for three years: \$299,392

- A. Personnel Salary (\$117,396). Funds are requested to cover the salary for: i) an Assistant Specialist I at 50% EFT for every year of the project and ii) one Graduate Student Researcher (GSR) I for two Summer Quarters for the two first years of the project. The Assistant Specialist I and GSR I will be under the supervision of Dr. Manosalva and will conduct all the research activities for this project. Both personnel have been working since 2021 for the rootstock breeding program directed by Manosalva and have been trained to conduct all research activities in the field (Obj. 1) and greenhouse (Obj. 2) as well as to conduct statistical analyses. In addition, both personnel are in constant communication with our grower collaborators and organize & manage all the field and GH activities for the program; thus, they are essential personnel to conduct all the activities for this proposal. Field data collection and harvest at all fields will be conducted with the assistance of Manosalva. Dr. Arpaia will oversee the data collection at Pine Tree with Manosalva and the Assistant Specialist and GSR.
- **B. Fringe Benefits (\$43,787).** Employee benefits are estimates, using the composite benefit rates agreed upon by the University of California. The composite benefit rate for the Assistant Specialist I is 43.7% and for the GSR I is 2.1%. Subsequent years include escalations based on recommendations by our campus administrative office.
- **C. Domestic travel (\$46,362).** Funds are requested to cover the travel expenses for the Assistant Specialist and the GSR to travel at all 10 field trials described in **Table 1**. Funds requested include the cost of a cargo van rental from UCR fleet services at a monthly rate of \$552.1 to collect field data from all the plots (12 days/once per year). *Notice is cheaper rent by month than by day (\$55.21/day).* A cargo van is required to fit all the equipment required for tree measurement, coolers for samples, bins for harvest, digital scale, etc. Trips conducted to Southern California plots have been budgeted as single day trips. For Northern trials, travel cost include lodging with an average rate of \$180/night at the hotel in Ventura area and meals at a per diem rate of \$79/day. Travel cost includes the travel expenses for Dr. Arpaia to oversee the data collection at harvest at the Pine Tree trial including reimbursement for mileage at a rate at currently approved IRS rates, hotel at \$160/night, and meals at \$70/day.
- D. Supplies (\$29,177). Funds are requested to purchase avocado experimental trees (liners) described in Obj. 2. Ungrafted and 'Hass'-grafted trees corresponding to commercial rootstocks will be ordered from Brokaw Nursery (\$22/tree) to conduct two replicated experiments for publication purposes. Funds will be covered supplies to measure stem water potential (SWP) as described in Obj.1 & 2 including the purchase of: i) 2 small portable (\$266/unit) & 1 big (\$355/unit) nitrogen gas tanks to be used with our pressure chamber instrument, ii) Nitrogen gas refills (\$35/refill for big tank), and iii) SWP bags at \$40 per pack of 50 bags. Money is requested for general supplies and consumables including UC Mix soil, chemicals to prepare solutions for salinity treatments, fertilizers, tree labels, tree sticks, ziploc bags for sample collection, media to prepare pathogen inoculum and for pathogen isolation, pipette tips, tubes, petri dishes, and gloves. These supplies were estimated based on historical amounts and cost of similar research projects and activities at the Manosalva Laboratory.
- E. Services and others (\$62,670). Funds are requested to conduct irrigation water suitability analyses (\$95/sample) and soil comprehensive analyses (\$57/sample) for all fields once per year and will be done by Fruit Growers Lab (FGL, Santa Paula). Money is requested to cover ionomic leaf and root profile analyses at a rate of \$75 per sample (FGL, Santa Paula). These analyses will be conducted in trees evaluated under Obj. 1 to correlate their salinity responses to leaf and root ionomic profiles in order to elucidate the mechanism(s) explaining these responses and phenotypes. We are requesting funds to conduct leaf and root Na+ and Cl- content in trees exposed to salinity under greenhouse conditions (Obj. 2) at a rate of \$45/sample (FGL, Santa Paula). Finally, funds are also requested to cover UCR greenhouses fees at a rate of \$200/month (~300 Sq ft) for 6 months (each salinity experiment) described in Obj. 2. Manosalva used regularly these greenhouses with better temperature and humidity controllers to conduct water stress experiments in avocado (drought and salinity).

# **Project Narrative**

**Project Title**: Assessing irrigation management tools and strategies on avocado fruit quality and yield impacts

**Project Lead:** Ali Montazar, Irrigation and Water Management Advisor, UCCE San Diego, Riverside, and Imperial Counties; email: <u>amontazar@ucanr.edu</u>.

**Project Cooperator:** Ben Faber, Subtropical Crops Advisor, UCCE Ventura and Santa Barbara Counties; email: <u>bafaber@ucdavis.edu</u>.

Executive Summary: Careful water management is critical to ensure optimal yield and highquality avocado fruits. This is even more important under avocado crop production systems in California due to uncertain water supplies, mandatory reductions of water use, the rising cost of water, and increasing salinity in water sources. We have conducted extensive data collection and analysis over the last three years on 12 avocado commercial sites. Through this past study, seasonal crop coefficient (Kc) curves have been updated for California avocados, as well as an evaluation of avocado crop water consumption conducted under different environments and orchard features. While we developed more accurate seasonal Kc values and a better understanding of the efficacy of irrigation tools in CA avocados, a second phase of this study needs to be carried out assessing the developed Kc values in regards with avocado fruit quality and yield impacts. This is a necessary phase that may provide growers with a high level of confidence to adopt the information and enhance the efficiency of water use in avocados. This new study intends to evaluate the impact of irrigation management using the developed seasonal Kc curve and other cost effective and user-friendly tools in California avocados. It is expected that the tools and information under development by this study will enable more efficient resource- use irrigation management and long-term sustainability in avocado production.

List of specific project objectives: This project aims to assess the impact of irrigation tools and management strategies to optimize water-use efficiency and economic productivity in avocado production systems. Enhancing water-fertilizer, and energy-use efficiency, water conservation, water quality, and economic gains of avocado growers are the primary goals that this study will address. The project specifically aims to:

- verify the developed Kc seasonal curves for California Hass avocados in regards with avocado fruit quality and yield impacts.
- assess the impact of irrigation tools (ET-based irrigation, OpenET satellite data, soil moisture sensing, Implexx Sap Flow sensor) and irrigation management strategies (various water application rates) on yield and fruit quality of avocados.
- quantify water use efficiency enhancement following improved irrigation management practices.
- disseminate project findings to growers and stakeholders.

# List of specific project deliverables:

• evaluation of ET-based irrigation scheduling using the developed Kc values on avocado fruit quality and yield impacts.

- evaluation of irrigation management using OpenET satellite data on avocado fruit quality and yield impacts.
- the effectiveness of soil moisture sensing and Implexx Sap Flow sensor on improving avocado irrigation management.
- evaluation of various irrigation regimes on avocado fruit quality and yield impacts.
- assessing the impact of irrigation tools on water use efficiency and water conservation.
- assessing leaching requirements of avocado orchards over season/s.

**Background:** The PI of this project has recently completed an irrigation study to better understand the impacts of environmental and plant factors on crop water use and to develop more precise crop coefficient values for California Hass avocado production systems. The study was conducted in 12 avocado sites in southern California (Fig. 1).



Fig. 1. A demonstration of flux tower monitoring station and some of the instrumentation set up.

While a similar crop water use pattern was found over the course of the measurement seasons in avocado experimental sites, considerable differences were found in the seasonal ET (actual evapotranspiration) amounts determined across avocado sites and seasons. For instance, an 11.4-in difference in the seasonal consumptive water use was determined amongst the four avocado sites in 2024 (Fig. 2).

The results of this study clearly show that avocado crop water use varies spatially and temporally. The greatest seasonal crop water consumption was determined at an avocado site (site A) with the features of coarse sandy loam soil texture, 44% south facing slope, average elevation of 758 ft. above mean sea level, plant density of 120 trees per acre, mean canopy coverage of 88.7% and tree height of 23.2 ft. In contrast, the least seasonal crop water use was observed at an avocado site (site D) affected by coastal climate with the features of loamy soil

texture, 3% southwest facing slope, average elevation of 164 ft. above mean sea level, plant density of 254 trees per acre, mean canopy coverage of 75.9% and tree height of 12.5 ft.

The results illustrate that avocado has the lowest crop coefficient values during the summer months, increasing gradually from late September to a maximum in mid-winter, again gradually reducing during spring to a minimum in mid-summer (Fig. 3). To be more precise, the findings revealed greater crop coefficient values of avocados during flower bud development, and flowering through fruit set growth phases than the fruit development phase.

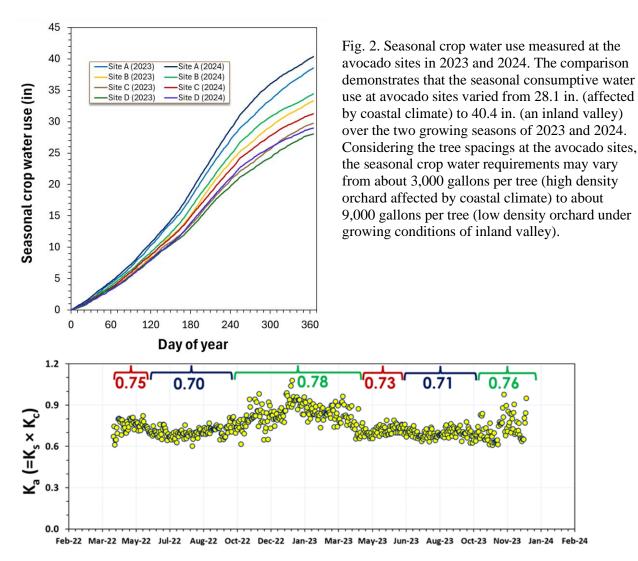


Fig. 3. Avocado crop coefficient curves over two growing seasons in a high water-use avocado site.

**Work Plan and Methods:** The field experiment will be conducted in two avocado research sites equipped with the flux tower over a three-year period, one in Temecula and one in Escondido. The seasonal Kc curve had been already developed for these sites. Four irrigation strategies will be arranged in a Randomized Complete Block Design with six replications (six trees per irrigation strategy: for the analysis, we will consider three tree sets consisting of two trees per set per each irrigation treatment to consider soil variability and the impact of top-bottom of slope)

(Fig. 4). The irrigation strategies will consist of (1) grower practice the entire growing season as control treatment, (2) 100% ETc, (3) 80% ETc, and (4) irrigation based on the best OpenET model identified for avocados (an assessment of OpenET models will be undertaken for avocados using the flux tower data and the results will be used for the irrigation strategy 4). ETc will be determined using the Kc values developed for the sites and spatial CIMIS ETo data (ETc=Kc × ETo). It needs to be noted that the leaching requirements will be added to ETc in irrigation treatments 2-4. The assumption is that grower irrigation practice provides an over irrigation strategy in this study. Our earlier date collected from several avocado sites verifies this assumption.

The soil water status will be monitored within the soil profile, depths of 6 through 36 in., in each treatment using two different types of soil moisture sensors measuring soil water potential and volumetric water content. A precision irrigation system will be set up to accurately monitor water applied (using digital flowmeter) and deliver irrigation water in each treatment. EM-38MK2 will be run to develop salinity maps in the experimental areas of each site. Soil salinity will be evaluated twice per year, mid-August and early May and the required leaching will be performed as needed. In addition, soil solution access tubes will be installed at the depths of 1 to 3 ft to monitor ECe, chloride, and nitrate-N of soil solution on a regular basis.

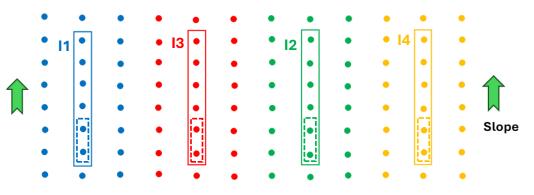


Fig. 4. Layout of experimental sites. Dots with similar colors demonstrate avocado trees under a similar irrigation strategy. Six central trees in each irrigation strategy (I1 - I4) will be considered for monitoring and yield assessment (three sets of trees consisting of two trees per set per each irrigation treatment to consider soil variability and the impact of top-bottom of slope). The experiment will be conducted in about 0.6 acres in two different mature avocado sites. All experimental tress in each site will be on the same row (predominant slope) orientation.

Implexx Sap Flow sensor will be utilized to measure trees transpiration as well as Leaf Porometer to monitor stomatal conductance. Monitoring plant water status will be conducted using dendrometers on a continuous basis along with pressure chamber readings (three times per month between May and September). In addition, the difference of canopy temperature versus air temperature recorded by fixed view-angle infrared thermometers along with aerial imagery and analysis will be used to evaluate crop water stress indices. Continuous normalized difference vegetation index (NDVI) values will be measured by Spectral Reflectance sensors. All data will be measured and transferred using telemetry devices on a continuous basis. Canopy reflectance in the visible and near infrared regions of the electromagnetic spectrum will be measured through high-resolution, multi-spectral, and thermal cameras that will be carried by an unmanned aerial system on three different days per season. Water distribution uniformity will be evaluated using the standard evaluation methods for micro irrigation systems.

The agronomical performance of irrigation strategies will be also assessed during the seasons by monitoring fertilizations, foliar nutrient content and fruit yield. Avocado fruits are gradually harvested from February to April to assess yield and water productivity. To evaluate the fruit size (i.e. indicative of commercial quality), fruits are analyzed and classified into different size-classes according to their weights. The percentage of dry matter is also analyzed in 10 randomly selected fruits per irrigation treatment with a Near Infrared Analyzer (NIR).

**Project Outreach:** A robust outreach program will be developed to disseminate project findings to growers and stakeholders. We will hold three avocado irrigation workshops. The findings will also be presented at the grower meetings of the CAC and at the Avocado Café. Results will be published as extension publications in *Topics in Subtropics* and *Extension Connection* newsletters, CAC- *from the Grove Magazine*, and UC blogs and as scientific articles in peer-reviewed journals. The PI will participate and present the project findings at the 11<sup>th</sup> World Avocado Congress and the American Society for Horticulture Science (ASHS) annual conference.

**Milestone Table:** The project milestones of Year 1 - Year 3 are given in Table 1. Starting this project from July 2025 provides the research team with better time management to gather a three-year yield data and ensure a more comprehensive assessment of irrigation strategies. It might be a little bit weird, but to stay with the CAC fiscal years, we need to consider Year 1 – Year 3 as follows; Year 1: July 1, 2025 – October 31, 2026 (15 months), Year 2: November 1, 2026 – October 31 (12 months), 2027, and Year 3: November 1, 2027 – June 30, 2028 (9 months).

| Milestone     | Activities  | Time       | Estimated budget |  |
|---------------|---|------------|------------------|--|
|               |   | completion | amount (\$)      |  |
|               | Purchase the special purpose equipment.             | Jul 2025   | 29,500           |  |
|               | Field visits to finalize the exact locations of     | Jul 2025   |                  |  |
|               | experimental sites.                                 | Jul 2023   |                  |  |
|               | Set up field experiments in two avocados sites      |            | 5,000            |  |
| M1 – Year 1   | including irrigation treatments and sensor          | Aug 2025   |                  |  |
| MI = I cal I  | installation.                                       |            |                  |  |
|               | Run irrigation treatments.                          | Mar 2026   | 40,913           |  |
|               | Regular data collection (soil, plant, water, yield, |            |                  |  |
|               | aerial imagery), sensor and equipment maintenance,  | Mar 2026   |                  |  |
|               | and data analysis. Conduct salinity survey.         |            |                  |  |
|               | Run irrigation treatments                           | Oct 2026   |                  |  |
|               | Regular data collection (soil, plant, water, aerial |            |                  |  |
| M2 – Year 1   | imagery), sensor and equipment maintenance, and     | Oct 2026   | 40,912           |  |
| NIZ - I ear I | data analysis.                                      |            | 40,912           |  |
|               | Hold Avocado Irrigation Workshop.                   | Jul 2026   |                  |  |
|               | Publish extension article.                          | Sep 2026   |                  |  |
| M1 – Year 2   | Run irrigation treatments.                          | Mar 2027   | 28,000           |  |

Table 1. Project milestones of Year 1 – Year 3. Each year consists of two milestones (M1 and M2).

|             | Regular data collection (soil, plant, water, yield),<br>sensor and equipment maintenance, and data  | Mar 2027 |        |
|-------------|---|----------|--------|
|             | analysis.<br>Develop University of California blogs and various<br>web-based platforms to share science-based<br>information.                     | Feb 2027 |        |
|             | Run irrigation treatments.  | Oct 2027 |        |
|             | Publish extension article.  | Sep 2027 | -      |
| M2 – Year 2 | Regular data collection (soil, plant, water, aerial<br>imagery), sensor and equipment maintenance, and<br>data analysis. Conduct salinity survey. | Oct 2027 | 29,370 |
|             | Hold Avocado Irrigation Workshop.   | Jul 2027 |        |
|             | Publish extension article.  | Sep 2027 |        |
|             | Participate in and present the project findings in national/international conference.   | Sep 2027 |        |
|             | Run irrigation treatments.  | Mar 2028 |        |
|             | Regular data collection (soil, plant, water, yield),<br>sensor and equipment maintenance, and data<br>analysis.                                   | Mar 2028 |        |
| M1 – Year 3 | Publish extension articles.   | Mar 2028 | 32,000 |
|             | Develop University of California blogs and various<br>web-based platforms to share science-based<br>information.                                  | Mar 2028 |        |
|             | Run irrigation treatments.  | Apr 2028 |        |
|             | Regular data collection (soil, plant, water, aerial<br>imagery), sensor and equipment maintenance, and<br>data analysis. Conduct salinity survey. | Apr 2028 |        |
| M2 – Year 3 | Hold Avocado Irrigation Workshop.   | May 2028 | 13,415 |
|             | Participate in and present the project findings in national/international conference.   | May 2028 |        |
|             | Publish extension articles.   | Jun 2028 | ]      |
|             | Publish peer-reviewed journal article.  | Jun 2028 |        |

# **Project Budget**

**Budget Detail:** A total budget of \$219,110 is requested for conducting this project (July 1, 2025 – June 31, 2028). The details of the budget can be found in Table 2.

|   | Budget (\$) |          |          |          |             |
|---|-------------|----------|----------|----------|-------------|
| Item  | Year 1      |          | Year 2   | Year 3   | Total       |
|   | FY 24-25    | FY 25-26 | FY 26-27 | FY 27-28 | budget (\$) |
| Personnel                                   |             |          |          |          |             |
| Staff research associate salary             | 7,250       | 29,000   | 29,000   | 21,750   | 87,000      |
| Staff research associate fringe benefits    | 4,205       | 16,820   | 16,820   | 12,615   | 50,460      |
| Graduate student salary and fringe benefits | -           | 6,250    | 6,250    | 6,250    | 18,750      |
| (to be determined)                          |             |          |          |          |             |
| Personnel subtotal                          | 11,455      | 52,070   | 52,070   | 40,615   | 156,210     |
| Supplies                                    |             |          |          |          |             |
| 3-D sonic anemometer (no=2)                 | 7,500       | -        | -        | -        | 7,500       |
| CR3000 datalogger (no=6)                    | 3,000       | -        | -        | -        | 3,000       |
| digital flowmeter (no=6)                    | 2,000       | -        | -        | -        | 2,000       |
| soil moisture sensor (no=9)                 | 9,000       | -        | -        | -        | 9,000       |
| Implexx Sap Flow Sensor (no=12)             | 8,000       | -        | -        | -        | 8,000       |
| Supplies subtotal                           | 29,500      | -        | -        | -        | 29,500      |
| Travel                                      |             |          |          |          |             |
| Travel to the experimental sites            | 2,000       | 3,000    | 3,00     | 2,500    | 10,500      |
| Other costs                                 |             |          |          |          |             |
| Scaffolding structures to set up sensors    | -           | 16,000   | -        | -        | 16,000      |
| above canopy (no=2)                         |             |          |          |          |             |
| Soil/water/plant lab analysis               | -           | 1,500    | 1,500    | 1,500    | 4,500       |
| Cell phone modem services                   | -           | 800      | 800      | 800      | 2,400       |
| Total                                       | 42,955      | 73,370   | 57,370   | 45,415   | 219,110     |

## **Budget Narrative:**

1- Personnel: A Staff research associate (SRA) will be recruited for the project who will help the research team with setting up and performing the irrigation treatments, monitoring stations and sensors in the experimental orchards, tuning up the instruments, collecting field data and conduct analysis, performing other field activities and sensors maintenance, and participating in the outreach program. For a three-year period, the average annual salary of the SRA is estimated to be \$58,000 and the fringe benefits are assumed at 58% of salary. We expect this project to support 50% FTE of the SRA salary and fringe benefits in each year over a three-year period.

A graduate student will be hired to work 750 hours at a projected average rate of \$25 per hour (fringe benefits included) to help the research team with aerial imaging and data analysis.

2- Supplies: While the PI will use some available sensors and equipment in his lab for this study, there are still some supplies that need to be purchased by this project including 3-D sonic

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anemometer (81000 RE), CR3000 datalogger, digital flowmeter, soil moisture sensor, and Implexx Sap Flow Sensor.

3- Travel: The PI, SRA, and graduate students have several multiple-day (an average of two days per trip) trips for installation of monitoring equipment and sensors at the experimental sites, data collection, aerial imaging, take down of the monitoring stations, grower meetings, and workshops. A total of 32 trips is estimated with an average of 310 miles per trip. The project estimate for travel expenses is 9,920 miles (\$0.67 per mile), 18 nights lodging (\$170 per night), 16 days per diem (\$60 per day).

4- Scaffolding structures for monitoring towers are required. Renting materials, dismantling scaffolding and demobilizing assembling is at an average flat rate of \$8,000 per site.

5- Soil/water/plant lab analysis: soil, water, and plant analysis will be conducted by the UC Davis laboratory. The project will have an estimated 120 samples which will each be analyzed for five factors/parameters. The cost per sample is an average cost of \$15 for each factor analysis.

6- Cell phone modems will be used to transfer real time data of monitoring stations. The monthly phone service for each cell modem has an average rate of \$200 per year for each cell modem (Verizon wireless service). This service requires four cell modems over a three-year period.



# **Project Narrative**

**Project Title** 

Chloride Removal and Sequestration System for Irrigation Water

**Project Lead** 

John Skardon, Ph.D. CEO, Founder Tailwater Systems LLC

**Project Cooperators** 

Maureen Cottingham Cam Lam Farms 2577 Hilltop Land Camarillo, CA 93012

**Executive Summary** 

Excess chloride in existing aquifers and irrigation water damages many types of fruit trees including avocados. To date, there has been no demonstrated method that can cost-effectively remove chloride from irrigation water and sequester it so it cannot re-enter the local ecosystem. The purpose of this test is to demonstrate the effectiveness of our solution to solve this long standing environmental challenge facing California's avocado growers.

For this project, Tailwater plans to install a <u>YieldMax</u> 5 GPM chloride removal system with a 100 unit <u>PhytoVap</u> evaporator array near the city of Camarillo, CA. The property is managed by our cooperator- Cam Lam Farms. We will install and validate the system by October 30 2025, and plan to operate the system continuously during the 2026 growing season: Feb 1st 2026 to August 1 2026. The test will consist of at least two lines of avocado trees. Group 1 (control) is irrigated using the existing water supply. Group 2 (treatment) will be irrigated using treated water from their contaminated well (CI > 190mg/L). Both groups will follow the historical irrigation schedule and nutrient application. Weekly water samples will be taken from the treatment system during operation and mailed to Tailwater. Weekly NDVI images of the test area and surrounding grove will be acquired via drone to provide a time lapse view of the possible effects of the chloride free water application.



# Scalability and Throughput

The system has two major components that work together. The YieldMax unit strips the chloride from the irrigation water, storing it in a concentrated brine solution. The companion PhytoVap array evaporates the brine and sequesters the chloride. The plants in our PhytoVap array tolerate 9,000-10,000 mg/L of chloride easily.

YieldMax systems can be scaled easily to 100's of gallons per minute by purchasing a commercial off-the-shelf <u>deionizer</u> or equivalent system in a lead-lag configuration. Tailwater adds the resin and necessary installation and ammonia recovery subsystem for chloride removal. The actual throughput required is driven by the amount of chloride removal. For example If chloride is near 200 mg/L and we want to lower it by 50%, then the smartest approach is to treat ½ of the flow to remove all the chloride then blend the treated and untreated streams during irrigation.

Large PhytoVap systems use 24 liter buckets to contain our cordgrass. Plants in the larger buckets can evaporate 2 or more gallons per day. A 1000 bucket system can evaporate 2000 gallons per day of brine during normal operation. At 4% brine flow (worst case) this equates to about 50,000 gallons per cycle (normally 8 hours) of treated water or 100 GPM flow rate. Our most recent project required 5,000 ft2 for 1200 buckets (20 laterals with 60 buckets per string). A key point about PhytoVap- <u>once water enters this system it never touches the ground</u>.

One approach to scaling up would have the customer installing an above ground reservoir to store the treated chloride-free water. In 3 days of operation between irrigation events, running only 1 shift per day (8 hours cycle time), the 100 GPM system would produce 150,000 gallons every 3 days. If we assume that we are treating 50% of the total flow, then *the total amount* of low-chloride water available for irrigation would be 300,000 gallons every 4 days.

Assuming 20 gallons of water per tree per day, this small 100 GPM system could irrigate 15,000 trees. Using the UC Davis estimate of 430 trees per acre ( high density planting) this would cover 35 acres. Using the conventional 145 trees per acre, this would satisfy about 100 acres.

Another approach along the same line of thinking is to operate the system very heavily when irrigation demand is lowest (November-February). Chloride-free water can be stored in above ground reservoirs over the winter months.

Finally, given the restriction of having to treat the brine and evaporate onsite, the size of the PhytoVap array could become limiting. With the planned extension of the brine line in Southern California, it may make financial sense to haul some of the brine to the brine line for disposal. In summary, there are many ways to leverage the functionality of our system that are beyond the scope of this proposal.



# **Project Objectives**

The objectives of this proposal are:

- 1. Demonstrate system ability to remove and sequester chloride
- 2. Document operating costs
- 3. Document changes in tree health with chloride-free water

# Deliverables

- 1. A 5 GPM (2400 GPD) YieldMax system with Ammonia Recovery Module (ARM)
- 2. 100 unit PhytoVap evaporator
- 3. A final report including statistical analysis of data collected
- 4. One or more on-site stakeholder outreach meetings and demonstrations to review system performance and address questions

# Work Plan and Methods

Experimental Plan for Tree Health

The fundamental question we want to answer with this project is: "are avocado trees irrigated with treated water as healthy as those that are treated with high quality water". This is an important question as many avocado stands from Ventura to San Diego have high chloride wells on their property. Getting these wells back "online" could provide a supplemental water source that is dramatically cheaper than "imported water", or in some cases, dramatically improve the fruit quality and yield in groves irrigated that must irrigate with existing high chloride water.

Our null and alternative hypotheses are:

Ho- There is no significant difference in NDVI (plant health) between trees irrigated with our treated water(group 1) and trees currently irrigated with the farm's existing clean, low-chloride water (group 2)

## Ha- Treated water from a chloride contaminated well does not maintain tree health

Variables collected weekly for this experiment from both groups of trees are:

- a. Dependent variable- NDVI for each tree
- b. Independent variables
  - i. Flow
  - ii. Treated effluent chloride level
  - iii. Week #
  - iv. Eto
  - v. Treated effluent Ec



Week is a proxy for date. For example week1 would be January 1-January 7. Flow is the total amount of water (gallons) applied for the week. Chloride level is the measured chloride in the treated water. Eto is self explanatory and will be collected from the nearest weather station close to the test area or from the customer if they have it onsite. Treated effluent conductivity (Ec) is helpful as YieldMax removes all the anions in the irrigation water and replaces them with a bicarbonate ion. Before reaching the trees, the bicarbonate may be treated with phosphoric or citric acid (customer will decide) to lower pH and replace bicarbonate with a citrate ion, for example<sup>1</sup>.

The challenge is that we do not know the underlying distribution of the NDVI variable (yet). Assuming the NDVI data appears normally distributed, we will pursue a strategy beginning with simple multiple linear regression. Generally written as:

$$NDVI = \beta 0 + \beta 1(Flow) + \beta 2(Cl -) + \beta 3(week) + \beta 4(Eto) + \beta 5(Ec)$$

Should this model not provide predictive ability, we plan to then try ANCOVA or analysis of covariance, or a mixed effects model (all regression types). If the NDVI data appears not to follow a normal distribution, we can transform the data using log, square root, arcsine, or perhaps Box-Cox. Should all of these fail we can shift to more sophisticated models that are better at handling non-linear data.

Once the dataset is built, we plan to use Python and its libraries to perform all the statistical analyses. There are other more sophisticated models (generalized additive models) that could be better suited for this problem but their complexity is a drawback.

# Soil and Tissue Samples vs NDVI- why the change?

We discussed the basic idea of using conventional tissue and soil samples at length with Fruit Growers Labs to generate our budget. They pointed out that it is <u>very unlikely</u> that we would be able to identify any meaningful differences between leaf analyses over the short term of the project. Soil samples would also be problematic as any repeated irrigation is likely to push soil bound chloride further into the vadose zone. Even if we increased the sample size from 5 to 25, we would probably need to do multiple leaf samples and multiple soil samples for each group. The cost to test the samples (2 sets of 25 trees) every week would have been prohibitive (> \$10,000). NDVI imagery, however, <u>can detect many changes</u> in the health of the trees <u>much sooner</u> than visual indications such as leaf tip burn.

The peer reviewed literature supports the Fruit Grower's Lab staff opinion. Bernstein (1965) showed that visual cues like tip burn may take several weeks or longer to appear as the chloride accumulates in the older leaves first. This is way too slow for us. Along this line of thought, Maas (1986) showed that chloride levels accumulate in leaves weeks before tip burn is visible. Lovatt and Zheng (1996) demonstrated that it can take weeks to months for visual chloride

<sup>&</sup>lt;sup>1</sup> High levels of bicarbonate can bind to soil minerals like calcium making them unavailable.



damage to appear. This last paper clearly suggests that we need a much more sensitive method to detect the effects of chloride-free irrigation on leaf and tree health much sooner. With this in mind and the knowledge that NDVI is already being used to better predict overall avocado yield (<u>CropCount</u>), weekly NDVI imagery is already the right tool for this project.

There are three ways to acquire the weekly NDVI maps:

- 1. Purchase a small agricultural drone (DJI P4 Multispectral). About \$7,000. NDVI software free with purchase. \$0.0 per flight.
- 2. Using a drone service. \$700-\$2000 per flight
- 3. Using Satellite Data + service

The challenge of using satellite data is the standard resolution (10m2) is roughly 3 trees (300 ft2 or 27 m3). Using the 1M or smaller resolution requires more expensive data sources. We need closer to <1 meter resolution, hence a low flying drone equipped with the right imaging camera is the best solution.

The next best solution would be to do monthly drone flights. At \$700 to \$2000 per flight, this becomes prohibitively expensive and does not provide enough granularity (6 data points per tree vs 28 using our own drone) to allow any meaningful analyses.

The simplest approach and best approach would be to purchase the DJI P4 multispectural ag drone (\$7000). The drone allows us to capture images every week. Depending on altitude, we can achieve a 1cm/pixel to 1meter/pixel resolution at altitudes of 60 and 300 meters respectively. This flexibility allows us to have multiple pixels for each tree. Final altitude for these imaging runs will be determined on site.

# Experimental Plan for Water Treatment System

Monitoring the treatment system over the 6 months of operation will provide extremely valuable data that will help inform us, the customer, and the commission if our treatment system produces repeatable results (0 or low Cl-) water. However, the cost of weekly samples with a large array of variables could be prohibitive (\$120 per irrigation suitability analysis x 28=\$3,360). Our plan is to focus on building a Shewart control chart that tracks chloride and Ec over all 28 weeks. These are very easy to do and provide very robust predictive power about the stability and repeatability of our design.

# **Project Outreach**

We plan to communicate to the local growing community by organizing at least 1 onsite visit, with the permission of the co-operator. This site visit will allow local growers to examine the system, study the results (to-date), and ask any questions about how the system can be scaled up and implemented for their growing areas. We could easily move the system to another grower for more piloting in the 2027 year.

We plan to publish an article in the Commission publication called "From the Grove" to



document the system's performance. Part of our communication, in addition to the chloride removal capacity would be a slide show or "movie" showing the vegetation index changes from the start of the treatment to end of the treatment period (Feb-August 2026) based on the drone camera images.

# Milestone Table

| No | What  | Completion | Budget   |
|----|---|------------|----------|
|    | 2024-2025 FY                                |            |          |
| 1  | Initial Site Visit (1 TWS Day)              | 7/1/2025   | \$1,712  |
| 2  | Order Parts for Treatment System            | 08/1/2025  | \$40,410 |
| 3  | Order drone                                 | 8/1/2025   | \$7,000  |
| 4  | Install System                              | 10/30/2025 | \$4,896  |
|    | Subtotal                                    |            | \$54,019 |
|    | 2025-2026 FY                                |            |          |
| 5  | Irrigation starts (chem costs)              | 3/1/2026   | \$1,358  |
| 6  | Weekly samples start                        | 3/1/2026   | \$2,100  |
| 8  | Monthly visits start (½ TWS Day)            | 3/1/2026   | \$8,856  |
| 9  | Out Reach Visit for Growers (1 TWS Day)     | 5/1/2026   | \$1,712  |
| 12 | Equipment tear down and removal (1 TWS Day) | 8/5/2026   | \$1,712  |
| 13 | Final report                                | 9/15/2026  | \$570    |
|    | Sub total                                   |            | \$16,308 |
|    | Grand total                                 |            | \$70,326 |



# Project Budget

# Budget Detail

|    | What                                  | QTY | Unit Cost | Total<br>Cost |
|----|---------------------------------------|-----|-----------|---------------|
| 1  | 5 GPM YieldMax System w/NH3 Recovery  | 1   | \$32,389  | \$32,389      |
| 2  | 100 Bucket PhytoVap System            | 1   | \$6,120   | \$6,120       |
| 5  | Pumping System                        | 1   | \$1,901   | \$1,901       |
| 4  | Purchase Drone                        | 1   | \$7,000   | \$7,000       |
| 7  | Installation (3 full day)             | 3   | \$1,632   | \$4,896       |
|    | Equip subtotal                        |     |           | \$52,306      |
| 6  | Monthly Visit (TWS ½ Day Site)        | 8   | \$1,082   | \$8,856       |
|    | Initial Site Visit (TWS ½ day)        | 1   | 1712      | 1712          |
| 8  | pH Adjust chems (CaO and H3Cit)       | 1   | \$500     | \$500         |
| 9  | Ammonium Bicarbonate powder (50 lb)   | 4   | \$127     | \$508         |
| 10 | Carbon Dioxide (per refill)           | 7   | \$50      | \$350         |
| 11 | Weekly water samples (3 pts)          | 84  | \$25      | \$2,100       |
| 12 | Outreach visit on-site (TWS full day) | 1   | \$1,712   | \$1,712       |
| 13 | Final Report                          | 8   | \$90      | \$570         |
|    | Equipment tear down                   | 1   | 1712      | 1712          |
| 14 | Other costs                           |     |           | \$16,308      |
|    | Grand Total                           |     |           | \$70,326      |



# Budget Narrative (1 page)

- The major change in our budget is driven by the purchase of the drone. Our initial ideas
  of sampling tissue and soil proved unworkable and extremely expensive: > \$10,000.
  The drone did increase our capital requirements but eliminated the extensive testing
  costs that would have been required.
- Site Visits. We will use the IRS 2025 mileage rate for this project (\$0.70/mile ). GSA M&I rates for Ventura county are \$65/person per day, Lodging is \$191/day per person for Ventura County. Daily lodging and meals cost per person is \$256.
- 3. Note that our site visits will always use 2 people for safety reasons. First, we require two people on site when handling chemicals or operating equipment. Second, since two of our contractors are women, we do not send them to remote sites alone.
- 4. Staff costs. Tailwater is family owned and operated. Family members are paid as contractors. The standard rate is \$90/hour for technical staff and \$120/hr for senior technical staff. For this project we are discounting these rates to \$50/hr for technical stall and \$90/hr for senior technical staff or \$140/hr blended rate.
- 5. Full Day Visit(2 people). A one full day site visit with 2 staff is \$1120 for labor and \$512 for lodging and meals for a total of \$1632 + \$392 for mileage.
- 6. Half day visit (2 people). \$392 for mileage, \$560 for labor, and \$130 for meals and incidental expense. Total is \$1082.00 per visit.
- 7. Bicarbonate expense is very conservative. If NH3 recovery works well at the site, this cost will be 85% lower.
- 8. CO2 will be 50lb cylinder delivered once a month.
- 9. Monthly visits. These are priced as a "Half day visit". During these visits our team, checks the system, swaps out the CO2 cylinder, refills any chemical supplies, does maintenance on the PhytoVap array- grass has to be trimmed monthly. Our staff also collects data from the several embedded dataloggers in our PhytoVap and YieldMax systems and the drone.
- 10. All equipment purchases will be done in the commission's 2024-2025 budget year. The vast majority of personnel and other expenses occur in the commission 2025-2026 budget year.
- 11. Pumping system. This is a high pressure irrigation pump along with piping and fittings that is used to send our treated water to the trees we are evaluating. The trees are isolated from the existing irrigation system so that they only receive water from our system. Cam Lam will provide nutrient solutions to be mixed with our treated water.
- Weekly water samples. Cam Lam staff will be collecting 3 x 10ml water samples every week and mailing them back to Tailwater for analysis. Our staff owns a Hach 1900 spectrometer. Chloride will be tested using the Hach standard TnTplus kit. Our TAT is < 24 hours after receipt. Price includes FedEx freight, cost of test kit, and our labor.



# References

- 1. Bernstein, L. (1965). *Salt Tolerance of Avocado Trees: Effect of Chloride and Sodium on Growth and Leaf Burn. Agronomy Journal*, **57**(6), 381-385.
- Goto, K., Goto, T., Nmor, J.C. et al. Evaluating salinity damage to crops through satellite data analysis: application to typhoon affected areas of southern Japan. Nat Hazards 75, 2815–2828 (2015). <u>https://doi.org/10.1007/s11069-014-1465-0</u>
- 3. Lovatt, C. J., & Zheng, Y. (1996). *Strategies to Minimize the Impact of Salinity Stress on Avocado Trees. California Avocado Society Yearbook*, **80**, 99-106.





March 10, 2025

Tailwater Systems 3855 Via Nona Marie, Suite 205 Carmel, Ca 93232

RE: Avocado Commission Grant Support for 2025-2026

Dear John,

Camlam Farms, Inc. will support the installation and operation of your chloride removal system on our ranch in Camarillo, CA during the 2025-2026 grant period.

We agree to provide the following:

- a. 120VAC service to power your system
- b. A source of "high chloride water"
- c. Permission to use drones, as required, to overfly the test area
- d. Permission for you and your staff to come to our property during the grant period
- e. A line of avocado trees to receive the chloride-free water
- f. Nutrient mix for the trees receiving the chloride free water.
- g. Collecting and sending water samples via Fed Ex weekly
- h. Host at least one site visit by other grower/members of the Commission as part of the outreach requirement.

Tailwater's responsibilities include:

- a. Will contact Camlam at least 24 hours before any visit to the property
- b. Remove all equipment from our property at the end of the test period
- c. Do not disclose any information about our site, staff, or methods without our written permission.

Any other local services required to assist in the project will be negotiated between our two companies.

Sincerely,

John B. Lamb President