

ALMOND BOARD OF CALIFORNIA

# RESEARCH UPDATE 2024



## **Advancing California Almonds Through Innovation**

Funded by grower assessments, Almond Board-funded research adds value to the industry by:

- Provide industry members with information & resources on options to maximize their efficiency, profitability and value for their operations
- Stimulate higher value and diversified almond co-product market development
- Drive global demand for California almonds
  - Strengthening our health halo for almonds
  - Amplifying California almond's environmental stewardship story
  - Marshall research to stimulate new uses of almonds

This report summarizes current research on almond production, environmental aspects of production, human nutrition, co-product biomass, almond quality, and food safety that supports these goals and objectives.

This research is directed by over 60 industry stakeholders who share their expertise through service on ABC committees and working groups:

- Strategic Agricultural Innovation Committee
  - Biomass Work Group
  - Production Stewardship Work Group
- Nutrition Research Committee
- Almond Quality and Food Safety Committee

These volunteers ensure research targets industry priorities and deliver practical and relevant information. We are grateful to the committee members for sharing their time and experience and invite others in the industry to consider service on Almond Board committees.

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# Table Of Contents

## Nutrition Research

Can almond nut consumption improve nocturnal glycemic control in women with gestational `diabetes mellitus?	9
Effects of almonds on glycemia in adults with elevated HbA1c concentrations	10
Effects of almonds in glucose-intolerant adults: a randomized controlled study on muscle mass and obesity, energy metabolism and lipidome, non-alcoholic fatty liver and inflammation	11
Almonds to improve gut health and decrease inflammation in metabolic syndrome	12
Effects of almond consumption on the gastrointestinal microbiota and postprandial glucose handling in adults with overweight and obesity	13
Effects of Whole Almonds on Immune Health and Responsiveness in Adults with Obesity	14
Effects of almond snacking on gastrointestinal health in adults with overweight or obesity	15
Almonds as a source of healthy fats: in vivo synthesis of gut microbiota-produced cyclopropane fatty acids following almond feeding	16
Effect of almond supplementation on gut health and glycemic control in adults with prediabetes in rural settings of Karnataka, India: impact assessment in a cluster parallel randomized trial	17
A randomized, comparator-controlled parallel study to investigate the dose response of an almond-enriched diet on lipid levels in a population with hypercholesterolemia	18
Effect of daily consumption of almonds on immune strength and response to flu vaccination in overweight middle-aged men: a randomized controlled study	19
Effects of almond consumption on innate myeloid and lymphoid cell composition and activity	20

Exploring the role of almonds in enhancing immune strength	21
Almonds and their impact on immune optimization to viral infection: a randomized controlled trial of vaccination model of immune response	22
Evaluating Almond Protein for Human Health Benefits	23
The Effects of Almond Consumption on Functional Performance Aerobic Capacity and Physical Activity in Overweight and Obese Active Older Adults	24
The Effect of Almond Consumption on Skin Collagen and Elastin in Women	25
Almond Supplementation in Mild to Moderate Acne Vulgaris	26
Effect of Almond Supplementation on Mild to Moderate Acne Vulgaris in a community living population in India	27
Effects of Almond Consumption on Quality of Sleep in Adults: A Randomized Controlled Trial	28
Almonds and the gut-brain axis: a randomized controlled trial to improve mental health, psychological distress and quality of life	29
Randomized controlled trial of almond supplementation vs isocaloric diet on cognitive functions in middle-aged (40-60 years) Asian Indians with Prediabetes	30
Almond Consumption and the Risk of Cancer Incidence and Mortality in a Population with a Wide Range of Intake	31
Deep Metabolomic Profiling of Almonds	32
<b>Almond Quality Food Safety Services</b>	
Effect of Water Activity, Temperature and Incubation period on Fungal Growth and Aflatoxin Production on Almond nuts	33
Full Demonstration of SmartProbe Technology for Early Detection of Insect Pests and Environmental Monitoring	34

## **Production Research**

Investigation CELF-Pretreated Almond Tree Waste for Valorization into High Value Co-Products to Curtail Tipping Charges to the Farmers	35
Allergenicity and Risk Assessment of Novel Almond Hull Food Ingredients	36
Cost Return Studies for California Almonds	37
Irrigation Management: Evaluating Current Sensor-Based Products and Remotely Sensed Information and Testing Thresholds for Delaying the Start of Irrigation in the Spring	38
Updating Information on Evapotranspiration (ET) and Crop Coefficients (Kc) of Micro-Irrigated Almond Production Orchards Grown in California for use in Water Resource Management and Irrigation Scheduling Decisions	39
Yield Prediction for Resource Management and Yield Optimization in Almond	40
Data-driven Smart Irrigation for Almond Orchards	41
Immobilizing Soil Nitrate Using Almond Shells in Winter-Fallow Vegetable Fields to Reduce Nitrate Leaching	42
Optimizing Potassium Management in Almond	43
Evaluating HFLC Nitrogen Management Strategies to Minimize Reactive Nitrogen Mobilization from California Almond Orchards	44
Optimizing Nitrogen and Water use Efficiency in Replanted Orchards After Whole Orchard Recycling	45
Improving Non-Fumigant-Based Approaches For Management of Almond Replant Problems	46
Effect of Partial Substitution of Fertilizer with Organic Matter Amendments on Nutrient Cycling	47

Cross-Project Approach to Accelerate Multidisciplinary Research on the Interaction of Nonstructural Carbohydrates (NSC) With Biotic and Abiotic Stresses Management Practices and Varieties in Assessing NSC's Dynamics Impact on Yield - an Ongoing Carbohydrate Observatory Project.	48
How to Irrigate Almond Orchards - for the Current Year's Expected Yield or for Maximum Yield Potential?	49
Determining Almond Tree Water Use and Stress using Surface Energy Balance Models with Unmanned Aircraft Systems	50
Remote-Controlled Evaluation of Distribution Uniformity and Stem Water Potential: Extending Imagery to Integrated Decision Support	51
Determining the Sensitivity of Fruit (nut) Set and Crop Load to Early Season Water and Carbohydrate Status, Developing a Water/Carbon Model for Fruit Set, and Evaluating Whether Low Set and Resulting Low Crop Loads Require Less Than Full In-Season Irrigation	52
Almond Variety Development	53
Regional Field Evaluation of New Almond Varieties & Selections – 3rd Generation	54
Regional Field Evaluation of New Almond Varieties & Selections – 4th Generation	55
Almond Culture and Orchard Management	56
Field Evaluation of Almond Rootstocks in the Southern San Joaquin Valley	57
Nickels Soil Lab Projects	58
Three-Dimensional Model-Based Analysis of the Impact of Variability in Almond Tree Structure and Configuration	59
The Application of Molecular Tools and Quantitative Phenotyping for Genomics-Assisted Breeding in Almond	60



Are Californian Almond Cultivars and Rootstocks Susceptible to PPV and Can Almonds be a Host for the Spread of Sharka in California?	61
Discovery of Genetic Variation in Related Self-Fertile Species of Almond	62
Field Screening of Size Controlling Rootstocks for Off-Ground Harvested Almond Orchards	63
Accelerated Assessment of Almond Variety Candidates	64
Evaluating New Breeding Material for Salinity Tolerance in Almond Rootstocks and Exploring Novel Sources of Salinity Tolerance in Prunus Germplasm	65
Resilient Prunus Rootstocks for a Changing Climate	66
Comparing Root Traits and Depth Distributions for Mature Almond Rootstocks; is There a Link Between Root Architecture and Propagation Method?	67
Improving Fruit Removal and Harvest Efficiency of Independence Almond Cultivar	68
Multi-Scale Evaluation of Stacked Regenerative Practices in Almond Systems	69
Avian Biodiversity in Almond Orchards of Central California	70
Effects of Almond Adjuvants and Phytochemicals on the Synergistic Toxicity of Fungicides and Insecticides on Honey Bees	71
Another Look at Pheromonal or Related Attractants for Leaffooted Bugs ( <i>Leptoglossus</i> spp.) Infesting California Nut Crops	72
Evaluating the Influence of Landscape Composition on Almond Orchard Susceptibility to Leaffooted Bug ( <i>Coreidae: Leptoglossus zonatus</i> ) Colonization in the Spring	73
Revisiting Trap Selection for Pheromone- Based Monitoring of Leaffooted Bugs ( <i>Leptoglossus</i> spp.) Infesting California Nut Crops	74

Biology, Monitoring, and Management of Native and Invasive Stink Bugs in Almond Orchards	75
Evaluation of Reduced-Risk Hull Split Sprays for Navel Orangeworm	76
Spatiotemporal Models to Evaluate the Potential Value of Sterile Insect Technique for Control of Navel Orangeworm	77
Initiation of First Stage Product Development of a Friendly Navel Orangeworm for Californian Almond Growers	78
Ecology, Monitoring and Management of Carpophilus Beetle	79
Implementing a Nematode Management System for Almond Using Chemical and Biological Treatments	80
Weed Research and Extension to Address Almond Grower and Industry Management and Sustainability Goals	81
Region-Wide, Disease Risk Forecasting System	82
Biology and Management of Almond Brown Rot Jacket Rot Shot Hole and Hull Rot	83
Investigation of Aspergillus Niger Causing Hull Rot and Conditions Conducive to Disease Development in Kern County	84
Epidemiology and Management of Phytophthora Root and Crown Rot of Almond	85
Epidemiology and Management of Almond Band Canker Disease in Young Orchards	86
Improve the Detection and Risk Prediction of Pseudomonas Syringae Causing Bacterial Blast and Bacterial Canker of Almond in California	87
Assessing Almond Rootstock Susceptibility to Ganoderma Wood Decay and Crown Gall	88
Assessing Nematode Control Soil Health and Tree Vigor in a Commercial Almond Orchard Four Years After Soil Biosolarization	89

# Can Almond Nut Consumption Improve Nocturnal Glycemic Control in Women with Gestational Diabetes Mellitus?

PROJECT NO: BGR-22-Flynn-NR-01

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**Summary**

Fasting hyperglycemia is a common challenge in managing gestational diabetes mellitus (GDM) and is thought to be linked to elevated nighttime blood sugar levels. Women with GDM are more likely to snack at night compared to those with normal glucose tolerance, and bedtime snacking has been associated with higher fasting blood sugar levels. However, the impact of snack type on nighttime blood sugar regulation remains unclear. The primary aim of this study is to conduct a randomised controlled trial to assess the impact of consuming whole almonds as an evening snack, compared to a nut-free snack, on nighttime blood sugar levels in women with GDM. The secondary aim is to evaluate how almond intake influences various metabolic markers, including fatty acids, amino acids, glycolysis-related metabolites, ketone bodies, and inflammatory markers in this population. As of September 5, 2024, we have received ethical approval and will begin recruitment following the capacity and capability (C&C) review.

# Effects of Almonds on Glycemia in Adults with Elevated HbA1c Concentrations

PROJECT NO: DMS-21-Mattes-NR-01

**Principal Investigator:**

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## Summary

The purpose of this study was to examine the potential for sustained almond consumption to reduce HbA1c concentrations among individuals with elevated values. A 16-week randomized, parallel-arm, controlled trial was conducted. Eighty-one adults with elevated HbA1c concentrations (>5.7%) were randomly assigned to incorporate 2 oz of raw almonds (A: n=39) or energy-matched snacks (C: n=42) into their daily diets. Body weight, body composition, plasma lipids, HbA1c, plasma vitamin E, glycemia (by meal tolerance test and continuous glucose monitoring), dietary intake, and hedonic responses to test foods were measured at stipulated time points. Participants consuming almonds ingested 253 kcal/d more than participants in the control group (P=0.02), but this did not result in a significant difference in body weight. No statistically significant differences were observed in HbA1c concentrations, blood chemistries, body composition, or glycemia over time or between groups. However, Healthy Eating Index scores improved within the almond group as compared to the control group (P<0.001). Additionally, the hedonic rating of almonds within the almond group did not decline as markedly as the control group's reduced liking of the pretzel snack. Alpha-tocopherol increased significantly, and gamma tocopherol tended to decrease in the almond group, indicating compliance with the dietary intervention. Overall, daily ingestion of 2oz of raw almonds in a self-selected diet for 16 weeks did not alter short-term or longer-term glycemia or HbA1c concentrations in adults with elevated HbA1c concentrations, but they were well-tolerated hedonically and improved diet quality without promoting weight gain.

# Effects of Almonds in Glucose-Intolerant Adults: a Randomized Controlled Study on Muscle Mass and Obesity, Energy Metabolism and Lipidome, Non-Alcoholic Fatty Liver and Inflammation

PROJECT NO: DMS-21-Kabisch-NR-01

## Principal Investigator:

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## Summary

Tree nuts – such as almonds – contribute to beneficial effects of the Mediterranean diet on risk for cardiovascular events, type 2 diabetes, dyslipidemia, hypertension, inflammation and non-alcoholic fatty liver disease. Almonds provide few carbohydrates, but lots of unsaturated fat and dietary fiber and may therefore have a significant impact on metabolic health. But to which extent and by which mechanisms may almonds improve all individual aspects of the Metabolic Syndrome, in particular glucose tolerance, liver fat and subclinical inflammation? Previous clinical trials showed weaker effects in humans compared to rodent studies, most possibly due to low statistical power and metabolically insusceptible patients. The Mediterranean diet is particularly effective in patients with obesity and poor overall lifestyle, while normal weight persons might benefit to a smaller extent. The 3-year AGAMEMNON project aims to investigate if 16 weeks of supplementation with almonds (vs. no treatment) in 150 patients with prediabetes and NAFLD leads to significant improvements in glycemia and liver fat, lipid metabolism, body composition and inflammation. The study is designed as an open-label two-arm single-blinded parallel randomised intervention study. The cohort structure is chosen in order to select a highly responsive group of patients with low confounding by medication. The isocaloric design will outrule effects of weight loss and will allow the analysis of metabolic pathways between fat depots, inflammation, insulin resistance and gut function. Oral glucose tolerance tests are facilitated to determine glycemic conditions during the study. State-of-the-art MR assessment will be used to measure body fat distribution. A psychometric battery will clarify the impact of behavior on initial metabolic state and responsiveness. Lipidomics will serve as novel predictors of progression and metabolic response. Study recruitment is currently underway with the study expected to be fully enrolled by 2026.

# Almonds to Improve Gut Health and Decrease Inflammation in Metabolic Syndrome

PROJECT NO: GUTH01

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## **Summary**

Metabolic syndrome afflicts ~35% of Americans and affects over a billion people world-wide. Metabolic syndrome is a group of conditions that together increase the risk of developing type 2 diabetes, heart disease, stroke, and fatty liver disease. The conditions associated with metabolic syndrome are high blood sugar, high blood pressure, high blood triglycerides, low levels of good cholesterol, and excess body fat around the waist. There is a critical need to develop strategies to stop these metabolic issues and prevent further health deterioration. Gut health and chronic inflammation are key factors in the development of metabolic syndrome that can be targeted with dietary approaches. Almonds, as a healthy snack, are easily incorporated into a diet, and almonds are a rich source of antioxidant and anti-inflammatory compounds that may significantly improve gut health and limit inflammation in people with metabolic syndrome. The beneficial compounds in almonds include vitamin E, polyphenols, healthy fats, fiber and minerals. Many of the US population, especially those with metabolic syndrome, are at risk for deficiencies of these micronutrients and dietary factors. The hypothesis tested was that daily almond consumption will improve vitamin E status, gut health, cardiometabolic health outcomes, and decreased inflammation and oxidative stress in persons with metabolic syndrome. Participants with metabolic syndrome were randomly assigned to an almond (n=38) or control intervention group (n=39). Participants consumed either 2 oz. of whole dry roasted almonds, or non-whole grain crackers (of equal caloric content) daily for 12 weeks. The crackers acted as a control food that lacks bioactive compounds such as fiber, healthy fats, polyphenols or micronutrients found in almonds. Our results show that almond consumption decreased total cholesterol, “bad” cholesterol (LDL cholesterol), and a modest but significant improvement in waist circumference (0.8 cm at week 4) relative to the cracker control group. Almond consumption also improved biomarkers of gut health in participants that exhibited elevated inflammation in their gut at the beginning of the study. Almond snacking was not associated with significant changes in participant’s weight, calorie intake, systolic or diastolic blood pressure, fasting glucose, or insulin levels, relative to the cracker control group. An improvement in vitamin E status was observed in response to almond consumption. Diet records also indicate almond snacking was associated with significant increases in dietary soluble fiber, copper, biotin, magnesium, riboflavin, and healthy fats (polyunsaturated fatty acids and monounsaturated fatty acids). Participants in the almond treatment group recorded these improvements in their diet at both the 4 and 12 week study visits, compared to baseline and the cracker control group. The improvement in gut health and vitamin E status did not cause significant changes in systemic plasma levels of inflammation or oxidative stress. Together, these results are significant because they support the benefit of almond snacking in persons with metabolic syndrome and provide an evidence base for dietary recommendations to increase the consumption of almonds to promote health.

# Effects of almond consumption on the gastrointestinal microbiota and postprandial glucose handling in adults with overweight and obesity

PROJECT NO: 19-HolscherH-NR-01

## Principal Investigator:

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## Summary

We completed a 12-week, randomized controlled trial to test our hypothesis that consuming 2.0 oz roasted almonds daily would help support gut and metabolic health in adults with overweight and obesity. Our primary objective was to determine the effect of almond consumption on gut health, focusing on increasing *Roseburia* spp. abundances and butyrate concentrations, and decreased microbially-derived secondary bile acids. We anticipated that supporting gut health would positively impact metabolic health by reducing inflammation and body fat and improving glycemic control. Approximately 500 people responded to recruitment advertisements expressing interest in the study. 40 individuals were deemed eligible to participate in the study, and 33 individuals elected to begin the study. Of the 33 individuals that started the study, 25 completed the 12-week intervention. Our results revealed that almond consumption did not enrich *Roseburia* or fecal butyrate concentrations. Subjective assessments of gastrointestinal health, including intolerance symptoms such as bloating and flatulence, were similar between groups. There were no differences in stool consistency between groups. There were no differences in measures of glycemic control or inflammation. Adiposity, bile acid and fecal metagenomic analyses are ongoing. Contrary to our hypothesis, there was no enrichment in fecal *Roseburia* concentration or short-chain fatty acids.

# Effects of Whole Almonds on Immune Health and Responsiveness in Adults with Obesity

PROJECT NO: ECP-Dhillon-NR-001

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**Co-PIs:** Victoria J. Vieira-Potter, Jeffery Woods

## Summary

This study was designed to explore the health advantages of consuming whole almonds compared to an isocaloric cookie snack over a span of six weeks, focusing on immune health indicators in adults with obesity. Participants in the almond group are provided with 57g (320 kcal) of whole almonds per day, while the participants in the control group are provided with an isocaloric cookie snack. We compared the effects of almonds versus control on immune health and inflammatory biomarkers in blood and adipose tissue, the cell-mediated immune response, and the gene expression of metabolites involved in immune health in adults with obesity (BMI: 30-45 kg/m<sup>2</sup>). In the first two years, the project achieved significant milestones, including obtaining IRB approval, assembling a full research team, and completing participant recruitment, dietary interventions, and data collection. To date, 60 participants have successfully completed the study, and 4 are still active, expected to finish in September 2024. Preliminary findings were presented at the American Society of Nutrition Conference in 2024. We anticipate publishing three manuscripts based on the project's specific aims, focusing on almond effects on 1) blood and adipose immune health markers, 2) cell-mediated immune response, and 3) gene expression of immune markers in obesity.



# Effects of Almond Snacking on Gastrointestinal Health in Adults with Overweight or Obesity

PROJECT NO:ECP-Nagpal-NR-001

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## Summary

The overarching objective of our project was to determine the extent to which almond intake (42.5 g/d) modulates gut permeability, intestinal/systemic inflammation, gut-derived hormones, and gut microbiome-metabolomic arrays in adults who have overweight or obesity. A secondary objective was to assess relationships between gut permeability, inflammatory markers, gut microbiomes, gut/plasma metabolites, hormones, bile acids, and measures of cardiometabolic health in adults with overweight or obesity consuming 42.5 g/d of almonds. The project involved ancillary analyses of previously completed ABC-funded studies conducted at Penn State University that showed that the almond snacking, compared to an isocaloric high-carbohydrate snack, for 4-weeks improves plasma lipid profiles in individuals who have overweight or obesity.

Dietary snacks contribute to over 20% of our total daily energy intake. Therefore, snack selection has an appreciable influence on our nutrition, weight status, and gut and cardiometabolic health. Heart disease is one of the leading causes of mortality worldwide and is associated with poor nutrition, obesity, and gut microbiome dysbiosis. Nut consumption has been correlated inversely with hypercholesterolemia, coronary heart disease, and mortality. Almonds are rich in dietary fiber, unsaturated fatty acids, and polyphenols and are known to modulate the gut microbiome in healthy subjects. However, whether and how almond snacking regulates intestinal and cardiometabolic health features in subjects with overweight and obesity, that are otherwise more predisposed to gut dysbiosis and cardiovascular disorders, remains unknown. To this end, ours is the first multi-omics study to comprehensively assess the effect of almond snacking on gut and cardiometabolic health in adults with overweight and obesity. With an overarching aim to specifically evaluate the role of almond snacking in mitigating the impairments caused by the average American diet, we examined a wide array of health features including gut microbiome, gut-blood metabolome, bile acids, intestinal epithelial integrity, inflammatory arrays, and blood lipid profiles. We found that almond snacking increases beneficial and functional gut microbes, including *Faecalibacterium prausnitzii* and suppresses opportunistic pathogens such as *Collinsella aerofaciens*, thereby favorably modulating the gut microecological niches through symbiotic and microbe-metabolite interactions. Moreover, almond snacking elevates the intestinal availability of health-beneficial monosaccharides and fosters bacterial consumption of amino acids, owing to enhanced microbial homeostasis. Additionally, almond intake enhances metabolic homeostasis by inducing a ketosis-like effect and leads to lowered inflammation while improving satiety-regulating hormones associated with anti-obesity effects. The findings demonstrate that even small but prudent dietary/lifestyle choices, such as including almonds as snacks into a typical American diet without exceeding energy needs, may be a simple strategy to foster a healthier gut microbiome homeostasis thereby ameliorating intestinal and cardiometabolic health. The study shall inspire and facilitate prospective studies aimed at elucidating the health benefits of almonds.

# Almonds as a Source of Healthy Fats: in Vivo Synthesis of Gut Microbiota-Produced Cyclopropane Fatty Acids Following Almond Feeding

PROJECT NO: GH-22-DebedatJ-NR-01

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**Summary**

This study is investigating how eating whole almonds may benefit health by exploring how the fats in almonds interact with our gut bacteria. Almonds are rich in a type of “healthy” fat known as monounsaturated fatty acids (MUFAs), and we believe that these fats can be transformed by gut bacteria into unique fat molecules called cyclopropane fatty acids (CpFAs). Based on results in cell culture studies showing activities of CpFAs in signaling pathways associated with support of metabolic health, our hypothesis is that these CpFAs may play a role in improving metabolism in the body following almond consumption.

After a week of following a diet low in CpFAs, participants were asked to consume either whole almonds or a similar snack low in MUFAs. We collected blood and urine samples over an 8-hour period and stool samples before and after the feeding day to track CpFA production. Participants also completed detailed dietary questionnaires to help us understand how different eating habits might affect CpFA levels. Additionally, we have analyzed the gut bacteria in each participant to determine whether specific bacterial species are associated with higher CpFA production. All participants have now successfully completed the study, and we are in the process of analyzing the data. Specialized chemical analysis to determine levels of CpFA in the blood and stool are almost complete, and will be linked to gut bacteria data and to almond intake. The results of this study will help us understand how almonds, gut bacteria, and diet together influence metabolism and contribute to overall metabolic health.

# Effect of Almond Supplementation on Gut Health and Glycemic Control in Adults with Prediabetes in Rural Settings of Karnataka, India: Impact Assessment in a Cluster Parallel Randomized Trial

PROJECT NO:ECP-VAIDYA-NR-001

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## Summary

This study aims to observe the effect of almond consumption for 16 weeks on blood glucose levels and gut health in rural Indian adults with prediabetes, compared to traditional snacks like cereal and pulses. It took place in six villages in Karnataka, involving 152 overweight adults with prediabetes who weren't taking any diabetes medication. The research is registered in India's clinical trial database. So far, all 152 participants have completed the almond intervention of the study. Blood samples were taken at the start, after 8 weeks, and after 16 weeks to see how almond consumption affects their health. Stool samples from 63 participants were also collected to understand the impact of almonds on gut health and beneficial bacteria. The data is currently being analyzed. The study faced challenges, such as cultural differences and gaining trust in the community, but the team overcame these hurdles and achieved 97% adherence to the almond diet. To give back to the community, the researchers also started a diabetes awareness program focused on self-care and managing diabetes complications.

A paper on the study's protocol was published in BMJ Open this year. Interim results were shared at international conferences, where the study won two "Best Scientific Paper" awards. Early findings show that about 30% of the community has prediabetes, with a diet high in carbohydrates but low in protein and fiber. In a sub group of participants, almond consumption has shown a positive trend in increasing beneficial gut bacteria, which helps regulate blood sugar and cholesterol levels. Final results will be available next year.

# A Randomized, Comparator-Controlled Parallel Study to Investigate the Dose Response of an Almond-Enriched Diet on Lipid Levels in a Population with Hypercholesterolemia

PROJECT NO: HH-21-CROWLEY-NR-01

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## Summary

Modifications to dietary habits are recommended for individuals with higher lipid levels to lower heart disease risk. While almond consumption has been shown to improve some risk factors for heart disease, the effects of almonds on high-density lipoprotein (HDL) cholesterol, also known as “good cholesterol”, are less clear. The objective of this study was to determine the amount of almonds that would provide the best increase in HDL cholesterol in adults with high cholesterol. This randomized, comparator-controlled study consisted of a 16-week period in which participants were randomized to one of three groups (25 per group) including high dose almonds (2.5 oz/day), low dose almonds (1.5 oz/day) or nut-free diet (cookies with the same caloric content as the 1.5 oz/day). Two almond amounts were selected based on existing scientific evidence for their effects on cholesterol levels. Blood lipids and other blood markers (apolipoproteins), heart disease risk estimate (Framingham Cardiovascular Disease Risk Score), anthropometric measures (weight, body mass index, waist circumference, sagittal abdominal diameter and waist:hip ratio), blood pressure and blood concentrations of vitamin E were assessed.

Participants who consumed the high or low dose of almonds had a decrease in their estimated risk for developing heart disease after 16 weeks compared to their baseline score despite no increases in HDL cholesterol. Further, systolic blood pressure significantly decreased after 8 weeks for the high dose almond group compared to the comparator, who consumed cookies. The participants in the comparator group had an increase in sagittal abdominal diameter, a marker of abdominal obesity, after 4 weeks compared to baseline which did not occur for those in the almond groups. Findings from this study suggest almond consumption may support dietary strategies for improving CVD risk factors in adults with hypercholesterolemia.

# Effect of Daily Consumption of Almonds on Immune Strength and Response to Flu Vaccination in Overweight Middle-Aged Men: a Randomized Controlled Study

PROJECT NO: IH-21-SabateJ-NR-01

## Principal Investigator:

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## Summary

Nutrition and diet plays an enormous role on the immune system. It is now recommended and might become essential to have an adequate intake of immune-boosting nutrients to strengthen the immune system throughout the life-cycle. Recent evidence suggests that plant foods that are also minimally processed may provide a wide range of immune enhancing nutrients and can be a useful strategy for individuals that seek to improve their immunity and body's defense mechanism against pathogens. Given that plant foods such as almonds have a plethora of nutrients that individually and in synergy provide several health benefits they may have a role to play in enhancing immune functions. Almonds are a good source of several immune-enhancing micronutrients like fiber, zinc, copper, vitamin E, magnesium, selenium, folate, and antioxidant polyphenols, and when regularly consumed as a snack, are likely to enhance immunity and reduce the risk of upper respiratory tract infections and symptoms such as those associated with the seasonal flu. This study is working to determine if almond consumption in overweight men and post-menopausal women enhances immunity. This will be assessed by examining changes in lymphocyte populations and subsets such as T cells, T cytotoxic (Tc) cells, ratio of Th to Tc cells, T helper cells, B cells, natural killer (NK) cells, regulatory T cells (Treg) and the naive and memory Treg cells. Since total lymphocyte count is a measure of nutritional status, changes in lymphocyte populations will provide important information on prior nutritional status of the subjects and on nutritional improvements as a result of almond consumption. Inflammatory biomarkers (CRP, IL-1 $\beta$ , IL-6, IL-10, TNF- $\alpha$ , IFN- $\gamma$ , MCP1, Eotaxin, E-selectin, RANTES and Pentraxin-3) involved in low-grade, systemic inflammation will be assayed in this study. The study also includes an assessment of the effects of almond consumption on antibody and immune response to influenza vaccination. Data collection is finished for all 61 participants and final analyses are being completed and summarized into a manuscript for peer-reviewed publication.

# Effects of Almond Consumption on Innate Myeloid and Lymphoid Cell Composition and Activity

PROJECT NO: IH-21-BULLOM-NR-01

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## Summary

Nutrition is known to affect the immune system and modulates resistance to infection, making adequate and appropriate nutrition essential for bolstering immune defenses and enhancing pathogen resistance. The AlmondIS study aimed to evaluate the effect of regular almond consumption on innate and adaptive immune system in healthy individuals with overweight or obesity who follow a Western-style diet and consume unhealthy snacks. Additionally, the study sought to determine whether changes in miRNAs mediate the influence of almonds on the activity of innate lymphoid cells and other indicators of immune function in both the innate and adaptive immune systems. The first participant was randomized 20th September 2021, and the last participant's visits was conducted 2nd May 2024. No significant differences in anthropometric and circulating inflammatory markers were observed between the intervention groups. Preliminary results indicate a significant increase of monocytes (CD14+ CD16-) in the almond group compared to controls. Since these type of cells can differentiate into macrophages and dendritic cells, which play crucial roles in pathogen clearance and the initiation of adaptive immune responses, this could be interpreted as an advantageous initial immune response to infections. On the other hand, increased CD14+CD16+ monocytes typically indicate a state of chronic inflammation and reflect immune system activation in response to ongoing inflammatory stimuli. Elevated levels of these monocytes are often associated with chronic diseases and metabolic disorders. Therefore, the reduction in CD14+CD16+ monocytes observed following the almond intervention could be interpreted as a potential mechanism for improving metabolic status and reducing inflammation. This suggests that almond consumption might contribute to better metabolic health by modulating inflammatory responses and potentially mitigating chronic inflammation. Since the context in which changes in immune cells occur is crucial for interpreting their significance and implications for health, incorporating in vitro PBMC cytokine production data will offer a better comprehensive view of immune function. Finally, incorporating miRNA information into the analysis will help to elucidate the molecular pathways through which almonds affect immune responses, offering insights into the regulation of inflammation and immune homeostasis.

# Exploring the Role of Almonds in Enhancing Immune Strength

PROJECT NO: IH-21-KERNM-NR-02

## **Principal Investigator:**

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## **Summary**

The COVID-19 pandemic highlighted the importance of good nutritional status for immune strength and for reducing comorbidities that increase morbidity and mortality of infections. Almonds likely confer numerous immune strength advantages over common snacks; however, little empirical research is available. We aimed to determine the impacts of almonds on immune health. We conducted an 8-week parallel-arm trial of 48 overweight/non-obese (BMI=24-30) adult (aged 40-65 years) men (n=24) and women (n=24) tested to be free of almond allergy/sensitivity who were randomly assigned to consume daily isoenergetic portions of almonds (15-20% of energy intake) or a common snack (pretzels). Almonds promoted higher intake of PUFAs, MUFAs, fiber, vitamin E, calcium, magnesium, phosphorus, and potassium. No effects on complete blood count differentials, proliferation of peripheral blood mononuclear cells or cell-mediated cytokine production were detected. Furthermore, the percentage of T lymphocytes, B lymphocytes, CD4+ T cells and subpopulations, CD8+ T cells and subpopulations, and Natural Killer T cells (NKT) and NK cells in the blood were similar; however, a time by trial interaction ( $p < .05$ ) was observed for CD8+ T effector memory (TEM) cells and CD8+ naïve cells. No major difference in blood lipid profiles, blood pressure, plasma cytokines, markers of inflammation or endothelial function were detected. In conclusion, almonds promoted consumption of key nutrients, but exerted little impact on immune function or cardiovascular risk factors of apparently healthy individuals who were at the upper end of normal weight through overweight.

# Almonds and Their Impact on Immune Optimization to Viral Infection: a Randomized Controlled Trial of Vaccination Model of Immune Response

PROJECT NO: IH-21-DIMIDIE-NR-01

## **Principal Investigator:**

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**Co-PIs:** Sarah Berry, Wendy Hall, Esperanza Perucha, Victor Turcanu, Kevin Whelan

## **Summary**

**Background:** Almonds have the potential to beneficially impact immune function. They are high in immunoregulatory nutrients (e.g. vitamin E and zinc). They may also influence the gut microbiota and their metabolites, which are known to interact with the immune system. Moreover, as a healthful snack option high in fibre and micronutrients, they can positively alter an individual's dietary intake, and in turn immune function. In order to study the impact of dietary interventions on the immune response, a vaccination model of viral infection is widely used. For example, studies exist assessing the impact of specific immunoregulatory nutrients (e.g. selenium and zinc), prebiotic and probiotic supplements on the immune response following influenza vaccination; however, currently, there are no published studies in this area administering a whole food that has the potential to impact immune function, such as almonds.

**Objective:** This project is a parallel group, randomized controlled trial (RCT) that aims to investigate the impact of 8-week consumption of almonds (56 g/d) or a control of a calorie-matched pretzel snack (86 g/d) on the immune response using a vaccination model of viral infection in healthy, middle-aged adults aged 40-64 years old. After 4 weeks of consuming the dietary intervention, participants received the seasonal influenza vaccine. The immune response is assessed by measuring rates of seroconversion and geometric mean antigen-specific antibody titres at 4 weeks post-vaccination. Participants are also followed-up at 2- and 3-months post-vaccination to assess the incidence of self-reported upper respiratory symptoms, providing valuable clinical data to support the biomarkers of immune responses.

**Key results so far:** The recruitment target has been met, with 90 participants randomized. Results for study outcomes will be made available once the sample and data analysis is complete.

**Discussion:** Sample and data analysis is underway, and is foreseen to continue to February 2025. The project will provide extremely valuable data, with the potential to inform public health messages regarding almonds on the immune response following influenza vaccination.



# Evaluating Almond Protein for Human Health Benefits

PROJECT NO:PHYS01

## **Principal Investigator:**

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**Co-PIs:** Angelos Sikalidis, Aleksandra Kristos, Elisabet Borsheim, Scott Reaves

## **Summary**

Considering how best to feed the world's growing population while conserving the limited resources that are available is of critical importance. Essential nutrients must be a focus of the strategies employed since they not only sustain human life, but have also a profound impact on health, longevity, and well-being. Protein is one of the most abundant elements in tissues in the body that must be supported by a consistent adequate intake/flow of protein of a certain composition. Protein source has important implications in terms of sustainability but also quality of protein with animal-based being of higher quality but also more taxing on the environment compared to plant-based. While plant-based food alternatives extend several environmental benefits, it is critical that nutritionally, they function in a similar manner to animal-based protein sources. Our study uses a combination of well-established techniques that we have used in published work recently as well as novel techniques to evaluate the quality and effects of plant proteins (almond and rice) relative to animal protein when consumed by study participants. Furthermore, we explore how enhancing almond proteins can improve their quality and functionality bringing their attributes closer to those of whey, an animal-based protein considered the "gold standard" in terms of protein quality supplement. The aim is to provide a high-quality environmentally and vegetarian friendly plant-based protein as a good alternative to whey, catering to consumer sensitivities over the environment as well as choice. In our study, healthy female undergraduate college students are recruited. Dietary assessment including protein intake is conducted at baseline while measurements including anthropometrics, nitrogen balance and body composition will be performed. After creating similarly homogeneous groups each receives a protein supplement whereby it is isocaloric and isonitrogenous with the difference in the protein source (animal vs. plant). The different supplementation regimes are: 1. Whey protein powder (WP) a standard animal based high quality protein to serve as control; 2. Almond protein powder (AP); 3. Almond protein powder enhanced; After a 12-week supplementation whereby participants will be receiving their supplement in the form of a drink daily, assessments post intervention will be conducted including those at baseline as well as blood amino acid composition. Muscle "virtual biopsies" will be conducted on week 2, whereby the extent, profile, and mechanism of protein incorporation to the muscle will be evaluated.

# The Effects of Almond Consumption on Functional Performance Aerobic Capacity and Physical Activity in Overweight and Obese Active Older Adults

PROJECT NO: 19-ArjmandiB-NR-01

## Principal Investigator:

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## Summary

Our overall objective was to determine the effect of 12 weeks of almond consumption (ingested as a snack twice daily) on energy expenditure, performance, and functional-related outcomes, in active, overweight/obese older adults compared to 12 weeks of an isocaloric matched control snack. The central hypothesis of this parallel arm study centered on the idea that daily consumption of an almond snack for 12 weeks would contribute to improvements in energy expenditure, physical and functional performance, vascular function, inflammation/oxidative stress, sleep quality, mood status, and body composition. Recruitment is complete and data is currently being analyzed to achieve the three following aims:

- 1) To determine the extent to which 12-week consumption of roasted almonds affected functional performance and aerobic capacity. To assess this aim we administered the Continuous-Scale Physical Functional Performance (CS-PFP) test and a submaximal aerobic test.
- 2) To determine whether 12-week consumption of roasted almonds affected physical activity, sleep quality, mood status, and body composition. To assess this aim we measured resting metabolic rate (RMR) and substrate utilization, daily physical activity, and sleep quality via Actigraph accelerometry and Pittsburg Sleep Quality Index [PSQI], subjective mood status via Profile Mood States Questionnaire (POMS), and body composition via dual-energy X-ray absorptiometry (DXA) to assess android, gynoid, and total lean and fat mass.
- 3) To determine whether 12-week consumption of roasted almonds affected vascular function, and inflammation/oxidative stress. To assess this aim we measured markers of vascular function with flow-mediated dilation (FMD), pulse wave velocity (PWV), and pulse wave analysis (PWA).

# The Effect of Almond Consumption on Skin Collagen and Elastin in Women

PROJECT NO:SH01

## **Principal Investigator:**

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## **Summary**

The purpose of this study is to investigate how almond consumption impacts skin health in premenopausal and postmenopausal women, focusing on collagen production, skin elasticity, wrinkle severity, and pigmentation. Almonds are rich in nutrients such as vitamin E, polyphenols, and healthy fats, which have been shown to offer protective benefits against oxidative stress and skin damage. Previous studies have suggested that almonds may help reduce wrinkles and improve the skin's resistance to sun damage, particularly in postmenopausal women, whose skin often shows more signs of aging due to decreased estrogen levels.

The study will include women aged 35 to 70, with half being postmenopausal (defined as having had no menstrual periods for over a year) and the other half premenopausal. Participants must have maintained a stable diet and skincare routine for at least six months before starting the study and must continue these routines throughout the study. Exclusion criteria include nut allergies, current use of collagen or certain supplements, pregnancy or breastfeeding, recent cosmetic procedures, and significant skin conditions. Participants will be randomly assigned to two groups: one consuming 60 grams (about 2 ounces) of almonds five times a week, and the other consuming a similar amount of non-nut-based food with matched calories and protein. To measure the effects of almond consumption, research coordinators will take high-resolution facial photographs to evaluate wrinkles and skin pigmentation. These images will be analyzed using advanced facial imaging systems to track changes in wrinkle severity and pigment intensity. Participants will need to come to their study visits without makeup or skincare products to ensure accurate measurements. Blood samples will be collected at three different visits to analyze various health markers, including those related to skin health. Participants will fast overnight before these blood draws, which will be done before noon. A small skin biopsy will also be performed to assess the levels of enzymes related to collagen breakdown, specifically matrix metalloproteinase 1 (MMP1) and tissue inhibitor of metalloproteinases (TIMP1). This will involve a local anesthetic, a small punch tool to remove a skin sample, and stitching to close the wound. The biopsy aims to understand how almonds might affect collagen degradation in the skin.

# Almond Supplementation in Mild to Moderate Acne Vulgaris

PROJECT NO: SH-21-Sivamani-NR-03

## **Principal Investigator:**

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## **Summary**

The purpose of this study is to analyze acne lesion counts, byproducts of microbial activity in the blood (short chain fatty acids), and changes in the types and amounts of various bacteria and fungi in the gut with the use of oral almond ingestion, compared to a control diet. The study will include men and women age 15 to 45 years old that have mild to moderate acne, as judged by a doctor. They should have been using the same acne treatment for at least four weeks, unless the medication is one that is on the exclusion list for which they may choose to undergo a washout period. The exclusion criteria include if they have severe acne, a nut allergy, or are not willing to stop taking oral probiotics, topical antibiotics, benzoyl peroxide, or vitamin E. Additionally, participants will be excluded if they have taken oral antibiotics for acne in the past month, are pregnant or breastfeeding, have changed their hormonal birth control in the last three months, are using supplements that contain nuts, have used isotretinoin in the last three months, or are current or heavy smokers. Participants have been randomly assigned to two groups: one consuming 60 grams (about 2 ounces) of almonds daily, and the other consuming a similar amount of non-nut-based food with matched calories. To measure the effects of almond consumption, research coordinators will take high-resolution facial photographs to evaluate acne progression. Participants will need to come to their study visits without makeup or skincare products to ensure accurate measurements. Blood samples are collected at three different visits to analyze various health markers, including those related to skin health. Participants will fast overnight before these blood draws, which will be done before noon. Skin microbiome and stool are collected to analyze changes in microbiome at three different visits. A genetic sample is collected by swabbing the inside of the cheek at two visits to collect cells to look for the presence of specific gene variations that relate to aging and inflammation. The study has fully recruited 60 participants and is now fully enrolled.

# Effect of Almond Supplementation on Mild to Moderate Acne Vulgaris in a community living population in India

PROJECT NO: SH-22-Udipi-NR-01

## Principal Investigator:

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## Summary

Acne appears to flare in response to high glycemic load diets and foods that stimulate the insulin cascade. Almonds have the potential to lower the glycemic load of foods and augment the short-chain fatty acid deficiency that has been noted in mild to moderate acne vulgaris. People with acne have been reported to have deficiencies in short-chain fatty acid-producing bacteria, gut dysbiosis and microbiome shifts. Nuts are rich in prebiotic fiber and polyphenols and have proven benefits for human gut health and gut microbiota. The hypothesis of this study is that the daily supplementation with 60 grams of almonds will improve mild to moderate acne vulgaris compared to those that receive control supplementation.

This study is a 20 week randomized, controlled, parallel-group study to evaluate how the oral ingestion of almonds influences mild to moderate acne vulgaris, skin and gut microflora, and associated biomarkers like plasma short-chain fatty acids, inflammatory markers, blood glucose levels, and lipid profile. The primary endpoint is total acne lesion count (a combination of inflammatory and non-inflammatory lesions). 200 participants will be screened for 100 inclusions (50 inclusions per group to get 30 complete cases). After obtaining their consent, participants will be randomized into almond or control groups by computer-generated randomization. Participants will be monitored and daily follow-ups done over video calls to determine compliance and any problems encountered associated with the intervention. Recruitment and data collection for this project is ongoing.

# Effects of Almond Consumption on Quality of Sleep in Adults: A Randomized Controlled Trial

PROJECT NO: OTR-22-DESAIS-NR-01

## **Principal Investigator:**

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**Co-PIs:** Dr. Shobha Udipi, Research Director and Head. Hon Director, Integrative Nutrition and Ayurvedicals Kasturba Health Society-Medical Research Centre , Dr. Rama Vaidya; MD PhD Reproductive Endocrinologist Director, Division of Endocrine and Metabolic Disorders Medical and Research Centre -Kasturba Health Society

## **Summary**

This study is a randomized controlled trial examining the effect of almond consumption on quality of sleep in adults with poor sleep quality aged 21-55 years in Mumbai, India. Sleep patterns are assessed using Pittsburgh Sleep Quality Index (PSQI) and Epworth Sleep Scale (ESS), polysomnography and actigraphy. The study will also determine the effect of almond consumption on associated biochemical measurements like serum serotonin and melatonin, brain derived neutrophic factor (BDNF) and cardiometabolic risk factors (blood glucose and insulin levels and inflammatory cytokines and lipid profile).

To date 758 adults between 21 and 55 years of age have been screened (Phase 1). Among these 451 were found to have a higher PSQI and/ ESS scores and were eligible for the study. Of these 227 consented to participate in the second phase of intervention and therefore, underwent polysomnography. All reports have been certified by Dr. Lancelot Pinto, Consultant Sleep Study Physician for the project. Some participants were diagnosed with severe sleep apnea and therefore, could not be included further leaving a total of 209 participants included in the intervention phase. Endline data collection is remaining for four participants in the almond group and six in the control group. The majority of participants have finished with the intervention and endline data collection including anthropometry measurements, biochemical measurements, physical activity questionnaire, quality of life and 24-hour diet recall. Once all participants have completed all study-related interventions data analysis will be completed and final results will be available.

# Almonds and the Gut-Brain Axis: a Randomized Controlled Trial to Improve Mental Health, Psychological Distress and Quality of Life

PROJECT NO:22-Dimidi-NR-02

## Principal Investigator:

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**Co-PIs:** Professor Kevin Whelan, Dr Joseph Chilcot

## Summary

Symptoms of depression and anxiety have seen a recent marked global increase. Despite growing evidence for the modulating role of healthy dietary patterns in managing symptoms of depression and anxiety, further research is required. Consumption of snacks now contributes to roughly 17% and 21% of daily energy intake for men and women respectively. Almonds are a healthy snack option and are readily consumed worldwide. They are high in a range of nutrients and other bioactive compounds, some of which are known to elicit physiological responses that can affect brain function. For example, our meta-analysis has shown that almonds may affect gut microbiota composition, and our recent study has shown they can also modulate metabolite production (e.g., short-chain fatty acids), both of which are involved in the regulation of the gut-brain axis. However, there is little evidence of the effect of almonds on other aspects of mental health such as symptoms of depression and anxiety. Therefore, given: (i) the high content in key nutrients that affect oxidative stress and inflammation that are linked to psychological distress, (ii) prebiotic effect on the gut microbiota and their metabolite production, and the impact of these on gut-brain signaling, and (iii) potential displacement of unhealthy snacks with almonds, there is a strong rationale underpinning the potential effect of almonds as a low-cost approach for optimizing mental health. The NutriMood study aims to investigate the effects of whole almonds on mental health and the gut-brain axis compared to a commonly consumed snack, in adults with mild to moderate symptoms of depression and anxiety. Eighty four participants aged 18-45 enrolled in a 12-week parallel randomized controlled trial. Participants either consumed 56g of whole almonds per day or 2 iso-caloric muffins per day as a control snack. Mental health, including symptoms of depression, anxiety, mental well-being, quality of life, sleep quality, functional impairment and psychological distress, were assessed at 3 time points (weeks 0, 6 and 12) of the intervention. Stool and blood samples were collected at two time points (weeks 0 and 12) for the analysis of the gut microbiome and serum vitamin E levels. Data collection was complete in May 2025 and results are being analyzed. This study will provide unique and valuable data to the rapidly expanding field of nutritional psychology. There is a potential rationale and gap for singular foods to operate as an adjuvant treatment for mental health alongside traditional talking therapies or medication. The use of singular foods as an intervention for mental health would provide a highly accessible, low-cost intervention without the need for specialist dietary counseling which is integral to the success of whole-diet interventions.

# Randomized Controlled Trial of Almond Supplementation vs Isocaloric Diet on Cognitive Functions in Middle-Aged (40-60 years) Asian Indians with Prediabetes

PROJECT NO: CFP-21-MISRA-NR-01

## Principal Investigator:

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## Summary

The primary objective of this study is to evaluate the effect of almond supplementation on cognitive functions using computerized neuropsychological assessment systems (CANTAB). The co-primary objective is to evaluate changes in neuroimaging (blood flow, and the expression of functional brain networks during cognitive demands) using functional MRI. The secondary objectives are to observe the mean change in structural MRI, hippocampal volume, white and grey matter changes, to evaluate resting-state connectivity using resting-state MRI, and to correlate oxidative stress with neurocognitive measures. This is a randomized controlled parallel arm study on free-living Asian Indians, selected based on inclusion criteria. The sample size is 60 subjects (n, 30 in each arm). The measured parameters pre and post-intervention include 24-hour diet recall, food frequency questionnaire, physical activity assessment, anthropometry including circumferences, height, and weight, blood parameters including blood glucose (fasting and 2 hrs.), serum insulin (fasting and 2 hrs.), lipid profile, inflammatory parameters, and the parameter of oxidative stress, cognitive function assessment using CANTAB, functional MRI and, hippocampal volume assessment. Recruitment for this study is complete. We have done MRIs for 60 subjects at baseline and 38 subjects have completed the intervention. We will start the data analysis once we have completed the data collection.



# Almond Consumption and the Risk of Cancer Incidence and Mortality in a Population with a Wide Range of Intake

PROJECT NO: OTHN01

## Principal Investigator:

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## Summary

This study will examine the relationships between almond consumption, independent of other dietary factors or lifestyles, and the risk of developing or dying from cancer in the Adventist Health Study-2 cohort and will examine if the purported effect of almonds on cancer is distinct for men and women. As malignant neoplasms (cancer) are the second leading cause of death in the United States of America, a large portion of scientific study is understandably directed at both primary and secondary prevention of these diseases. In the field of nutrition, increased consumption of nuts have been related to cardiovascular health, with almonds being one of the nuts having a strong evidentiary basis for this association. However, while many of the nutrients present in almonds have also been identified as protective against cancer, research into the potential protective effect of almonds as a whole food has been much less abundant. The Adventist Health Studies have landmark publications on nuts, and a strong history of cancer research in the epidemiological field. The Adventist Health Study-2 (AHS-2) prospective cohort has been ongoing since 2002, with an initial food frequency questionnaire which contained comprehensive information on over 200 food items, including almond consumption, completed by more than 96,000 study participants upon enrollment. There have been linkages with the National Death Index for nationwide vital statistics, and state-level cancer registries for data on cancer incidence. With such ample data available for analysis and a cohort profile that has both a low presence of notable confounders like alcohol and tobacco use, and a wide range of almond consumption, the AHS-2 presents an ideal opportunity for research that may uncover an association between almonds and cancer prevention if one exists. Cox proportional hazards regression will be performed to investigate the effects that almonds have on the risk of cancer incidence and mortality. Validated food frequency data from the AHS-2 will be used, with multiple imputation for missing data, and regression calibration on the exposure of interest, almond consumption. Models will be built a priori, with identical statistical analysis conducted between the primary and secondary objectives, with the latter being comprised solely of the female participants.

# Deep Metabolomic Profiling of Almonds

PROJECT NO: NUTC01

## Principal Investigator:

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## Summary

This study focuses on analyzing three almond varieties—Nonpareil, Independence, and Monterey—to uncover their potential health benefits by profiling bioactive compounds. These compounds, known as secondary metabolites, promote health in various ways. The research has four main goals: to perform a detailed analysis of the bioactives in almonds, identify and measure these health-promoting compounds, compare the metabolite composition between the varieties, and use the findings to improve the nutritional value of almonds. The study highlights almonds as a nutritious food, rich in bioactives alongside healthy fats, proteins, vitamins, and minerals. Using Brightseed’s advanced Forager® technology, researchers identified 212 metabolites and 318 lipids across different almond samples, discovering four times the number of compounds previously known. Notably, 17 bioactive compounds with proven health benefits were identified, six of which were found in almonds for the first time. These compounds support key areas like immune, metabolic, gut, cellular, and brain health. Overall, the research positions almonds as a valuable functional food that can contribute to multiple aspects of human health. Future research can explore the specific health benefits of these compounds and how they can be applied to health innovations.

# Effect of Water Activity, Temperature and Incubation period on Fungal Growth and Aflatoxin Production on Almond nuts

PROJECT NO:22-GizachewD-AQFSS-01

## Principal Investigator:

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## Summary

*Aspergillus parasiticus* is a fungus that can infect almonds and produce carcinogenic aflatoxins (AF). The objective of this study was to determine the growth and total AF production of *A. parasiticus* on three types of almond kernels (in-shell, shelled and split) under different combinations of temperature (20, 27 and 35 °C), moisture (65%-99%). Measurements of fungal growth and aflatoxin were conducted after 10, 20 and 30 days of incubation. The in-shell kernels supported the least amount of growth and AF production. The fungus grew moderately at 90% and 95 % moisture and produced moderate (<50 µg/kg) amount of total AF at 95% on in-shell almonds. On shelled kernels, growth was also limited to 90% and 95%. Aflatoxin production reached high levels at 0.95 aw at all three temperatures on shelled almonds (324.3, 325.4, and 139.8 µg/kg at 20, 27 and 35 °C, respectively).

The fungus grew rapidly and produced high levels of AF (>300 µg/kg) on split kernels over a wide range of conditions 80% to 95% moisture and 20 to 35 °C temperature. Contour plots revealed that the optimum conditions for AF production on split kernels were at 90%-95% moisture and 20-27 °C temperature. Aflatoxin production also depended on incubation time. By day 30, AF production on split kernels exceeded 300 µg/kg at 80% at all temperatures. There was no fungal growth and AF production at 65% and 75% moisture level for up to six months of incubation period.

These findings indicate that maintaining the moisture level to 75% or less during transport and storage can reduce the risk of infestation and aflatoxin production by *A. parasiticus* on inshell, shelled and split almond kernels.

# Full Demonstration of SmartProbe Technology for Early Detection of Insect Pests and Environmental Monitoring

PROJECT NO:22-PanZ-AQFS-01

## Principal Investigator:

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## Summary

The goal of this project was to fully demonstrate a new wireless AI based SmartProbe technology (SPT) for early insect pest detection, monitoring environmental conditions and moisture content of stockpiled almonds and to make the technology commercially available for implementation in the almond industry to reduce concerns and economic losses from all the problems caused by insect infestation and moisture. Three different models of SmartProbe (Model L for stockpiles, Model S for bins, Model H for processing room) were successfully demonstrated at four different almond processing facilities. This project also identified the starting locations of insect infestation in stockpiled almonds and the significance and rate of insect damage in almonds under different conditions. The key findings and recommendations from this research project are summarized as below:

1. SmartProbe was an effective tool that detected the insect pests (red flour beetles, Indian meal moths, gnats, ants and drugstore bugs) in the early stage of the infestation in almond stockpiles, processing room, storage bins and storage warehouses, but no insects were expected or detected by human visual inspection.
2. It was found that the insect damage rates of stockpiled almonds ranged  $4.6\pm 0.6\%$  to  $14.0\pm 1.4\%$  and the rates increased by 1.95% on average in two weeks.
3. The insect activities started in the surface layer in the top location in the stockpiles due to warmer temperature.
4. SmartProbe also effectively monitored the product moisture through temperature and relative humidity.
5. SmartProbe detected insect pests much sooner and had better catching ability compared to the conventional traps in the processing rooms. SmartProbe with dust prevention worked well in the dusty environment of processing rooms.
6. SmartProbe system now is commercially available at AIVISION FOOD INC. to be used for early detection of insect infestation, product moisture management, and monitoring of environmental condition and fumigation treatment effectiveness with AI insect identification, automatic alert and report, and reduced labor cost and product loss.

# Investigation CELF-Pretreated Almond Tree Waste for Valorization into High Value Co-Products to Curtail Tipping Charges to the Farmers

PROJECT NO: BIOM01

## Principal Investigator:

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Co-PIs: Charles Cai

## Summary

Almond orchard waste was evaluated as a potential input for fabric. Fabrics like viscose/rayon and lyocell are normally made from the pulp of trees from forests or grown for single-use. Today, 300M+ forest trees are logged annually to produce pulp for the apparel industry. Overall, almond byproduct is a promising candidate for additional process optimization as the out-of-spec parameters are not too far off from desired quantities.

The Hurd Co's agrilose® is a 100% agriwaste-based feedstock pulp used by apparel brands to make fabrics like viscose/rayon and lyocell, which are normally made from forests. In addition to being forest tree-free, The Hurd Co's patented technology is dramatically more sustainable: the zero emission process uses 50% less water and 90% less energy than conventional pulping processes. Agrilose is a one-to-one replacement for forest tree pulp and will be available at a cost-competitive price, making it an easy substitute for manufacturers and an easy choice for brands.

The key transformative technology implemented by The Hurd Co is called CELF (Co-solvent Enhanced Lignocellulosic Fractionation), developed and invented by Dr. Cai at UC Riverside. CELF is a new generation biomass fractionation, pretreatment, and pulping technology that cleanly and efficiently separates cellulose, hemicellulose, and lignin fractions from lignocellulosic biomass feedstocks. CELF is capable of fractionating single or mixed biomass residues from the agricultural and forestry industries to yield high quality intermediates that can be further manufactured into fungible liquid fuels and high value co-products. Impactful products from CELF intermediates include fuel ethanol, biogasoline, sustainable aviation fuels (SAF), green construction materials, bioplastics, commercial adsorbents, and lipids/fatty acids.

The Hurd Co and the biomass R&D team at UC Riverside are internationally recognized as leaders in biorenewables and providers of transformative solutions for upgrading biomass-based waste. Feedstocks that have been successfully valorized in prior R&D projects utilizing CELF technology are poplar, maple, pine, corn stover, sugarcane bagasse, switchgrass, hemp, palm, and almond husks.

Overall, almond tree waste shows significant promise as a potential starting material for production of dissolving pulp for use in the apparel industry. The Hurd Co recommends pre-washing raw samples before subsequent processing. The optimization of pulping conditions is needed to realize lower lignin and hemicellulose content.

The current Kappa # is too high and post-bleach washing is recommended to reduce total mineral ash content in order to meet dissolving-grade pulp specifications. Iron and silica content is still high after bleaching and optimization steps are required to achieve proper thresholds.

The Hurd Co further recommends sieving followed by coarse-filter screening and caustic wash. The overall viscosity is too low and more optimization is necessary to increase numbers. This includes additional filtering to remove fines and post-bleach washing to lower silica and iron. With additional process optimization, there is a bright future for a waste-to-wear product made from almond waste.

# Allergenicity and Risk Assessment of Novel Almond Hull Food Ingredients

PROJECT NO: BIOM02

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**Summary**

The project aims to identify potential allergenic proteins in almond hull samples and conduct an allergenicity risk assessment of almond hulls as novel food ingredients, including almond protein residue analysis and in vitro antigenicity and cross-reactivity assessment. In the absence of specific guidelines for allergenic protein identification in novel whole foods, we applied the Codex Alimentarius (CODEX) guidelines for foods derived from modern biotechnology, which recommend a weight-of-evidence (WoE) approach involving multiple analyses to form scientific conclusions. SDS-PAGE and Western blot analyses using anti-almond IgG antibodies (2B4) revealed positive immunoreactivity in Butte and Nonpareil South samples, while one Nonpareil sample showed no positive immunoreactivity. These findings suggest the potential presence of almond or almond-like allergenic proteins in almond hulls or the possibility of cross-contamination during processing. Further analysis is necessary to confirm these observations. Additionally, proteomics results indicated a possible relation between almond hulls and other Prunus species, such as apricots. Based on a literature review and database search, future IgE-based studies will assess cross-reactivity using anti-almond human plasma, along with plasma from cherries, peaches, and apricots.

# Cost Return Studies for California Almonds

PROJECT NO: ECON02

## **Principal Investigator:**

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## **Summary**

Almond production practices and costs continually evolve due to changes in technologies and products available, regulations, pest and disease pressure, input prices and other relevant factors. To remain competitive, California almond producers must have up-to-date decision-making tools that are relevant to each almond-growing region in the state. In recent years, relatively low almond prices have emphasized the need for accurate cost of production assessments for almond operations to remain economically viable. In this project, we conducted five sample costs and returns studies for almond production across almond-producing regions and production practices. This project builds on prior almond cost and return studies.

Comparing the studies completed in 2024 with those from 2019, we find that operating costs have increased by 38-40% depending on the region. Operating cost increases are due to increasing labor, fuel, fertilizer and pesticide costs, as well as increased interest on operating loans. Overhead costs (cash and non-cash) have increased by 57-60% between 2019 and 2024. Much of the increase in overhead costs is due to higher interest rates in 2024 (8.25% compared to 6% in 2019) which increases capital recovery costs (non-cash overhead) associated with investments. However, regulatory and environmental fees and office expenses also increased, which added to higher cash overhead costs. The price of almond meats used in the 2024 studies (\$1.60/lb) was 36% lower than that used in 2019. The almond price is assumed to be a weighted average of nonpareil and pollinizer varieties. Due to the higher costs and lower almond prices, net returns above operating and total costs were negative for all of the conventional almond studies.

# Irrigation Management: Evaluating Current Sensor-Based Products and Remotely Sensed Information and Testing Thresholds for Delaying the Start of Irrigation in the Spring.

PROJECT NO: HORT22

## **Principal Investigator:**

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## **Summary**

A fully automated closed-loop irrigation control system was developed that independently maintained trees at a desired target level of water deficit (moderate and severe levels of SWP). The target midday SWP levels for this initial year were about 18 bars for moderate and about 25 bars for severe stress. FloraPulse sensors were used to continuously monitor SWP, and periodic checks showed close agreement to the pressure chamber at midday. Automated daily irrigation was based on comparing the previous days' 24h average SWP to the 24h average that corresponded to target value of midday SWP. 24 hour averages were used to as the basis for control because they would be less subject to short term variations in weather conditions, but would still be indicative of long term trends in tree stress. The system changed irrigation amounts depending on how weather and soil water availability affected the trees, and resulted in remarkably stable daily patterns of SWP. Kernel weight was closely related to seasonal average SWP, but yield was not. This was not surprising because kernel weight is strongly dependent on within-season water stress effects, whereas yield is influenced by carry-over effects on spur growth and bud development. This was the first year of such an experiment and 2023 was also a low crop year, so yields in 2024, compared to the control treatment, will determine if the targets of 18 and 23 bars, which will be imposed in 2024, should be maintained or reevaluated for 2025. In the final analysis, this experiment will be the first step in determining the appropriate SWP set-points for automated, plant-based irrigation control.



# Updating Information on Evapotranspiration (ET) and Crop Coefficients (Kc) of Micro-Irrigated Almond Production Orchards Grown in California for use in Water Resource Management and Irrigation Scheduling Decisions

PROJECT NO: HORT52

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## Summary

Energy flux data was collected in 27 orchard-seasons from eight different locations in the Central Valley. All of the almond orchards were mature and micro-irrigated located in the upper Sacramento and San Joaquin valleys. Weather and reference evapotranspiration (ET<sub>o</sub>) data were collected from representative CIMIS stations to develop crop coefficient (K<sub>c</sub>) values for calculating potential ET<sub>c</sub> = ET<sub>o</sub> × K<sub>c</sub>. The energy flux station data were collected by various groups, but all used the residual of the energy balance method with measured net radiation, ground heat flux, and sensible heat flux to calculate half-hourly values of latent heat flux (LE). The LE was then converted to ET in mm per half hour, and daily ET<sub>c</sub> was calculated as the sum of the 48 half-hourly periods from midnight to midnight.

A goal of the project was to determine if micro-climate differences affect almond K<sub>c</sub> values. Datasets came from the north and south regions of the Central Valley, but the ET<sub>o</sub> values were nearly identical for all CIMIS stations except Durham #12, which had mid-summer daily ET<sub>o</sub> values 0.8-0.9 mm lower than the other stations. The lower ET<sub>o</sub> values were likely due to surrounding orchards that blocked the wind. Three of the study orchards had mid-season K<sub>c</sub> values between 1.10 and 1.20, whereas the other four other study orchards had mid-season K<sub>c</sub> values near 1.00, which suggests that the orchards with mid-season K<sub>c</sub> less than 1.10 were not at potential ET<sub>c</sub>. The K<sub>c</sub> differences are most likely due irrigation practices.

The main goal was to develop and disseminate improved K<sub>c</sub> information to growers. However, because there is uncertainty as to why the study orchards had different mid-season K<sub>c</sub> values, an application program "KcGen.xlsx" was developed to help users identify accurate K<sub>c</sub> values by presenting the K<sub>c</sub> curves for the seven study orchards and letting the users select which curve most likely matches their orchard(s). The KcGen program outputs K<sub>c</sub> information with several time-steps in tabular and graphical formats to provide data for existing irrigation programs. KcGen also utilizes historical monthly means of daily ET<sub>o</sub> data from local CIMIS stations to calculate mean ET<sub>c</sub> values for irrigation planning purposes. The program also utilizes 1-7-day ET<sub>o</sub> forecasts using (FRET) data from the National Weather Service with leads of 1 to 7 days to allow for prospective irrigation scheduling. In addition, the KcGen application program was developed to allow the estimation of potential ET<sub>c</sub>, but it also includes the ability to estimate ET<sub>c</sub> reductions due to regulated deficit irrigation during the growing season by letting users input information on timing of deficit irrigation and applying a recommended K<sub>c</sub> reduction. This feature improves the ET<sub>c</sub> estimation during deficit irrigation periods such as prior to and during hull-split and nut harvest. It is impossible to completely account for lower crop coefficients during deficit irrigation periods without inputs from growers because there is a lack of information. This limits accuracy of the Cal-SIMETAW model for developing projections of seasonal water use for almond orchards and ET of Applied Water (ETAW), which is used in California for water resources planning; however, a 2-3-week period of reduction in ET due to planned deficit irrigation will likely result in over-estimating seasonal ET<sub>c</sub> and ETAW. The project team is working with Department of Water Resources scientists on how to resolve this problem. The collaborators on this project are associated with UCCE and we expect to devote considerable effort towards the dissemination of results. The project team is currently working with UCANR to develop a web repository to facilitate dissemination of updated information on ET<sub>c</sub> and K<sub>c</sub> for almond production.

# Yield Prediction for Resource Management and Yield Optimization in Almond

PROJECT NO: HORT66

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**Co-PIs:** Sat Darshan S. Khalsa, Yufang Jin, Mason Earles, Stavros Vougioukas Vougioukas

**Summary**

The single tree yield analysis for almonds revealed significant variations at the tree, cultivar, and regional levels. 'Nonpareil' and 'Monterey' displayed varying fruit-set percentages at the Woodland site, with 'Monterey' showing higher fruit-set but not resulting in higher yields, despite similar kernel weights. Conversely, 'Monterey' outperformed 'Nonpareil' in yield at the Madera site, possibly due to its higher kernel weight. Tree size and flower abundance are significant factors influencing the fruit-set and yield relationship. Annual trunk growth varied between sites, with younger trees exhibiting higher growth rates. Interestingly, the correlation between trunk growth and tree yield was consistent across both sites, with higher trunk growth in the previous year correlating with increased yield in the current year. However, a comprehensive understanding of the growth-yield association in almonds requires assessments of blooming time, synchronization with pollinizers, bloom density, and the effectiveness of pollination at the tree level.

In 2023, the Vougioukas Lab focused on simultaneously measuring the yield volume and weight of individual trees at commercial harvesting speeds. UC Davis designed and built a yield measurement system that was mounted on a commercial off-ground harvester. The system was tested and validated during the 2023 almond harvest at several orchards in California, and it proved to be accurate to within 7.5% of manually collected ground truth measurements. The system used a laser line profiler mounted over a conveyor to measure volume and a weighing conveyor to measure mass. Additionally, a GPS unit, vibration sensor, and ground speed radar were used to determine the location of each tree and match the yield with the tree location. Development of an algorithm began which subtracts leaves and debris from point-cloud images of the almond stream to improve measurement accuracy.

In 2023 Jin Lab focused on forecasting the tree-level yield variability and understanding the water use efficiency within an orchard. We integrated individual tree yield and time series of multispectral satellite imagery from Planet with Convolutional Neural Network (CNN) deep learning methods to predict individual tree almond yield with various cadence. Further analysis was done to quantify the water use efficiency at 30m. The CNN model predicted the tree level yield very well especially after bloom imagery was incorporated and captured in-field yield variation patterns. This is the first scalable study attempted for yield forecasting at a fine scale with Planet satellite imagery. Tree level yield estimation over the whole orchard also allowed for within-field water use efficiency (WUE) assessment at the end of the season, filling an important gap in site-specific resource management. Work is ongoing to build more robust yield forecasting models to capture both spatial and year-to-year variability of almond yield, by pooling the multi-year yield data and remote sensing observations.

Through 2023, the Earles lab developed a ground-based sensing device equipped with RGB cameras, a GNSS system, lights, a microprocessing unit, and a custom PCB board to collect geo-located imagery. Over three years, approximately 3.6 million images were captured at three phenological stages (bloom, fruit set, and harvest) from three locations. A flower detection model was developed using a deep learning model to estimate the number of flowers per tree at bloom. A deep learning model was employed to develop a flower and fruit detection algorithm, achieving an internal performance  $r^2$  of 0.85 for flower estimation and 0.60 for fruit counting. When compared to manually measured kernel yield for around 120 trees, the algorithm showed an external performance  $r^2$  of 0.46 for flower counting and 0.19 for fruit counting. These images were then processed to create spatial distribution maps for attributes including flower/fruit count, fruit set ratio, kernel weight, and yield prediction.

# Data-driven Smart Irrigation for Almond Orchards

PROJECT NO: HORT69

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## Summary

The project evaluated stem water potential (SWP) sensors within commercial almond orchards across California's Central Valley, during the 2020 and 2022 growing seasons, with a focus on both Osmatic Cell (Saturas) and Microtensiometer or MT (FloraPulse) sensors embedded into almond tree trunks and branches. Performance against pressure is reported.

At various farms, including CAPEX, Gruenwald, Nickels, Sharma, UC Kearney, and CSUC, OC sensors exhibited good correlations and statistical significance compared to the pressure bomb. However, OC sensors performed poorly at Westwind and Clark farms. In contrast, MT sensors consistently demonstrated good agreement with the pressure bomb, strong correlations, and statistical significance across all sites, showcasing reliability with an average error of 0.22 MPa. This research demonstrated the robustness of MT sensors for continuous measurement of SWP, making them a promising tool for helping growers optimize almond orchard water management. One of the main advantages of the MT sensors is the ability to show recovery from water stress following irrigation as demonstrated at Jasleen farm. Stress recovery within one to two days was observed, highlighting the real-time monitoring capability of MT sensors. This dynamic feedback presents promising prospects for revolutionizing precision irrigation in almond orchards. Additional details on microtensiometer evaluation in almonds can be found in this publication by Kisekka et al. (2024) (<http://dx.doi.org/10.2139/ssrn.4713202>).

Integrating smart irrigation technologies, which combine soil-plant-weather monitoring, is of interest to many almond growers, especially under SGMA (Sustainable Groundwater Management Act). Using a machine learning model called Random Forest (RF), we identified solar radiation, temperature, neutron count, and soil water content as the main factors influencing changes in SWP. We developed a predictive model for SWP based on RF. The RF model consistently exhibited strong correlations with SWP measurements obtained from the Pressure Chamber. The machine learning model can be used to upscale SWP measurements from individual trees to the entire orchard. This type of model would be useful for guiding variable rate irrigation. Detailed results on SWP mapping using machine learning can be found in this publication by Savchik et al. (2024) (<https://doi.org/10.1007/s00271-024-00932-8>). In addition, research is underway to develop an iOS AI-enabled app for estimating canopy light interception which can be used as a proxy for orchard-specific crop coefficient.

The study also assessed satellite-based remote sensing of evapotranspiration (ET) using the SEBAL model. Commendable correlations between remotely sensed ET and eddy covariance were observed for three seasons from 2020 to 2022. While effective for estimating monthly to seasonal orchard responses to irrigation scheduling, challenges persist in achieving acceptable daily-scale accuracy. Research on integrating remotely sensed ET with ground-based measurements to reduce errors in daily ET is currently ongoing and will be reported in future reports.

The main findings to date are summarized below:

- Microtensiometers produce stem water potential values that are comparable to a pressure chamber (bomb).
- The ability of the microtensiometers to measure water stress recovery following irrigation is a powerful feedback for almond irrigation management.
- Remote sensing of evapotranspiration based on the SEBAL is accurate at the monthly and seasonal time scales, more research is needed to reduce errors at the daily time scale relevant to irrigation management.
- Artificial Intelligence models can predict stem water potential at individual trees and over the entire orchard from easily measurable variables e.g., solar radiation, temperature, evapotranspiration, and soil water content.
- Remote sensing data such as land surface temperature can be used in the delineation of irrigation management zones.

# Immobilizing Soil Nitrate Using Almond Shells in Winter-Fallow Vegetable Fields to Reduce Nitrate Leaching

PROJECT NO: INSH02

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## Summary

In the fall, incorporating nitrogen-rich cool-season vegetable crop residues (e.g., broccoli, cauliflower) allows rapid tissue decomposition under mild winter temperatures in Coastal California. The resulting pool of residual soil nitrate is vulnerable to leaching by rains during the winter fallow. When cover cropping is impractical, immobilizing nitrate by high-carbon amendment applications can help reduce nitrate leaching. Our previous studies found <2 mm sieved almond shells applied at 5 tons/acre as one of the most effective immobilizers but were cost-prohibitive. The incubation study examining a range of application rates (0, 5, 10 tons/acre) and almond shells' particle sizes (unground, ground to ½", ¼" and 2 mm) suggested <¼" sieved almond shells at 5-7.5 tons/acre as a lower-cost effective practice. Based on these results, a non-replicated strip field trial was established to evaluate a 0, 5, and 10 tons/acre rate of applied almond shells ground to ¼ inch particle size in a large-scale commercial field in Gonzales, CA. The material was applied in one strip the length of the field (30 feet wide by 900 feet long), which allowed us to evaluate one pass with a soil chisel vs. two passes to incorporate the material into the soil. Unfortunately, the Salinas River flooded the trial in January and March 2023, and the sampling was terminated after the second flooding. Monthly soil nitrate monitoring at 0-3 feet depth, until the field was flooded in March 2023, suggests that there was reduced residual nitrate in the 10 tons/acre almond shell treatment incorporated with both one and two passes of a soil chisel. However, soil nitrate concentrations were highly variable, and no clear difference was found in the total amounts of nitrate-nitrogen across 3' soil profiles between the control and all almond shell treatments (e.g., 360-460 lb-N/acre/3 ft soil profile before flooded in Nov. 2022 and 43-68 lb-N/acre/3 ft soil profile after first flood in Feb. 2023). This might suggest a limitation of this approach when nitrification and nitrate leaching occur before immobilization due to the moist soil in the fall and early winter rain. A lab incubation study examined the effect of soil texture and soil organic matter (SOM) content on nitrogen immobilization and remineralization. Regardless of soil type and SOM content, < 2mm ground almond shells immobilized 86-100% of soil inorganic nitrogen at 2 weeks. However, the soil type and SOM content affected the nitrogen's remineralization rate at 4 weeks and beyond; the sandier the soil, the faster the remineralization, and the higher the SOM content, the faster the remineralization. It showed that the soil texture and SOM content can affect nitrogen remineralization and nitrogen availability for a successive crop.

# Optimizing Potassium Management in Almond

PROJECT NO: INSH03

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**Co-PIs:** Daniel Geissler

## Summary

Limited knowledge of K dynamics in plant and soil compromises soil and plant sampling technologies and limits our ability to manage orchard K effectively. Even in relatively small areas with the same fertilization history, non-uniform patterns of K have often been observed. The availability of plants in soil K is affected by a multitude of different factors, such as soil moisture, soil-forming processes, soil mineralogy, soil pH, presence of soil potassium solubilizing microorganisms, plant growth and yield, management factors, and land use. The combination and the interactions between these factors create variability in the spatial and temporal distribution of soil K and ultimately complicate K management in agricultural settings. Moreover, in the context of micro-irrigated tree crops, in which wetted zones are concentrated in limited portions of the soil, the dynamics between soil and plant processes that affect K availability can be particularly non-homogeneous within the orchard. As a result of inaccurate tools for quantifying K soil and a limited understanding of soil K dynamics, despite the known K variability, growers default to applying uniformly high rates of K applications. With the recent dramatic increases in K fertilizer prices, however, excessive applications are wasteful and not economically viable. It is essential to develop and make available a way to cost-effectively map soil and plant K variability to improve the efficiency of K fertilization. Variability also in K leaf status has been observed between samples, dates, and locations. Little literature has focused on the uniformity of distribution of leaf K within the canopy, despite its implication for the accuracy of leaf testing and on the subsequent identification of critical values, which are then used as the baseline for fertilization decisions. We are conducting a study in two productive almond orchards located in Woodland (CA) and Madera (CA). The study, through intensive soil and plant surveys, aims to improve our understanding of the complex dynamics of K in the soil and in the plant with the ultimate objective of improving K management and K use efficiency.

Our results, so far, suggest that there is a consistent interaction between soil exchangeable K with cation exchange capacity (CEC), moisture, organic matter, and saturation, that could be modeled and used to predict exchangeable K spatial distribution. On the other hand, we did not identify any clear correlation between soil solution K and other soil or plant factors. The distribution of both forms of soil K was non-uniform throughout the orchards, without any clear obvious explanation. We have started a collaboration with Dr. Elia Scudiero of UC Riverside, with whom we are mapping our orchard using Gamma Ray and EMI sensors. The confluence of new remote sensing and soil sensing technologies and their integration in this project represents a unique opportunity to improve K analysis.

Our results also suggest that the distribution of leaf K is non-uniform throughout the canopy, with the top of the canopy having the lowest leaf K concentration. Leaf K is also affected by the sampling time throughout the day. Once we gather a larger dataset about the distribution of leaf K within the canopy with a clear understanding of the physiological processes involved, modeling can be integrated to guide the leaf sampling and detect the nutritional status of the whole canopy, taking into consideration also the temporal variability in leaf K that has been observed. Then, with the implementation of new non-destructive technologies, such as hyperspectral images, plant nutritional status can be predicted and mapped on a larger scale and with more precision.

# Evaluating HFLC Nitrogen Management Strategies to Minimize Reactive Nitrogen Mobilization from California Almond Orchards

PROJECT NO: INSH04

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**Co-PIs:** Patrick Brown, Isaya Kisekka, Christine Armstrong, Spencer Jordon

**Summary**

This project provides the first comprehensive assessment of groundwater nitrate impact from a best management practice (in this case, HFLC) comparing three monitoring approaches to assess nitrate impact to groundwater: (1) Groundwater monitoring, the regulatory gold standard to assess pollution sources. Groundwater sampling is performed at 20 monitoring wells (screened at 7-14 m below ground surface). (2) Vadose zone monitoring provides immediate feedback on potential groundwater nitrate discharge but can be labor-intensive. Vadose zone monitoring is performed at 7 multi-level sites (0-3m depth) where soil moisture, nitrate, and ammonium fluxes are measured. (3) The NUE or nitrogen balance is a tool familiar to growers under the ILRP but its relationship to actual groundwater nitrate discharge is not well understood. Monitoring data is used to calculate water and nitrate (N) mass balance, as employed by the ILRP.

HFLC has shown promising results during the first five seasons of implementation (2018-2023). On average, reported NUE has increased by 17% and kernel yields have increased by 15% when compared to the previous five years of pre-HFLC orchard management. Residual N mass in the first 60cm of the orchard soils and pore-water nitrate concentrations in the vadose zone (measured to a depth of 280cm) have both shown decreases following the switch to more efficient nutrient management. Numerical models suggest that although nitrate concentrations quickly decreased in the shallow vadose zone, there will be an up to 25-year delay between the start of HFLC and an observable decrease in groundwater nitrate concentrations. This delay is caused by the long transport time that nitrogen experiences between surface application and groundwater leaching. Our unsaturated zone models suggest that this transport time can be between 15 and 25 years, depending on soil types (i.e. water and N move slower in clayey soils than in sandy ones). Then, once in groundwater, there will be a further delay of about five years before observable dilution will occur. Simulations also suggest that small pockets of irrigation distribution non-uniformity, along with a heterogenous geology, contribute to the highly spatially distributed groundwater N concentrations measured in the monitoring wells.

# Optimizing Nitrogen and Water use Efficiency in Replanted Orchards After Whole Orchard Recycling

PROJECT NO: INSH06

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## **Summary**

The orchard recycling practice has been adopted widely across California and promoted by public agencies including USDA-NRCS and CDFA. Food supply chains have developed sustainable initiatives promising to source products that improve soil carbon storage and mitigate water quality impacts using best management practices. There is a need to quantify trends in soil health status on a broad scale to validate and promote continued support for this practice. Soil health metrics were strongly associated with WC treatments across all sites. Locations with a WC amendment rate > 50 tons / ac had significantly higher values for all indicators in comparison to conventionally (CN) prepared soil in all categories, while the lowest 35 ton/ac application only increased soil organic matter levels. Notably, WC treatments resulted in significantly higher bioavailable N and total N suggesting WC creates conditions for N retention for a longer period in the soil which may enhance tree uptake, nitrogen use efficiency, and minimize N losses to leaching. A better understanding of the complex soil environment interrelationships stimulated by wood chip (WC) amendments is critical to determining best management recommendations such as the appropriate fertilization rate for newly established orchards following WOR. In the first four years of the nitrogen rate trial, no significant association was established between N rate and tree growth, yield, or nutrition for either control or wood chip treated soils. We expect the N treatment results to separate as trees mature and yields increase. Trees grown in WC amended soils have shown significantly better growth since planting, with significantly higher yields in 2023 in the 5th leaf.

# Improving Non-Fumigant-Based Approaches For Management of Almond Replant Problems

PROJECT NO: PATH1

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## Summary

During this project, replant trials were concluded near Chowchilla and at California State University Fresno (CSUF) and continued at the Kearney Agricultural Research and Extension Center (KARE) to test rice bran (Rb)- and ground almond hull and shell (Ahs)-based alternatives to soil fumigation. Depending on the treatment, Rb and Ahs were used as soil amendments only or as “drivers” for anaerobic soil disinfestation (ASD). Almond whole orchard recycling (WOR) chips were tested as a soil amendment. Rb, Ahs, and WOR chips were shallowly incorporated at 9, 9, and 60 tons/acre (t/ac) treated, respectively, while the fumigants, 1,3-dichloropropene plus chloropicrin, were injected deeply by shanks (332 plus 200 lb/ac, respectively) (all rates were per treated acre of row strips, which comprised ~50% of orchard land area).

Costs were estimated for the Rb, Ahs, and fumigation treatments. Amendment-only treatments with Rb and Ahs were estimated to cost \$1,271 and \$ 947 per orchard acre, respectively, whereas ASD treatments that used Rb and Ahs with water and tarp applications (Rb+WT and Ahs+WT, respectively), were estimated at \$2,191 and \$1,867 per acre, respectively. Cost of the strip fumigation was estimated at \$1,443 per acre without tarp.

At KARE, by the third growing season after planting, preplant ASD treatments Rb+WT and Ahs+WT and the amendment-only Rb treatment increased ‘Nonpareil’ kernel yields by factors of 2.9, 2.0, and 2.1, respectively, and increased ‘Monterey’ yields by factors of 4.5, 3.1, and 2.2, respectively (compared to the non-treated control). Preplant soil fumigation increased ‘Nonpareil’ and ‘Monterey’ kernel yields by factors of 2.1 and 3.6, respectively. WOR amendment had small effects on tree growth and yields. At CSUF, ‘Shasta’ kernel yields accumulated from 3 years of harvest were increased by preplant Rb+WT, Ahs+WT, and soil fumigation by factors of 1.8, 1.8, and 2.2, respectively, compared to the control.

Three first-year postplant fertilizer programs were tested for added benefits to the preplant treatments at KARE. Extra nitrogen (N) in the first growing season (5.5 oz/tree N) generally did not increase tree growth or kernel yield by first harvest, compared to a standard first-year program (3.5 oz/tree N). However, adding phosphate (2.2 oz/tree P) to the standard N program improved tree growth; kernel yields were increased by factors of 1.2 to 1.3, compared to the standard N program. The fertilizer treatment effects were generally the same across all preplant soil treatments, suggesting broad applicability to replant settings.

Net revenue impacts were assessed for selected preplant treatments. By the first year of harvest at KARE, none of the preplant treatments had generated net revenue increases, compared to the non-treated control. However, given typical yield progressions as orchards develop, Rb, Rb+WT, and fumigation treatments were likely to increase net revenue, compared to the control, by the second or third year of harvest. With ‘Shasta’ at CSUF, by the third harvest, accumulated net revenue increases for Rb+WT, Ahs+WT, and preplant non-tarped soil fumigation treatments, compared to the control, were \$1,899, \$1,715, \$4,348 per acre, respectively.

Statewide monitoring of Phytophthora disease incidence on almond was completed in partnership with UCCE Farm Advisors with almond responsibilities. Our findings were: (1) multiple species of Phytophthora, some relatively “new” to California almond production, were contributing almond tree death in young orchards, especially *P. mediterranea* and *P. niederhauserii*; (2) stocks from multiple nurseries have sometimes been infested with Phytophthora species; (3) some hybrid almond rootstocks are relatively susceptible to Phytophthora crown rot; and (4) in some young orchards suboptimal soil water management contributed to infection by Phytophthora. Also, perennial Phytophthora canker disease, which results in tree death from scion invasion by the pathogens, was a continuing problem in Kern County.



# Effect of Partial Substitution of Fertilizer with Organic Matter Amendments on Nutrient Cycling

PROJECT NO: PREC7

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**Co-PIs:** Vivian Wauters, Amelie Gaudin, Patrick Brown, Michelle Perez

## Summary

The impact on environmental quality and profitability of synthetic fertilizers in California almond orchards may be lessened by the use of organic matter amendments (OMA). In the short term, OMA can act as partial substitute to synthetic nitrogen (N) fertilizer while over the long-term increase soil organic matter and other essential nutrients like phosphorus (P) and potassium (K). Our OMA compost trial with composted manure and green waste compost was split into two N treatment starting in 2019; an Intensive rate at 180 lb N ac<sup>-1</sup> (120 units from fertilizer and 60 units from irrigation nitrate) and a Reduced rate at 135 lb N ac<sup>-1</sup> (75 units from fertilizer and 60 units from irrigation nitrate) in plots previously amended with the same OMA sources since 2015. We measured leaf nutrient status, N availability, soil organic matter, and almond yield. This project also leveraged these ABC funds as a match to test almond hulls and shells as an OMA using FFAR and WSARE grants to host new trials in San Joaquin and Yolo Counties. Hull and shell trials started in 2020 where we measured leaf nutrient status, soil exchangeable K, soil microbial composition and tree water stress over time. This final report includes data through 2023.

In 2019, 2020, 2021, 2022 and 2023, we collected almond yield across all plots in the compost trial. There were no significant differences between amended and unamended plots at both N rates. On average, yields under composted manure at the intensive N rate were the highest followed by the unamended control and then green waste compost at both N rates. Challenges from this project include high irrigation nitrate inputs of up to 60 lb N ac<sup>-1</sup>. This additional N may be reducing the differences in the N rates leading to a range that is too narrow to accurately quantify potential differences in cumulative N availability and almond yields. This five-year data set shows that OMA use has had no negative impact on overall orchard performance, but green waste compost use combined with lower N fertilizer rates may impact tree performance.

Furthermore, we observed greater leaf N, P and K concentrations for both OMA sources compared to the control. In 2021, soil moisture sensors measured higher volumetric water content at 15 cm depth in the composted manure plots compared to the control. In 2021, we also measured cumulative N availability across all plots. There were no significant differences between amended and unamended plots at both N rates suggesting much of the excess N is being incorporated into soil total N. Soil organic matter was significantly higher for both OMA sources compared to the control reported as SOC and TN concentrations from 0 to 10 cm.

Funding from this project matched our proposal on almond hulls and shells as a soil amendment, K availability, and the role of off-ground harvest in facilitating soil health practices. There were no statistically significant differences in leaf N, P, Ca, S, B, Zn, Mn, Fe, or Cu. In the amended trees, leaf K was significantly higher. Soil exchangeable K in hull and shell plots were significantly greater compared to the control from 2019 through 2021. Stem water potential was significantly less negative in the hull and shell treatments than the unamended control in 2021. These results show that OMA sources did not improve almond yields, but do affect soil fertility, moisture and microbial composition, as well as tree nutrient and water status. Overall these studies as a whole demonstrate benefits to orchard soil and tree conditions, but do not significantly increase almond yields.

# **Cross-Project Approach to Accelerate Multidisciplinary Research on the Interaction of Nonstructural Carbohydrates (NSC) With Biotic and Abiotic Stresses Management Practices and Varieties in Assessing NSC's Dynamics Impact on Yield - an Ongoing Carbohydrate Observatory Project.**

PROJECT NO: PREC8

## **Principal Investigator:**

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## **Summary**

The goal of the project is to use the Carbohydrate Observatory analytical capacity to develop, coordinate, and implement a cross-project analysis of NSC for ABC funded projects to accelerate our understanding of the NSC role in almond production and to provide support to individual projects' principal investigators. Specifically, we developed the chemometric technique to reduce cost of NSC analysis to fraction of the enzymatic digestion technique. We are following and developing better understanding of environmental impact (loss of chill hours, wildfire smoke, rainfall) on NSC content on almond trees and analyze physiological long term-repercussion on yield potential. In addition, we provide a no-cost analysis to multiple projects supported by ABC.

# How to Irrigate Almond Orchards - for the Current Year's Expected Yield or for Maximum Yield Potential?

PROJECT NO: WATER17

## **Principal Investigator:**

Name: Maciej Zwieniecki

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## **Summary**

**Main Goal:** Determine the impact of matching irrigation to the spring estimates of the current year's yield on final yield and the following year's yield potential. The development of an accurate yield prediction model (YPM) that accounts for specific characteristics of the orchards (age, variety, location, management plan) and stochastic environmental conditions (precipitation, temperature, frost, persistent air pollution, pollination, etc.) requires large-scale, multi-year datasets that capture most of the variability. We are collecting data from multiple orchards across the Central Valley, including hourly temperature, precipitation, and spectral images from Google Earth Engine datasets; bloom time using a lab-developed satellite-based model; air pollution data from MODIS satellite OD47; non-structural carbohydrates (NSC) from the Carbohydrate Observatory; orchard parameters (variety, age, location); irrigation data and actual yield data from 152 orchards. This data collection spans from 2019 to the present, allowing us to account for various climatic scenarios and environmental perturbations.

This year, we have focused on developing a yield prediction model for almond orchards, incorporating orchard-specific characteristics (age, variety, location, management plan), environmental conditions (precipitation, temperature, frost, pollution), and tree biology (non-structural carbohydrates, phenology). This model requires extensive data to account for year-to-year climatic variability and perturbations (smoke exposure). Specifically, this year, we incorporated bloom prediction model to improve the accuracy of yield predictions. Using ground observations and Sentinel-2 imagery, we developed the Enhanced Bloom Index (EBI) and Normalized Difference Vegetation Index (NDVI) to measure bloom intensity and greenness. From 2019 to 2022, our method achieved a mean absolute error of 1.9 days in detecting peak bloom, demonstrating the potential of satellite-based monitoring for agricultural management.

Almonds are threatened by warmer winters, which can disrupt the necessary accumulation of chill for productive blooms. Using our bloom prediction model based on carbohydrate metabolism (Sperling et al.2019), we estimated the impact of past and future climate on bloom timing and success. We made progress in the analysis of irrigation's impact on yield, finding a significant correlation between end-of-season water deficit and yield. A 300 mm water deficit was associated with a 400-pound yield penalty. We aim to refine this model by incorporating more data and exploring various time series models to account for historical conditions. However, our progress was affected by the discovery that prolonged wildfire smoke exposure (years 2020 and 2021) reduces the accumulation of carbohydrates prior to dormancy and future yields, thus requiring incorporation of such stochastic perturbations into the analysis of the impact of management practices on yield.

# Determining Almond Tree Water Use and Stress using Surface Energy Balance Models with Unmanned Aircraft Systems

PROJECT NO: WATER16

## **Principal Investigator:**

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**Co-PIs:** Andrew McElrone, Bill Kustas, Kyle Knipper, Nicolas Bambach

## **Summary**

This ABC project focuses on the estimation of water use and stress in almond orchards using unmanned aerial vehicles, ground measurements, and validation sources (Eddy Covariance, leaf water potential, other measurements). The project aims to demonstrate the advances in drone technology (avionics, sensors, and data collection protocols) to surface energy balance components, towards the estimation of consumptive water use (Evapotranspiration) and stress (by separating the almond tree transpiration component from ET). The project methodology is based on the extensively used Two-Source Energy Balance Model (TSEB) model and previous multiyear experience in commercial vineyards across California. This ABC project supports components of the US Department of Agriculture Agricultural Research Service T-REX (Tree-crop Remote sensing of Evapotranspiration eXperiment); an umbrella project that aims to integrate ground, sensors, drones, and satellite data to provide a comprehensive integration of information on consumptive water use and stress for commercial almond sites across California Central Valley. 2023 was Year 3 of the project, with significant data collection efforts and analysis of the collected information in coordination with the T-REX partners including the University of California – Davis, USDA – ARS scientists and Utah State University AggieAir UAV program at the T-REX.

In terms of data collection, 2023 UAV data collection was focused on two of the sites, namely Vacaville and Woodland, near the Sacramento area. For this year, upgrades on the AggieAir technology and FAA permissions allowed expand the surveyed acreage and time in the air of the camera sensors, expanding from 200 to more than 350 acres every 30 minutes and from 30 mins of flight time to 60 mins (allowing two consecutive surveys per UAV flight). In addition, we incorporated a new image sensor on board to the AggieAir fixedwing (AgEagle AltumPT + Dual-Camera). The first camera allows the development of vegetation indices along with thermal measurements (for evapotranspiration estimates), while the Dual-Camera allows to approximate the spectral signature of almond vegetation and ground, opening opportunities to explore semi-hyperspectral analysis for phenological as well as stress conditions.

In addition, this year, similarly than 2022, we adapted air temperature and humidity sensors to be flown over USDA SAWS and AggieAir multicopter drones to collect atmospheric profile of temperature, useful for complementary USDA efforts to model and account advection conditions and its influence over Evapotranspiration estimates by satellite and drone information.

Regarding drone data research, we have evaluated the ground information acquired by the T-REX team and already published Leaf Area Index (LAI) models in almonds, to propose an update to the estimation of LAI at a single almond tree space. This update takes advantage of the pixel information detail (2 inches/pixel) provided by the AggieAir drone information. Using information collected across T-REX sites, a new LAI model that uses NDVI and tree fractional cover has been proposed and validated across the T-REX sites. This analysis goes along with an assessment of drone flight time conditions for an adequate characterization of the surface conditions (illuminated / shaded conditions in the farm) and estimation of water use (evapotranspiration).

# Remote-Controlled Evaluation of Distribution Uniformity and Stem Water Potential: Extending Imagery to Integrated Decision Support

PROJECT NO: WATER18

**Principal Investigator:**

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**Summary**

The Almond Board of California (ABC) has established an industry goal to reduce the water used to grow a pound of almonds by 20% by 2025. The simplest path for achieving this goal on time is by optimizing irrigation application and scheduling through energy-efficient irrigation scheduling using stem water potential (SWP) at key stages (“when to start”, hull-split, “when to end”), adjusting irrigation in response to actual evapotranspiration, and increasing distribution uniformity via improved system testing, maintenance, and application flow metering. This project addresses obstacles and knowledge gaps through new remote sensing tools, outreach, and integrated irrigation decision support to transform thermal, multispectral, and hyperspectral imagery to actionable information. As a part of the larger Tree Crop Remote Sensing of Evapotranspiration Experiment (T-REX), our team conducts applied remote sensing missions using off-the-shelf UAV platforms on a frequent basis at T-REX eddy covariance tower locations. Our team is using these data to develop and validate high-resolution actual evapotranspiration and energy balance maps at the tree scale. Our high-resolution models continue to demonstrate that off-the-shelf UAV platforms can provide maps of high-resolution actual evapotranspiration at acceptable and actionable levels of precision and accuracy to inform smart irrigation in California almonds.

Our overarching goal is to address research gaps, provide training, and develop decision support tools for CA growers and almond industry and water data service providers (e.g., CCAs). Key milestones for year 3 include: (1) We concluded data collection for water stress component of this project that will develop easy to use, early indicators of water stress using hyperspectral and thermal imagery validated by comprehensive physiological data (SWP, Florapulse sensors, photosynthesis) from the almond variety and delayed irrigation trial in Butte County. We submitted two studies related to the SWP work. Additionally, our preliminary results continue to show a connection and promise for using SIF, PRI, and other spectral indices to predict stem water potential using aerial approaches. (2) For T-REX, we continued to fly several UAV missions using our off-the-shelf platform in conjunction with Intensive Observation Periods (IOPs) at Butte, Woodland, and Ripperdan sites as well as a newly instrumented site in Westley, CA. In addition, we had our first T-REX article accepted for publication and developed a data fusion workflow that we will be continuing to refine and validate over the remaining project period. (3) We have been working with all T-REX flight data to develop a preliminary approach for identifying distribution uniformity problems using both UAV and satellite data to identify anomalies or changes within a field scene. This approach will be refined and tested with ground data from uniformity tests in 2024-2025. (4) We conducted a series of three irrigation extension clinics in Chico, Modesto, and Bakersfield in partnership with ABC where we focused on the fundamentals of irrigation management including evapotranspiration, scheduling, and managing soil infiltration and moisture with soil health practices.

# **Determining the Sensitivity of Fruit (nut) Set and Crop Load to Early Season Water and Carbohydrate Status, Developing a Water/Carbon Model for Fruit Set, and Evaluating Whether Low Set and Resulting Low Crop Loads Require Less Than Full In-Season Irrigation.**

PROJECT NO: WATER19

## **Principal Investigator:**

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**Co-PIs:** Maciej Zwieniecki, Andrew McElrone, Bruce Lampinen

## **Summary**

This was the first year of a study to determine whether deficit irrigation could be safely applied to almonds with a low crop load. Because stem water potential (SWP) is the most accurate measure of tree water stress, trees maintained in a well irrigated condition (about 10 to 12 bars SWP, except for hull split and harvest periods) were compared to trees maintained under moderate (about 18 bars) or severe (about 25 bars) water stress using daily irrigation. An important result in this first year was the demonstration that irrigation could be fully controlled using automated SWP sensors (FloraPulse). This appears to be the first successful proof-of-concept test of a fully automated, plant-based irrigation system. A target of 50% crop thinning was also applied to half of the trees in the moderate and severe stress treatments. Thinning was not found to be a factor influencing crop stress levels, but crop load was low and variable across treatments due to poor weather at bloom in 2023. Hence, this result will need to be interpreted together with future years results under more typical crop load conditions. For instance, it is possible that crop load did not affect the level of water stress because crop load was below a threshold for this effect. If this is found to be the case, then the results of this study will be used to identify this threshold. The poor weather at bloom was the result of substantial rainfall (10") that occurred in February/March, but because irrigation was based on SWP, this also allowed for a substantial delay in the start of irrigation in the control (mid-May) compared to a normal year, and a further delay in the moderate (mid-June) and severe (late-June) stress treatments. In addition to these delays, much less irrigation water was applied to the control (32") compared to a normal year, with remarkably low irrigation amounts required in the moderate (9") and severe (4") stress treatments. The fact that these water stress treatments had very little effect on yield must be interpreted with caution, because stress is known to have significant carryover effects, and these may not be apparent for 2-3 years. In addition to yield, starch levels in the wood were also not affected, although again, these effects may require time to be expressed. One unanticipated advantage of the plant-based daily deficit irrigation approach used in this study was the ability to continue irrigation during hull split and harvest without posing a danger to the harvested crop and apparently without predisposing the trees to shaker injury.

# Almond Variety Development

PROJECT NO: HORT1

**Principal Investigator:**

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**Summary**

The UCD almond breeding program has developed a series targeted genetic solutions for current and emerging challenges to California almond production and market expansion. Targeted traits include self-fruitfulness, improved water-use-efficiency and salinity tolerance, improved disease and pest resistance, and improved kernel quality and market value. Because genetic solutions are incorporated into new varieties and rootstocks, they offer unique economic and environmental advantages ranging from stand-alone solutions (such as self-fruitfulness to simplify orchard management including a reduced dependence on honeybee pollinators) to specialized tree and nut architectures to facilitate catch-frame harvest (as a strategy to minimize orchard dust and nut contaminations). Many traits, such as self-fruitfulness, were unavailable in traditional breeding germplasm requiring the breeding program to transfer desired traits from related germplasm, including related species such as peach. To make this newly enriched germplasm more widely accessible to public and private breeding programs we are currently working to fully document major breeding lineages and, concurrently, develop molecular markers for critical new traits such as self-compatibility, disease resistance and water-use-efficiency. Because the acreage of Nonpareil and its pollinizers have recently expanded to over 80% of all plantings driven by Nonpareil's high productivity and market value, the breeding program is also working to introduce new pollinizer varieties with improved market quality and productivity. Over 40 elite candidates have been planted in multi-year regional grower evaluation trials to identify and eliminate selections with serious deficiencies such as rootstock incompatibilities or susceptibility to Bud-failures, before any release for large-scale grower plantings.

# Regional Field Evaluation of New Almond Varieties & Selections – 3rd Generation

PROJECT NO: HORT2-3rd

**Principal Investigator:**

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**Co-PIs:** Luke Milliron, Roger Duncan

## Summary

The current Regional Almond Variety Trial includes 30 varieties, planted in the winter of 2014 in Butte, Stanislaus, and Madera counties. At all three locations, Nonpareil was planted alongside the test and standard varieties. The selected rootstocks at Butte, Stanislaus, and Madera sites were Krymsk 86, Nemaguard, and Hansen 536 respectively, with exceptions as listed in Table 1 of the full report. This trial consists of four replications of 11-12 trees of each variety or selection at each of the three sites. The peak bloom dates of all pollinizers in 2022 coincided with Nonpareil, except for the extremely early blooming variety UCD 3-40, which was dropped from the trial in the previous years. Despite three freeze events, yield for some varieties at the Butte trial were exceptional (4432 for Y117-86-03), however, yield was much lower for other varieties (1103 kernel pounds per acre for Supareil). In general, the bloom overlap has been good at all sites. Yields in the Salida trial continue to be moderated by irrigation water that is high in bicarbonates. More mature almond orchards grown under favorable weather conditions resulted in higher yield in 2022 than the previous year at the Madera site. In 2022 the most common kernel defects were doubles, twins, navel orangeworm, creases, discoloration, and mold. Varieties no longer under evaluation are UCD 3-40, UCD 1-232, UCD 1-271, UCD 1-16, Self-frP13.019, UCD 8-27, Self-fr P16.013, Y121-42-99, and UCD 7-159 due to poor yields, poor harvestability (high mummy counts), or insufficient replications.



# Regional Field Evaluation of New Almond Varieties & Selections – 4th Generation

PROJECT NO: HORT2-4th

## Principal Investigator:

Name: Roger Duncan

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## Summary

This project is the fourth generation of almond variety trials conducted by the University of California and sponsored by the Almond Board of California. The overall goal of this project is to enhance supply diversification by identifying better almond varieties that respond to different market demands and provide enhanced traits such as self-compatibility, pest resistance, and high yield efficiency. These trials will enable growers to make informed decisions when choosing varieties for their new orchards. The specific purpose of this project is to evaluate the most promising experimental almond selections from public and private breeding programs throughout the world and compare them to new and standard commercial varieties in side-by-side field trials in three California almond growing regions.

The field trials in Kern and Stanislaus Counties were planted in fall 2022 and the trial in Butte County was planted in spring, 2023. Therefore, there are no data to report yet. However, a few observations were made during the first leaf.

- Planting at the Stanislaus and Kern County sites went well with no problems and trees got off to an excellent start. At the Butte County site, trees were planted very late out of cold storage and approximately 20% will be replaced in spring 2024.
- The winter and spring of 2022-23 was very wet with several frost events. In the Stanislaus County trial, newly emerging shoots of many varieties died back or were killed by bacterial blast (*Pseudomonas syringae* confirmed by F. Trouillas lab). The most blast-affected varieties were Yorizane, Aldrich, Shasta, Vela, Florida, UCD B2 and UCD B15. A few trees had to be replaced but most recovered fully.
- Trees of UC Davis selections B6 and B11 (Stanislaus & Kern) and B3 (Kern County) had very poor bud break in spring 2023, where sometimes only one or two buds pushed, often only inside the carton or at the very top of the tree. The decision was made to remove UCD B11 from the experiment altogether due to extensive bud break problems. Trees were replaced with industry standards Monterey or Fritz in the Kern trial, Bennett-Hickman in the Stanislaus trial, and Monterey on Viking in the Butte trial. Affected trees of B6 and B3 were left in the trials and retrained up the stake where necessary.
- Foliar symptoms of potassium deficiency on the Spanish variety 'Constanti' became evident in the Kern County trial by midsummer in the first leaf. Potassium deficiency was confirmed by leaf analyses, indicating 1.1% K in leaves of Constanti compared to 1.8% in Nonpareil. Potassium deficiency symptoms were not evident on Constanti in the Stanislaus or Butte County trials

# Almond Culture and Orchard Management

PROJECT NO: HORT3

## Principal Investigator:

Name: Almond Workgroup Chair

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**Co-PIs:** Mohammad Yaghtmour, Mae Culumber, Franz Niederholzer, Katherine Jarvis-Shean, Phoebe Gordon, Franz Niederholzer, Cameron Zuber

## Summary

This project consists of four separate programs by four farm advisors working from Orland to Fresno.

### Project 1: Does adding phosphorus fertilizer increase almond yield?

Project leader: Franz Niederholzer, UCCE Colusa and Sutter/Yuba Counties

Project: Apply phosphorus fertilizer to a low soil P (3-7 ppm P by Olsen), double drip-line irrigated orchard planted in 2017 in Colusa County. Measure yield and leaf P to determine if adding P fertilizer increases yield.

Summary: Adding 70-120 lbs P<sub>2</sub>O<sub>5</sub>/acre over three years to Nonpareil, Aldrich, and Kester almond trees has not increased yield of any variety over that of unfertilized trees. A similar study adding 75 lbs P<sub>2</sub>O<sub>5</sub>/acre/year for two years did not increase Nonpareil yield, although soil P and leaf P levels increased significantly.

### Project 2: Monitoring soil C changes, overall soil health, tree growth and productivity in replanted orchards after recycling.

Project leader: Mae Culumber, UCCE Fresno County

Project: Chipping old orchards and incorporating the chips before planting a new orchard allows orchard renewal under air quality-based burning bans. What does adding 25-85 tons of wood chips mean to soil health and nutrient availability? Is mixing tens of tons of wood chips per acre helpful or harmful to soil health and tree growth and production?

Soil samples (0-6") along with leaf samples and yield data have been taken 2020-2022. In addition, soil nitrogen (N) availability and total content have also been measured along with soil organic matter and readily available carbon. Summary: Yield and tree growth data are weakly correlated to soil health measurements, but the trees have not yet reached full production. Measurement will continue. Soil health metrics were strongly associated with WC treatments across all sites. Locations with a WC amendment rate > 50 tons / ac had significantly higher values for all indicators in comparison to conventionally (CN) prepared soil in all categories, while the lowest 35 ton/ac application only increased soil organic matter levels. Notably, WC treatments resulted in significantly higher bioavailable N and total N suggesting WC creates conditions for N retention for a longer period in the soil which may enhance tree uptake, nitrogen use efficiency and minimize N losses to leaching.

### Project 3: Generating video content for extension efforts related to almond farming, production, and management

Project leader: Cameron Zuber, UCCE Merced County

Project: Two goals for project were 1) compile and collect short video to create a library to support ongoing extension efforts for almond production and 2) developing longer-form videos to be posted to web.

Summary: Overall, goals were accomplished as compilation, collection, and development were done though actual posting was delayed as time commitments with editing were underestimated. However, videos are being finalized and will be posted.

### Project 4: Irrigation Continuum Workshops

Project Leader: Curt Pierce, UCCE, Glenn, Tehama, Shasta, and Colusa Counties

Summary This project proposed to develop and conduct a series of workshops based on best practices for drought management and the ABC's "Irrigation Continuum" publication. The irrigation workshops were proposed to be a series of three individual events, each offering instruction and CEU's in line with the three stages of irrigation management outlined in the Irrigation Continuum. Funds were to cover costs related to conducting the events: venues, equipment, materials, refreshments and lunch.

After the project was accepted by the ABC, Tom Devol of the ABC proposed a revision to the project that would instead offer three workshops statewide, followed by a collaboration to revise the "Irrigation Continuum" publication to better reflect the evolving needs of producers. Three workshops were subsequently held in Spring 2023. One in each region of the state north (Orland), central (Modesto), and another south that was not a part of this project. Advisor PIERCE co-organized the North and South workshops with Tom Devol of ABC and presented at both events. Excess funds from the first stage were used to purchase tree and soil moisture measuring components, both for use in the trainings, and subsequent use in assisting growers during farm visits.

# Field Evaluation of Almond Rootstocks in the Southern San Joaquin Valley

PROJECT NO: HORT4

## Principal Investigator:

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## Summary

Trees were planted on October 22, 2019, in fumigated soil. The experiment is part of a replanted orchard on a sandy loam soil where orchard recycling was performed. Also, high winds in this part of the valley are one of the biggest challenges. Trees continued to grow well, and a significant difference in tree circumference was observed among the different rootstocks. While differences in tree trunk circumference between a few rootstocks are no longer statistically different compared to the early period of orchard development, the previous trend is still the same as the orchard is moving into the production phase. Rootstocks Flordaguard x Alnem and peach-almond hybrids except for Cornerstone expressed the biggest growth; and Krymsk 86 had the smallest trunk circumference among all rootstocks. Growth of Flordaguard, resistant to a population of peach root-knot nematode, was not statistically different from other peach-almond hybrids such as Hansen 536 and Bright's hybrid 5. Furthermore, the yield trend followed the tree size as expressed by tree circumference with trees grafted on Viking, Krymsk 86, and Rootpac R producing the least yields. Furthermore, Krymsk 86 had the highest number of kernels per ounce, which will correspond to smaller kernels.

Mid-July leaf analysis of Nonpareil shows that most of the trees in the orchard had nitrogen level below the adequate range values of 2.2-2.5% except for trees grafted on Krymsk 86, BB106, and FXA rootstocks. Significant differences in leaf nitrogen and potassium levels were detected, and phosphorus levels were not statistically different between trees grafted on the different rootstocks. We have not noticed any deficiency symptoms on the trees or any other negative effect on tree yield and general health. Chloride and sodium leaf content in 2023 shows chloride accumulation in leaves of trees grafted on Krymsk 86 or Flordaguard. This is less than the results in earlier years, however, sodium was not detected in leaf samples on average except in very low amounts in few peach-almond, peach and peach hybrid, and Rootpac R rootstocks. Both elements are below the critical values to cause leaf symptoms and affect tree health. Furthermore, no significant differences in hull boron content between the different rootstocks were detected with most values below the critical value. Unlike 2022, we did not detect any significant differences in midday stem water potential among the different rootstocks before harvest in 2023. In 2022, trees grafted on rootstocks showing the biggest tree growth as measured by tree trunk circumference had the biggest stress compared to the trees with the least tree growth. We have also evaluated the progression of hull split for Nonpareil on the different rootstocks during the 2023 season. Unlike in 2022, We did not see obvious differences in hull split initiation and progression except for trees grafted on FXA rootstock showing delayed hull split progression compared to the rest of rootstocks. Furthermore, we measured tree anchorage. Trees grafted on Krymsk 86, Rootpac R, Hansen 536, and Viking, are most the straight trees, while rootstocks Empyrean-1, BB106, Flordaguard, and Cornerstone were leaning the most in the orchard.

# Nickels Soil Lab Projects

PROJECT NO: HORT6

## **Principal Investigator:**

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**Co-PIs:** Bruce Lampinen

## **Summary**

The Nickels Soil Lab (NSL) is a privately owned, commercial orchard SW of Arbuckle in Colusa County. It was established with a bequest from Mr. Leslie Nickels to conduct applied research in support of local agriculture. While the property is owned by the Nickels Trust, UC ANR coordinates research at the site under an MOU with the Trust. A total of 160 acres on 2 properties constitute the NSL; 80 acres of almonds, 70 acres of open ground, 8 acres of walnuts, and 2 acres of olives. [29 acres of older almonds and 7 acres of older walnuts were removed in June, 2023.] There are 11 on-going projects at NSL conducted by UC and USDA personnel. The operating budget for the Nickels Soil Lab comes primarily from crop sales. The Almond Board of CA provides regular support for two projects, providing extra funds for non-commercial projects. The Almond Board also provides critical support for large ticket items (irrigation infrastructure) when market conditions make it impossible for NSL to pay for the project. This report covers three projects at NSL; 1) organic practices demonstration, 2) production and health comparison of Independence and Nonpareil/pollinizer plantings, and 3) in-row spacing trial on two different rootstocks.

In 2023, overall almond yield at Nickels Soil Lab was off from the excellent crop in 2022, due to alternate bearing (off year) in the older blocks (50% of acres) and very bad navel orangeworm damage, despite winter sanitation and insecticide sprays. Organic yield was 70-80% of conventional. Yield reduction appears to be due to conditions other than differences in organic and conventional practices. Deficient N levels are a continuing challenge in the organic block as organic N sources are extremely expensive. Independence vs Nonpareil+pollinators results showed equal yields in Independence, Aldrich, and Nonpareil while Sonora had another poor crop. This may have been related to extended by weather following full bloom. No difference existed between gross income/acre for Independence vs Nonpareil +pollinizers orchards. This comparison excluded differences in costs as well as insect damage. In the spacing trial, the Titan peach/almond (P/A) hybrid rooted block (7th leaf) and the Rootpac-R plum/almond hybrid (PI/A) rootstock planting (6th leaf) had very good years. All plantings in this block are 50% Nonpareil, 25% Aldrich and 25% Kester. No significant yield differences with tree spacing were measured for Kester on Titan or Rootpac-R rootstock at all tree spacings (12', 14', 16', or 18', all at 21' row spacing). Nonpareil on Titan and Aldrich on Rootpac-R showed significantly greater yield at closer spacing (12 or 14') compared to wider spacing (18').

# Three-Dimensional Model-Based Analysis of the Impact of Variability in Almond Tree Structure and Configuration

PROJECT NO: HORT45

## Principal Investigator:

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**Co-PIs:** Thomas Gradziel, Ted DeJong, Bruce Lampinen, Eric Kent

## Summary

This project seeks to develop and use state-of-the-art modeling tools to quantify and predict interactions between traits (phenotypes) and environmental conditions in the context of almond production. The scope of this project focuses more specifically on tree growth, structural development, and yield dynamics as it pertains to canopy configuration and architecture. The modeling framework represents tree development in three-dimensions, explicitly representing every leaf and branch as it evolves in time. This approach allows for direct representation of the effects of inter- and intra-crown competition for resources such as light, and how that competition drives shoot growth and spur dynamics.

Field measurements of structure and leaf physiology were collected on four different varieties (Aldrich, Nonpareil, Independence, Sonora) with varying tree architectures (upright, balanced, sprawling) to aid in model testing and calibration. LiDAR scans were also collected at a long-term configuration trial near Oakdale, CA where within-row tree spacing, pruning practices, and rootstock were varied. A framework for modeling the 3D structure of shoots during growth has been implemented and is being extended to full tree structural development.

We used the model to explore several illustrative orchard configurations that were hypothesized to improve orchard productivity or efficiency. The first was whether using an upright variety in conjunction with Nonpareil trees could increase profitability relative to an orchard with all varieties having similar growth habit as Nonpareil. It was hypothesized that by allowing Nonpareil trees more space to grow, it would increase their yield which is more valuable than pollinizer nuts. Simulations indicated that this was unlikely to occur in practice. At best, productivity of the Nonpareil trees stayed the same, and the overall orchard productivity and water-use efficiency was either similar or lower relative to a comparable orchard with all varieties having similar structure as Nonpareil. The best performing upright-Nonpareil case had East-West rows and reduced in-row spacing of the upright trees, but overall profitability is likely similar to a more uniform orchard.

We also examined a similar case in which a compact variety was interplanted with Nonpareil trees, with the logic that the smaller pollinizers would allow Nonpareil trees more space to grow and thus increase profitability or efficiency. While light interception of the Nonpareil trees did increase slightly, their photosynthesis was about the same, and water-use efficiency declined. Provided that the in-row tree spacing of the compact trees was reduced, whole-orchard productivity could be maintained. An East-West row orientation provided the biggest benefit to the Nonpareil rows.

Finally, we examined the hypothetical orchard configuration consisting of a potential (yet to be developed) self-compatible upright variety planted at high density, which could be viable with off-ground harvesting technology. The logic was that, since light penetration to the orchard floor for drying is not necessary in this case, upright varieties will allow more light interception and productivity while maintaining adequate row spacing for equipment access. Simulations suggested that the upright structure could provide a slight increase in light interception, photosynthesis, and water-use efficiency relative to a comparable orchard with the same leaf area index (LAI) and row clearance between adjacent rows but a shorter crown.

Further work is needed to incorporate longer-term growth and spur dynamics into the simulations, as current results use light interception and photosynthesis as a proxy. The framework for accomplishing this goal has been put in place over the past year, and development and integration will continue over the next year to include these processes.

# The Application of Molecular Tools and Quantitative Phenotyping for Genomics-Assisted Breeding in Almond.

PROJECT NO: HORT46

## **Principal Investigator:**

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**Co-PIs:** Thomas Gradziel, Andreas Westphal, Julia Stover-Blackburn, Diane Mather, Michelle Wirthensohn, Pedro Martinez-Garcia

## **Summary**

This is the final year of a project to provide genomic and phenomic support for the UC Davis almond breeding program. Gina Sideli led this project for four years before accepting a faculty position at Rutgers, and Pat J. Brown is leading the final year of this project. Activities in the final year include completing bioinformatic analyses of data generated by Dr. Sideli, as well as generating additional genomic resources (genome assemblies and sequence data) to support future genomics-assisted breeding efforts in almond. It is anticipated that a future proposal combining genomics, breeding, and biotechnology will be assembled by Drs. Brown, Martinez-Garcia, and Gradziel.

Dr. Sideli's accomplishments during the first four years of the project are substantial. Stable quantitative trait loci were identified for shell hardness and lipid stability; molecular markers were then developed, tested and validated for these two traits. Marker sequences are available in the publications and will be added to the public database on the Fruit and Nut Center website. Using phenology data estimated from an unmapped aerial vehicle (UAV), genomic prediction models for forecasting almond bloom period were also developed and validated. Pedigree analysis for the UC Davis almond breeding program was completed utilizing the newly developed Arbor Biosciences exome sequence genotyping probes. Rootstocks with two years of data for root knot and lesion nematodes were sampled for a transcriptome experiment using TagSeq method, and analysis is underway to reveal genes that are differentially expressed between nematode-resistant and susceptible rootstocks.

# Are Californian Almond Cultivars and Rootstocks Susceptible to PPV and Can Almonds be a Host for the Spread of Sharka in California?

PROJECT NO: HORT48

## Principal Investigator:

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## Summary

Plum pox virus (PPV) is responsible for Sharka disease, which is one of the most important limiting factors for Prunus production (apricot, plum, prune and peach) in affected areas.

PPV was detected in the USA in 1999 and was declared eradicated by the USDA in October 2019.

Despite this official declaration of eradication, Sharka is still of great significance. It still requires major quarantine efforts, and millions of dollars have been spent on it, even though it has never been described in California.

Studies about Sharka on Californian almond cultivars and rootstocks are scarce, and the behavior of these cultivars and rootstocks against Sharka remains unknown. Several years ago, our own results showed a limited potential role for almond as a virus source in Sharka epidemics. However, Sharka is still a global threat for areas of stone fruit production including almond. So far, after 44 months of working on the project, we have completed testing 19 almond rootstocks against PPV, and 23 almond cultivars are still under PPV phenotyping.

The results obtained show that most rootstocks included in our assays are susceptible to sharka, displaying strong symptoms of the disease. Only two rootstocks—the interspecific hybrid 'GF677' and the almond seedlings 'DryStock One'—have proven to be fully resistant. These results point to the significant role that rootstocks could play in a potential outbreak of the Sharka disease in California. Regarding the almond cultivars, most are resistant, although at least ten tested positive by RT-PCR, without showing Sharka symptoms so far.

# Discovery of Genetic Variation in Related Self-Fertile Species of Almond

PROJECT NO: HORT49

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## Summary

The almond breeding program at the University of California at Davis is pursuing the development of new California adapted varieties solving current and emerging needs. These include mainly self-fruitfulness, improved disease and pest resistance, improved water efficiency and adaptation to a changing climate. In the long term the program has developed advanced introgression lines transferring self-fruitfulness from multiple independent sources including *P. webbii*, cv. 'Tuono', *P. mira*, and *P. persica* (peach). Other relatives (species) such as *P. fenzliana* or *P. argentea* have been used as a source for improved tree architecture or other traits (e.g. amaretto flavor market.) rather than self-fruitfulness. Several factors affect the breeding efficiency for self-fruitfulness as the degree and stability of the trait and the tedious and time-consuming backcrossing required for successful trait introgression. Although, a few low-density markers methods for marker assisted breeding have been used and a large number of promising new germplasm is being developed, the number of improved self-fruitfulness cultivars belonging to the almond breeding programs in California is low.

Advances in high-throughput sequencing technologies provide a great opportunity to bring molecular breeding to full application in the almond breeding program at the UC Davis. Molecular breeding is based on the efficient selection of genomic variants known or hypothesized to be associated (tightly linked) with alleles with superior phenotypic effects. A robust breeding program requires knowledge of the many alleles at each genetic locus in the *Prunus* gene pool. Many allelic variants will have a different effect depending upon the desired phenotype, such self-fruitfulness. These allelic variants are largely caused by or tightly linked to single nucleotide polymorphisms (SNPs), the most abundant genetic variations within the genome.

New advances have been achieved in the genetic characterization of S alleles in the founders, resolving inconsistencies, and confirming the self-compatibility introgressed from related species. The SFB genotyping results show allelic polymorphisms, supporting the self-compatibility observed in the breeding results. Additionally, the genome assembly of 27 founders has been completed, providing insights into genetic relationships and clades. Further analyses are recommended to ensure current and future breeding progress toward obtaining a self-compatible Nonpareil using the current self-compatibility sources in the program.



# Field Screening of Size Controlling Rootstocks for Off-Ground Harvested Almond Orchards

PROJECT NO: HORT50

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**Co-PIs:** Giulia Moreno, Chuck Fleck, Sarah Paris, John Slaughter, Grant Zaiger

## Summary

Some current off-ground harvesting systems for almonds require smaller stature trees to accommodate machinery that surrounds each tree and extends over the treetops. Conventional almond rootstocks likely have too much vigor and will require extensive, annual pruning to maintain trees small enough to accommodate similar machinery. This will likely increase cultural costs, reduce yield potential, and decrease the productive lifespan of the orchard.

Dwarfing rootstocks used in other stonefruit industries may not be compatible with almond. They may also exhibit different growth characteristics or may not be suitable for almond farming systems. In UC trials, lower vigor rootstocks have been less productive per acre and per canopy area than high vigor stocks under current, medium to low density almond farming systems. It is unknown if yields of low vigor rootstocks could be comparable to higher vigor stocks if planted in a super-high density system.

A replicated field trial was established at the UC Kearney Agricultural Research and Education Center in Parlier, CA. Sixteen experimental, purportedly dwarfing rootstocks were planted in fall, 2020 or 2021 and are being compared to industry standards Nemaguard, Viking, and Bright's 5. These experimental rootstocks include Controller 6, Controller 9, and FLxK2 (University of California), Citation, New Root 2, and 266LZ4 (Zaiger Genetics), Rootpac 20 (Agromillora), MP-29 (USDA), D63.182 and K37.068 (Wawona), Hybrid 3776, DA6, and Hybrid 29 (Sierra Gold), and ATAP, TRIO 25-07, and TRIO 22-07 (Fowler Nursery). Rootstocks are being monitored for horticultural attributes including compatibility with Nonpareil and Monterey scions, vigor, suckering, anchorage, and productivity.

Incompatibility symptom expression often differs from variety to variety. Many of the experimental rootstocks in this trial showed mild to severe signs of incompatibility, (early-mid season cessation of growth, rolled leaves with marginal necrosis and interveinal chlorosis, premature defoliation, and sometimes tree death), with one or both almond cultivars. Severely affected rootstocks included New Root 2 and 266LZ4 on which all or almost all trees of both varieties collapsed and died by midseason in the first leaf. Two thirds of Monterey trees on Citation died by the end of the third leaf while most Nonpareil trees had mild symptoms the first leaf but were essentially asymptomatic by the third leaf. About half of the Nonpareil trees on DA6 and Controller 9 have also died so far, with most of the rest continuing to decline, but Monterey trees on DA6 have shown minimal signs of incompatibility. Rootpac 20 has shown minor signs of incompatibility (mildly rolled leaves with interveinal chlorosis). MP-29 was the only fully replicated rootstock that produced a significantly smaller tree trunk through the third leaf without displaying obvious signs of incompatibility. None of the second leaf demonstration rootstock trees showed signs of incompatibility, but only trees on ATAP and TRIO 22-07 were numerically smaller than trees on Nemaguard.

Trees on Brights Hybrid 5 had the largest Nonpareil trunk circumference while trunk circumference of Monterey trees on BH5 were similar to Viking and Controller 6. MP-29, Controller 6, and D63.182 produced significantly smaller canopies than trees on Nemaguard for both varieties. Third-leaf yields for both Nonpareil and Monterey were highest on Viking, Nemaguard, Brights Hybrid 5, and Controller 6 rootstocks. DA6, Controller 9, and Rootpac 20 had the lowest yields. Yields for MP-29 and D63.182 were lower than Viking, Nemaguard, BH5, and Controller 6 but higher than Rootpac 20, Controller 9, and DA6. The most dwarfing rootstocks also had the smallest kernels while the highest yielding rootstocks tended to have the largest kernels.

# Accelerated Assessment of Almond Variety Candidates

PROJECT NO: HORT51

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**Summary**

The accelerated testing of advanced almond selections provides rapid detection, identification and early elimination of candidates that show unacceptable levels of regional vulnerability to important pests and diseases as well as historically important disorders such as Noninfectious Bud-Failure and rootstock-incompatibility that have a history of destabilizing almond production at both the grower and market level. Over 50 advanced UCD breeding selections are currently being evaluated in over 30 regionally diverse sites in the Sacramento and San Joaquin Valleys. Because most advanced selections have incorporated self-fruitfulness from related almond and peach germplasm, this advanced, California-adapted germplasm also possesses a wealth of new kernel and tree types with value for improved production efficiency and market expansion. These include improved water-use-efficiency and salinity tolerance, disease and pest resistance, and improved kernel flavor, roasting quality, and postharvest storability. The introduction and regional evaluation of new nut and tree traits is also facilitating the development of more economical and water-efficient orchard systems such as more compact yet productive tree architectures, catch frame harvesting and in-field hulling.

# Evaluating New Breeding Material for Salinity Tolerance in Almond Rootstocks and Exploring Novel Sources of Salinity Tolerance in Prunus Germplasm

PROJECT NO: HORT55

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**Co-PIs:** Jorge Ferreira, Don Suarez, Thomas Gradziel, Daniel Kluepfel

## **Summary**

The primary objective of our project is to breed new almond rootstocks that combine vigor, disease, insect resistance, and salinity tolerance. Dr. Tom Gradziel's breeding program at UC Davis has been pivotal, assessing multiple lines for traits such as growth performance and resistance to biotic and abiotic stresses. Our evaluation focused on salinity tolerance in these elite lines, categorizing hybrids by their trunk diameter's relative change, which ranged between 0.83 and 1.08. Notably, genotypes TG 34, TG 22, TG 26, TG 20, and TG 18 stood out for their robust salinity tolerance and lower leaf sodium (Na) and chloride (Cl) levels. Conversely, genotypes SG 100, SG 162, and SG 161, less resilient to salt, exhibited higher leaf ion concentrations. We also established the proline concentration ratio (salt/control) as a reliable biochemical indicator of salinity tolerance in Prunus. In the 2023-24 season, our research is broadening its scope to assess various variety selections along with a range of new rootstocks from our breeding program for their salinity tolerance.

# Resilient Prunus Rootstocks for a Changing Climate

PROJECT NO: HORT57

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## **Summary**

The primary function of rootstocks is to increase orchard performance by safeguarding trees from soil borne pests and diseases, provide tolerance to abiotic stress and expand into regions with different soil types. As California almond production is under considerable pest and disease pressures, as well as abiotic stresses resulting from a changing climate, the implementation of multiple breeding technologies, including marker-assisted selection, genetic transformation and gene-editing, provides a strategic pathway for almond rootstock development.

Genetic transformation is a breeding tool that can be utilized to introduce pest/disease resistance and abiotic stress tolerance genes into existing and/or newly developed Prunus rootstocks to safeguard almond orchards from biotic and abiotic stress. Plant transformation involves the transfer and integration of genes into the genome of a plant cell. Once transformed, a plant must be generated from the transgenic cell(s). Therefore, the first step in developing a plant transformation method is to establish a plant regeneration system(s). Plant regeneration systems are typically derived from developing organs, including leaves, stamens and stems. When a developing organ is removed from the plant and placed in tissue culture with the appropriate hormones, cell proliferation occurs at the cut site to give rise to dedifferentiated tissue termed callus. By manipulating the levels of auxin and cytokinin, two major phytohormones that regulate cell division and differentiation, as well as sugar, mineral elements and vitamins, proliferating callus cells can be induced to produce embryos or shoots to generate plantlets.

One of the major challenges with developing a plant transformation system in perennial crops, including almond, is the inability to regenerate plants from callus derived from developing organs. The inability to regenerate plants is often linked to the organ selected for callus production, chemical and hormone composition of the medium, as well as temperature and light conditions. Once a plant regenerative system is developed, the cells of the callus can be transformed using *Agrobacterium tumefaciens*, which transfer and integrate genes into the plant genome. In addition, particle bombardment is another method used to transform callus cells. The development of a plant transformation system will require the usage of selectable markers, including GFP and antibiotics. Taken together, generating genetically transformable almond rootstock callus with the capacity to undergo plant regeneration was the primary objective of this project.

In the HORT57 project we investigated methodologies for establishing a genetic transformation system for almond rootstock breeding and development. To develop a plant regeneration system, the project team successfully developed methods to initiate embryogenic callus from immature stamens and callus with a high regenerative potential from developing leaves. Unfortunately, we were unable to induce embryo germination and shoot development from callus produced from stamens and leaves, respectively. Results also showed that callus initiated from developing petals, stems and maternal seed tissues displayed a low reproductive potential. In this project, we also demonstrated that Nemaguard leaves could be transformed with *Agrobacterium* and particle bombardment. Further, the transformation efficiency was substantially increased when lipoic acid was used when leaves were transformed with *Agrobacterium*. Lastly, results suggest that Green Fluorescent Protein and hygromycin are suitable selectable markers that can be used to select for transformed plants. In a future project, it is recommended to use regenerative peptides and genes to induce embryo and shoot formation from callus derived from stamens and leaves, respectively.

# Comparing Root Traits and Depth Distributions for Mature Almond Rootstocks; is There a Link Between Root Architecture and Propagation Method?

PROJECT NO: HORT67

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## **Summary**

Propagation methods that do not use seeds (e.g., cuttings, tissue culture) have many advantages, such as uniformity of traits and a high rate of root production. However, one possible disadvantage of this method is the change in root architecture that can be expected when roots are generated artificially or develop as adventitious roots. Young trees that originate from cuttings or tissue culture tend to produce many roots, which helps increase transplanting success. All these roots originate close to the base of the sapling, which can potentially crowd out adjacent roots and cause cracking between roots as the tree matures. In addition to potential for increased cracking, clonally propagated root systems extend laterally from the base instead of vertically as would happen in a seedling. It is likely that shallower root frameworks do not develop as deep into the soil profile. More shallow root systems will need more frequent irrigation when evaporative demand is high (i.e., during summer months). This study is collecting field data to evaluate root traits of mature trees and a variety of rootstocks at three field locations as affected by propagation method. In addition, we will do a common garden experiment where we will plant both clonally propagated and seed generated rootstocks grafted with Nonpareil and expose them to control (adequate water) and drought conditions. Regular destructive collection of the trees will allow us to track root development as a function of genetic background, and assess the impact of water availability under field conditions.

# Improving Fruit Removal and Harvest Efficiency of Independence Almond Cultivar

PROJECT NO: OTR01

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## **Summary**

Incorporating an abscission agent into an almond early harvest strategy may improve harvest efficiency of some varieties and strongly benefit the almond industry by improving harvestability and reducing pest pressure from navel orange worm and hull rot. Improving harvestability may be particularly beneficial for the self-fertile cultivar Independence which has shown some fruit release problems. In this experiment, the ethylene precursor 1-aminocyclopropane -1 -carboxylic acid (ACC), commercially called ACCEDE (Valent BioSciences), was sprayed at two different doses (300 and 500 ppm) on fully and deficit irrigated almond trees. The effects of ACC on nut ripening (pull force and hull split), and its toxic effect (leaf drop, leaf chlorophyll, return to bloom) were evaluated at weekly intervals. We have found that the application of ACC accelerated ripening in both full and deficit irrigated trees, as did deficit irrigation alone. This agrees with previous studies that suggested ethylene involvement in the almond abscission process (Labavitch, 1987). Harvestability, evaluated as the ratio between the yield and the fruit that remained in the trees was very high in all treatments. An earlier harvest would be necessary to assess the treatments' effect on harvestability. The ACC application didn't produce significant toxicity. This is promising and in contrast to earlier Ethephon that produced gummosis and leaf drop (Labavitch 1987).

# Multi-Scale Evaluation of Stacked Regenerative Practices in Almond Systems

PROJECT NO: INSH05

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## Summary

Regenerative Agriculture as a production model is gaining momentum and aims to shift the emphasis away from just yield toward the management of a functioning ecosystem that actively restore soil health, biodiversity, ecosystems health and water quality while producing sufficient food of high nutritional quality (Grant 2017, Rhodes 2017). Almond orchards have potential to become flagship models for regenerative agriculture due to their perennial and low disturbance nature and the possibility to stack multiple practices in space and time with potentially low yield lags, if well managed. Practices such as compost and other organic amendments, whole orchard recycling, cover cropping and hedgerow plantings could form the backbone of regenerative almond systems. To date, these practices have been studied in isolation, with ABC funding and support, showing context-specific benefits to sustainability by allowing a decrease or substitution of inputs and/or increasing soil health and groundwater and biodiversity conservation with often no yield lags. However, the combination, or stacking, of multiple of these practices remains understudied, especially when considering multiple socio-ecological functions of relevance to growers and environmental integrity.

There is also considerable landscape-level variation that results in more granular constraints and opportunities to implement regenerative models across the vast almond growing regions. Landscape-level factors include edaphic conditions that affect the outcome of soil-focused practices, precipitation differences that impact non-irrigated plant growth potential, habitat connectivity for airborne insects, and land ownership that affects practice adoption and the level of individual and community impacts. Knowledge of the context specific feasibility and outcomes of these systems is therefore needed to better pinpoint opportunities, constraints and expected benefits for orchard multifunctionality. Our project is integrated with other research initiatives to explore both the within-field and landscape-level impacts of stacked regenerative practices in almond orchards across the CA Central Valley.

During this first year of the project, we implemented two complementary approaches:

- 1) Establishment of an on-farm demonstration site in collaboration with the East Stanislaus RCD with a controlled, replicated, multifactorial trial implementing three-way comparisons of establishment practices (conventional, Whole Orchard Recycling (WOR), and WOR + almond shell biochar), and soil management practice combinations (control (no-till + mineral N fertilizer) and soil health (control + cover crops + compost)). This year, the demonstration trial has been established along with baseline measurement of soils and tree carbon inputs.
- 2) Development of a gradient of growers' orchards across the two major growing areas of the Central Valley. We have connected with growers and identified 18 commercial orchards to compare outcomes across two gradients: a landscape potential gradient (defined by soil type, landscape heterogeneity and quality) and a within-orchard practice gradient (none, low, or high adoption of regenerative practices). Regenerative practices include compost and other OM additions, cover crops or resident alley vegetation and grazing. We have streamlined the team and methods to quantify orchard response to practice adoption with a focus on ecosystem functions, both as it affects the multifunctionality of the orchard systems, and the production-relevant aspects of the ecosystem that are of highest importance to growers.

# Avian Biodiversity in Almond Orchards of Central California

PROJECT NO: SUS01

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## Summary

This is a three-year study to assess biodiversity of bird fauna utilizing almond orchards during different seasons in the California Central Valley. Our study seeks to fulfill a gap in baseline knowledge of bird species utilizing almond orchards on an annual basis. While studies on bird diversity in almond orchards have been conducted in other places, like Australia, there is a deficit of data on the California Central Valley. The California Central Valley is an important stopover along the Pacific Flyway and serves as home to hundreds of bird species. We anticipate that this assessment of bird biodiversity in almond orchards will serve to quantify the value of these habitats to avian fauna, as well as seasonal differences in habitat use. We hypothesize that migratory song birds will use almond orchards as a stopover point in the early spring and fall during migration periods and there will be a population of non-migratory birds that reside in orchards year-round.

The study approach is to collect baseline data utilizing emerging technology of autonomous recording units (ARUs) to gather acoustic data of bird vocalizations. ARUs, when used appropriately, can provide an efficient, standardized, and unbiased data-collection procedure at lower cost than traditional site visits by skilled observers (Priyadarshani et al. 2018, Darras et al. 2019). ARUs are devices that record audio at user-defined times, sensitivity, and frequencies. The audio recordings are then digitized and processed with computer-aided signal recognition systems to identify recorded bird song to species. Isolating and identifying bird vocalizations can be a challenge because they are usually produced within a busy sonic environment. Background noise can include traffic, farm equipment, irrigation, wind, rain, and running water. Bird songs are extremely varied and complex, and when multiple species and individuals vocalize simultaneously, such as during the dawn chorus, elements of songs overlap in time, frequency, and amplitude. With ARUs, samples can be collected daily and year-round to provide a much more robust data set than can be achieved by intermittent traditional field surveys.

While it was anticipated that a baseline year of data would be collected during the first year of the study, unforeseen challenges with identifying willing almond orchard owners delayed deployment of the ARUs. Some orchards have been identified and ARU deployment is occurring in Year 2.



# Effects of Almond Adjuvants and Phytochemicals on the Synergistic Toxicity of Fungicides and Insecticides on Honey Bees

PROJECT NO: PATH21

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## Summary

When in bloom, almond orchards in California's Central Valley are visited by ~2.5 million colonies of *Apis mellifera*, the western honey bee, a number that represents ~70% of all U.S. commercial honey bee colonies. Although bees forage on the almond flowers for pollen and nectar, growers frequently apply fungicides to the flowers during wet winter and spring conditions. Despite the fact that these fungicides are considered safe for bees, beekeepers sporadically report significant losses, particularly during the larval and pupal stages, following fungicide applications. Factors other than direct toxicity of fungicides may be involved in this intermittent bee mortality. Bees often ingest pesticides, including fungicides, along with their phytochemical-rich floral diet, including pollen and honey. Most dietary phytochemicals as well as pesticides are detoxified primarily by cytochrome P450 monooxygenases, creating the potential for synergistic or antagonistic interactions between pesticides and phytochemicals. In addition, adjuvants have long been used with pesticides in almonds to enhance their efficacy by modifying their physical properties. The effects of adjuvants on non-target insects within the almond agroecosystem, particularly honey bees, are thus important to characterize.

This project aims to evaluate how specific almond-derived phytochemicals affect interactions of pesticides, adjuvants, and phytochemicals affect honey bee workers, particularly nurse bees, and queen quality in honey bee colonies. In several parallel behavioral experiments, we documented the effects of pesticides and adjuvants on nursing behavior. Co-administration of the fungus propiconazole and the insecticide chlorantraniliprole reduced both visitation time and nursing behavior in bees fed phytochemical-free sugar water. However, supplementation of sugar water with 250  $\mu$ M quercetin alleviated all impaired nursing behaviors across all pesticide treatments, with no statistically significant differences between treatment groups. In the absence of adjuvants and fungicides, worker bees fed control and almond honey (a source of phytochemicals) showed prolonged queen care behavior compared to those fed quercetin-laced sugar water. In addition, queen care behavior decreased in the presence of the adjuvant Dyne-Amic.

Path analysis consistently showed a reduction in total queen care behavior in the presence of Dyne-Amic. Additionally, the analysis revealed a positive relationship between almond honey and quercetin-laced sugar water with queen cell visitation time. Our study also demonstrated negative effects of consuming combined Dyne-Amic, propiconazole, and chlorantraniliprole on nurse bee olfactory responses to alarm and brood pheromones, suggesting disruption of social signaling. This disruption of social signaling may provide a mechanism to explain how insecticides and adjuvants affect the behavioral responses of nurse bees. The study findings were published in a peer-reviewed journal (Wu et al. 2023, <https://www.nature.com/articles/s41598-023-41818-7>).

In RNAseq analysis of queen ovary tissue, exposure to the adjuvant Dyne-Amic resulted in the downregulation of CYP450 genes (CYP9Q1-3, CYP6AS13, CYP6AQ1, and CYP336A1) and their partner cytochrome b5 involved in xenobiotic metabolism. Additionally, genes producing UDP-glucuronosyltransferases, critical for detoxification, were also downregulated. The downregulation of these genes, along with the juvenile hormone esterase gene, could impact detoxification, juvenile hormone levels, and energy metabolism in the queen ovaries. However, due to high inter-sample variation within each fungicide subset, only a few differentially expressed genes were identified, making it challenging to determine the effects of fungicides on queen gene expression. Further analysis, such as qPCR, may be necessary to clarify these effects.

Our project also characterized the phytochemical profile of almond pollen to identify unique compounds in almond pollen and their effects on bees. In collaboration with Washington State University, more almond pollen and beebread samples were collected for further experiments. Some almond beebread samples were sequenced to study the fungal communities in beehives in almond orchards in view of evidence that mutualistic fungi in these communities may contribute to xenobiotic detoxification (in prep.).

# Another Look at Pheromonal or Related Attractants for Leaf-footed Bugs (*Leptoglossus* spp.) Infesting California Nut Crops

PROJECT NO: ENT018

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**Summary**

Leaf-footed plant bugs (Hemiptera: Coreidae: *Leptoglossus zonatus*) (LFB) are a key pest of almonds, pistachios and pomegranates in California. The adults feed on new crop almonds, which can cause damage/discoloration to the kernel or even premature nut drop. This pest overwinters as an adult in large aggregations outside of the orchard, typically in sheltered areas. In the spring, adults will disperse from these aggregations and move into almond orchards, usually in early- to mid-April. Currently, monitoring for this pest involves beat sampling the tree canopy, as well as directly looking for adults and/or signs of crop damage (e.g. gumming). This approach is time and labor intensive, and in many cases is looking for after-the-fact signs of LFB activity. As such, we proposed the development of a trap and lure system for LFB based on male-produced pheromone compounds. Here we summarize our efforts over the past seven years.

Initial studies found that a hanging panel-trap was the most effective for monitoring LFB, and that it could be improved by adding fluon (a type of liquid Teflon) and changing the trap color to yellow. Analysis of the LFB pheromone demonstrated that it is composed of 10 compounds, and methods were subsequently developed to efficiently synthesize each of those compounds. One of the compounds was entirely new to science, and has been dubbed “leptotriene”.

The first field testing of synthetic lures took place in 2021 and found that leptotriene was quite attractive to LFB. Subsequent studies took place over the 2022 and 2023 growing seasons to confirm (i) season-long attractance, (ii) efficacy of leptotriene alone and in combination with additional pheromone compounds and (iii) attractance of a more simplified and cheaper to produce leptotriene compound. The collective results from these studies have shown that leptotriene is the key compound within the LFB pheromone, and that attractancy could be enhanced with the inclusion of additional compounds, but that may be cost prohibitive. LFB appear to be attracted to leptotriene throughout the growing season, and the cheaper version of this compound was just as attractive to LFB as a more refined version of the compound, which could help drive down costs of production.

Studies in 2024 will now revisit trap selection using the new leptotriene lure, as well as evaluate lures from multiple commercial entities who are in the process of trying to scale-up synthesis of leptotriene. All of this is with the hopes that an affordable lure can be made commercially available to growers and pest control advisors.

# Evaluating the Influence of Landscape Composition on Almond Orchard Susceptibility to Leaf-footed Bug (Coreidae: *Leptoglossus zonatus*) Colonization in the Spring

PROJECT NO: ENT038

## Principal Investigator:

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**Co-PIs:** Jocelyn Millar, Kent Daane, Jacob Wender

## Summary

Leaf-footed plant bug (Coreidae: *Leptoglossus zonatus*) (LFB) is a key pest of almonds early in the season. They feed on immature nuts, which can damage the developing embryo and/or lead to nut drop. Tools for monitoring LFB are currently very limited, as well as time and labor intensive.

Over the past six years, co-PIs Wilson, Millar and Daane have been able to develop a new pheromone-based lure that is attractive to LFB adults (project co-funded by the Almond Board of California and California Pistachio Research Board). Here, we are proposing to use this new lure and trap system to monitor a series of twenty almond orchards to gather data on LFB spring colonization activity, and then see if it is possible to relate those data to key features in the landscape surrounding the orchards. If successful, this information could then be used to develop a risk assessment model that would allow growers to prioritize LFB monitoring and management efforts in almond orchards located in areas with high risk of LFB colonization.

Over the past year, we successfully monitored for LFB in the spring (April/May) at 20 almond orchards that were spread across Fresno County, representing a diverse range of landscape contexts. We are now in the process of relating that trap capture data to the composition of the landscape surrounding each site, which should provide us with a better understanding of what key landscape features (e.g. citrus, natural habitats, residential areas etc.) are associated with spring LFB colonization.

# Revisiting Trap Selection for Pheromone-Based Monitoring of Leaffooted Bugs (*Leptoglossus* spp.) Infesting California Nut Crops

PROJECT NO: ENT039

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**Co-PIs:** Jocelyn Millar, Kent Daane

**Summary**

Epicarp lesion, nut abortion, and kernel necrosis (i.e. brown spot) caused by the feeding of a suite of true bug species is a major source of yield losses in California nut crops. Leaffooted bugs (Hemiptera: Coreidae: *Leptoglossus* spp.) (LFB) cause some of the worst damage, in part because their mouthparts are robust enough to penetrate maturing endocarp tissues. In California, LFB overwinter primarily as adults, and then in the spring (April/May) migrate into almond orchards where they feed and oviposit. Damage is unpredictable because LFB can rapidly migrate into almond orchards from surrounding areas, such as adjacent crops, native vegetation and/or residential areas. Because of these rapid buildups, and since bug damage may only become apparent after they have moved on, continuous monitoring of LFB populations is crucial in timing treatments. Current monitoring strategies for LFB are time and labor intensive, and so an improved trapping system based on pheromonal or related attractants for each species of LFB would be of great value for monitoring and potentially control purposes.

In 2021, our team demonstrated the first attraction of *L. zonatus* to synthetic pheromones in the field. In particular, the novel compound leptotriene, which took several years to isolate and identify from a composite extract prepared from thousands of male LFB, proved to be a crucial component of the blend for *L. zonatus*. The same compound is also present in extracts of *L. clypealis*, the other species of concern to California growers. In 2022-2023, we continued to test the attractancy of this novel compound, both alone and in combination with other possible pheromone components, en route to determining the optimal pheromone blend for a lure. In parallel, we improved the yield in the synthesis of leptotriene 4.6 fold, allowing this crucial chemical to be produced in larger quantities and at lower cost.

In 2022-2023, having identified and synthesized all possible components of the *L. zonatus* pheromone, our efforts focused on increasing the efficiency of production and optimizing the lure blend. We also started to work with pheromone companies (e.g. Sterling International, AlphaScents, Chemtica) to see if an affordable lure can be produced at scale. Confirming what we saw towards the end of 2021, our additional assays over this time period showed that leptotriene is a key component of the attractant pheromone of *L. zonatus*, and is likely synergized by one or more additional components. Furthermore, placing the lures in a hanging panel trap demonstrated the potential for this new monitoring system in orchards.

In 2024, we continued to evaluate new iterations of the lures, revisited the selection of an optimal trap type, and continued to work with commercial entities to develop an operational LFB trapping system for transfer to growers and pest control advisors. Findings from 2024 suggest that the hanging panel trap is still the optimal trap type for this pest. Initial commercial iterations of the pheromone lure have shown mixed results, and research in this area is still on-going. Additional field experiments scheduled through the fall will provide

# Biology, Monitoring, and Management of Native and Invasive Stink Bugs in Almond Orchards

PROJECT NO: ENTO23

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## Summary

In recent years, almond growers have faced a concerning trend: an increase in damage from multiple hemipteran 'true bug' pests, commonly known as "brown spot." Among these pests are leaffooted bugs, various native stink bugs, and the invasive brown marmorated stink bug (BMSB, *Halyomorpha halys*). These pests use their piercing-sucking mouthparts to feed on almond fruits, leading to nut abortion, drop in young fruits, and defects in kernels when feeding occurs during critical stages of the growing season. Additionally, smaller plant bugs like *Lygus*, *Calocoris*, *Phytocoris*, and false chinch bugs occasionally contribute to damage.

To address these concerns, we conducted surveys of 10-11 orchards every year, between 2021 and 2023, assessing hemipteran insect prevalence, the potential expansion of invasive BMSB, and the efficacy of monitoring and management tools. Our findings revealed that large hemipterans such as BMSB, green stink bugs, and leaffooted bugs are the top three economic pests in almonds. BMSB, in particular, has spread to the lower San Joaquin Valley (Fresno area) and is slowly expanding its range. To monitor BMSB populations and potentially predict the damage caused by this pest, we recommend using clear panel traps baited with BMSB lure supplemented with orchard scouting. Additionally, we developed a predictive model that helps to assess the potential damage at harvest based on BMSB seasonal monitoring information. We tested the efficacy of multiple insecticides over the years and presented robust lists of products that can be used to control hemipterans in almonds. These science-generated tools in addition to intentional educational outreach, we educated growers to implement an effective integrated pest management (IPM) program targeting the hemipteran pests in almond orchards.

# Evaluation of Reduced-Risk Hull Split Sprays for Navel Orangeworm

PROJECT NO: PEST02

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## Summary

Navel orangeworm is a significant pest of almonds due to its ability to penetrate through the shell and feed directly on the harvestable kernel. Kernels infested by NOW are also at increased risk of the presence of aflatoxins that are highly regulated in certain export markets. We conducted trials that evaluated the ability of insecticides to prevent infestation by navel orangeworm during the spring and at hull split. Spring trials were done by gluing mummies onto a strip of drip tape, immersing them into a pesticide solution, and then hanging them in commercial trees for up to 20 days in the spring where they were subject to egg-laying by wild navel orangeworm populations. The hull split trials were conducted in the variety Nonpareil and again in Monterey to evaluate the effects of foliar sprays made at hull split and again approximately two weeks later in each variety.

In the mummy dip bioassay, all but two insecticide treatments significantly reduced the number of mummies to 1% or less compared to 3.0% to 4.1% for the two checks. This included all three insecticides containing methoxyfenozide (Intrepid, Intrepid Edge, Enkounter), all three diamides (Altacor eVo, Shenzi, Tetraniliprole), the pyrethroid Brigade, the experimental active ingredient Plinazolin, and Avaunt eVo. The two exceptions were Spear-Lep + Leprotec (4.0%) and MBI-306 (3.0%), both of which are stand-alone microbial products. However, no live larvae were present in mummies treated with VCP-031, which represents the microbial product MBI-306 combined with the pyrethroid bifenthrin.

In the hull split Nonpareil trial there were significant reductions in kernel infestation compared to the untreated check in plots treated with products containing methoxyfenozide (Intrepid, Intrepid Edge, Enkounter), two of the diamides (Altacor eVo and Shenzi), Plinazolin, and BasinFlex + Leprotec. In the hull split Monterey trial, when major and minor damage were combined, the most effective treatment was Plinazolin (5.3%) compared to the untreated check (22.5%). Products that were statistically equivalent to the best treatment included the industry standards Intrepid, Intrepid Edge, and Altacor eVo; as well as VCP—031, Tetraniliprole and Avaunt eVo. Each of these products reduced damage to <10%. Products that had damage levels that were statistically higher than the best treatment, but were significantly reduced compared to the untreated check included Enkounter, Shenzi and BasinFlex + Leprotec.

These trials demonstrated that commercial products containing methoxyfenozide and that are in the diamide chemical class continue to be effective against navel orangeworm. To the contrary, the pyrethroid Brigade had relatively poor results across trials, which is consistent with reports of the development of resistance to insecticides that are classified as pyrethroids. Among the experimental products, Plinazolin was a stand-out product that repeatedly produced the lowest damage levels across all trials. This product is particularly intriguing because it is expected to also have activity against large bugs, like stink bugs and leaf-footed bugs, such that usage could simultaneously have value against multiple pests.

Microbial products entered into the trials had mixed results. Spear-Lep + Leprotec and MBI-306 failed to reduce NOW damage in any trial. BasinFlex (a cousin of Spear-Lep) + Leprotec reduced damage by 60% and 42% in the Nonpareil and Monterey trials, respectively, but was not available in time to include it in the mummy dip bioassay. VCP-031, which is a premix of MBI-306 with the pyrethroid bifenthrin, reduced damage to levels that were numerically or statistically lower than when either active ingredient was used individually. Avaunt eVo resulted in reductions in NOW damage of 50% across both hull split trials, with no damage in the mummy dip bioassay.

# Spatiotemporal Models to Evaluate the Potential Value of Sterile Insect Technique for Control of Navel Orangeworm

PROJECT NO: ENT028

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**Co-PIs:** Ran Wei, Brittney Goodrich, Charles Burks

**Summary**

This project succeeded in that it effectively brought together a research team to develop a spatially-explicit model of NOW that, while limited, can generate viable outputs for NOW abundance and crop damage estimates. These outputs were then used to develop economic analyses that help clarify the potential economic costs and benefits of using SIT within a broader IPM program for NOW in California tree nuts. As with many research projects, there are known and unknown barriers to be overcome, and this research team will continue to work through the bottlenecks encountered in the development of this modeling and econometric project. To be clear, and to reiterate, the model outputs and economic analyses presented in this report ARE NOT DEFINITIVE, but rather an example of the type of output this project intends to generate. More work is needed to make the models more efficient, which will then allow for higher statistical power in the output and subsequent analyses.

**Future Directions – Improve Model Efficiency**

As mentioned, the primary current limitation is computing power. Co-PI Wei has identified a pathway to improving model efficiency, and is currently recruiting a programmer to help with this process. Once this barrier has been overcome, individual scenarios can be run many more times over large spatial extents. The model run time will be reduced, and this will allow for the research team to investigate a broader series of scenarios, which is critical to better understanding the nuance of integrating SIT into the current IPM program for NOW.

**Future Directions - Model Validation and Sensitivity Analysis**

While the model currently generates outputs that appear to be feasible based on the experience of co-PIs Wilson and Burks, who have studied NOW population phenology for many years, the research team is also in the process of acquiring monitoring and/or crop damage data from a wide range of sites and dates in order to more effectively validate model output.

Additionally, once the model efficiency has been improved, we plan to run a sensitivity analysis to determine the relative weight of different model components. While this most immediately helps clarify the expected variance within the model, it also highlights parameters that need to be more accurately defined, and in this way provides a roadmap for additional lab or field research efforts focused on NOW ecology, biology etc.

# Initiation of First Stage Product Development of a Friendly Navel Orangeworm for Californian Almond Growers

PROJECT NO: ENT033

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## Summary

### Key Highlights:

- Oxitec's Friendly™ navel orangeworm is poised to become an innovative and sustainable tool for Californian almond and pistachio growers.
- An early-stage project to develop the tools to generate a Friendly™ navel orangeworm product has been successfully delivered.
- Completion of this first project lays a clear path to full development of a scalable, effective Friendly™ navel orangeworm solution.

Oxitec is the leading developer of sustainable biological solutions to control pests that transmit disease, destroy crops and harm livestock. Oxitec's Friendly™ biotechnology platform is a genetics-based biological approach for targeted, non-toxic pest management. Friendly™ insects are engineered to carry a self-limiting gene that enables production of male-only cohorts which, after release into the environment, find and mate with female pest counterparts. Their offspring (females only, or both sexes, according to the technology version) cannot survive, so, with sustained treatment, the pest population crashes in the treated area. The self-limiting genes are non-toxic and cannot establish in the environment. The two leading Friendly™ products – the Friendly™ Aedes mosquito control solution (Spinner et al. 2022) and the Friendly™ fall armyworm crop protection product (Reavey et al. 2022) – are approved for commercial use in Brazil (the latter in Paraguay also). Friendly™ Aedes is now serving households, businesses and cities across Brazil and Friendly™ fall armyworm has been piloted on thousands of acres of commercial corn in Brazil's key corn-growing regions.

At the invitation of the Almond Board of California, Oxitec has conducted a successful feasibility project assessing the navel orangeworm challenge in the state, how new solutions are needed, and how a future Friendly™ navel orangeworm solution could deliver significant benefits to growers statewide. Following the feasibility project's conclusion that the Friendly™ technology offers significant promise, this new project was launched to initiate the first steps towards developing a Friendly™ navel orangeworm. This early-stage project has successfully achieved all of its key goals:

- establishment of rearing processes for navel orangeworm in Oxitec's laboratories to sustain future strain development activities;
- identification of the genetic sequences needed to engineer designed traits in navel orangeworm;
- development of methods to deliver DNA constructs into navel orangeworm eggs; and
- mapping of the regulatory pathway of a future Friendly™ navel orangeworm product to bring it to market in California.

The successful completion of this project has prepared Oxitec to initiate a full Friendly™ navel orangeworm product development project. When built, this new solution is expected to become an innovative, effective and sustainable tool to control populations of the most devastating pest of nut crops in California.



# Ecology, Monitoring and Management of Carpophilus Beetle

PROJECT NO: ENT037

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**Co-PIs:** Jhalendra Rijal, David Haviland, Sudan Gyawaly, Raman Bansal

**Summary**

Carpophilus beetle (Nitidulidae: *Carpophilus truncatus*) is a new invasive pest of almonds and pistachios in California. Damage occurs when adults and larvae feed directly on the developing kernel, causing reductions in both yield and quality. Survey efforts conducted during the past year following detection have shown that this pest has already spread to almond and pistachio orchards throughout the San Joaquin Valley.

The carpophilus beetle is recognized as one of the top two pests of almond production in Australia, where growers typically experience 2-5% of kernels infested, and can sometimes experience damage exceeding 30%. Australian farmers primarily utilize overwintering sanitation as a way to reduce damage. Other management options, such as biological and chemical controls, have very limited or unknown efficacy.

This project was designed to (i) improve our understanding of the phenology and temperature requirements of this pest, (ii) determine the timing of its activity in orchards, (iii) confirm crop infestation patterns within the tree canopy and within the field (i.e. edge effects), (iv) evaluate chemical controls and (v) evaluate an experimental pheromone lure (developed in Australia) under California conditions, as well as (vi) educate growers and pest control advisors about this pest.

Over the past year, new information was generated on the timing of spring emergence of carpophilus beetles from remnant mummy nuts, as well as the initiation of activity on new crop nuts. At hull split, chemical controls were evaluated in almonds. At harvest, stratified samples will be collected to determine if infestation varies between the lower and upper areas of the tree canopy, as well as between the orchard edge and interior, which will contribute to the development of better monitoring strategies. Additionally, improved rearing methods have been developed, which allows for the production of large numbers of carpophilus beetle to support laboratory assays. Using these methods, a study to determine the lower and upper temperature thresholds of this pest, as well as its development rate, has been initiated and is currently underway. A new pest identification guide for carpophilus beetle in almonds was also developed, and this was complimented by dozens of extension and outreach events across the Central Valley to educate growers and pest control advisors about the ecology, monitoring and management of carpophilus beetle. This included an online webinar featuring research and industry personnel from Australia, who shared their experiences managing this pest in almonds over the past decade. Finally, the research team secured the necessary administrative and importation protocols to acquire an experimental pheromone lure for carpophilus beetle that was developed in Australia. Research and extension on this new invasive pest of tree nuts will continue in 2025, with continued focus on the development of pest ID and monitoring strategies, as well as cultural and chemical controls.

# Implementing a Nematode Management System for Almond Using Chemical and Biological Treatments

PROJECT NO: PATH22

## Principal Investigator:

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## Summary

The expanse of almond production on ~1.5 million acre includes different growth environments across California. This among other parameters, includes different soil types, and possibly different precrops for plantings of almond. Frequently almond follows its own species or other (specialty) crops that leave behind soil-borne plant pathogens. Plant-parasitic nematodes, e.g., *Meloidogyne* spp. (root-knot nematodes, RKN), and *Pratylenchus vulnus* (root lesion nematode, RLN) are often found in soils scheduled for almond planting. Traditionally, preplant soil fumigation, until 2005 with methyl bromide and since then with 1,3-dichloropropene (1,3-D) often in mixes with chloropicrin when following a prior *Prunus* planting was used to reduce the risk for damage by these soil-dwelling parasites. Preplant soil treatments were applied in addition to rootstock choice because of the lack of resistance against certain nematode problems. Increasing regulation in the application pattern of 1,3-D based on environmental and human health concerns, increase the expense and reduce the treatment efficacy of this method. The sustainable pest management road map (SMP) envisions further reductions of chemical input of 90% by 2050.

Alternative cost-effective nematode management strategies are urgently needed. In this project, materials and methods that have been identified in prior experimentation to be suppressive against RLN are used in field plots as part of nematode management systems. Although new chemistries have shown high levels of efficacies, these do not mirror the known performance of soil fumigation and management needs to include post plant remedies to avert the risk of nematode damage. Several experiments are in place where preplant strategies are coupled with post-plant treatments. The first experiment reaches harvest maturity in 2024 allowing comprehensive assessments of potential treatment program benefits.

Two chemical nematicides had shown promise in suppressing ring nematode (*Mesocriconema xenoplax*) – a difficult to manage nematode species – under greenhouse conditions. In this project year, these materials were further evaluated under microplot conditions. Especially an experimental material had high levels of efficacy. Further experimentation is underway to corroborate these promising results. In summary, this project makes progress in offering nematode management strategies when the proven tools become more difficult to use.

# Weed Research and Extension to Address Almond Grower and Industry Management and Sustainability Goals

PROJECT NO: HORT12

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## **Summary**

Weed management is an important component of orchard management practices and production expense on every acre of almonds in the state. At the same time, chemical and physical weed management practices can contribute to system sustainability challenges, environmental degradation, and domestic and export market concerns. Research and extension efforts continue to help the almond industry navigate these competing, and at times contradictory, challenges. Project HORT12 serves as part of the foundation for a statewide weed management research and extension program in orchards, vineyards, and other irrigated crops. This program is largely, but not exclusively, focused on conventional herbicides which is applicable to more than 95% of almond growers.

Major efforts during this reporting period included: conclusion of two-year evaluations of the postemergence herbicide tiafenacil and the preemergence herbicide pyroxasulfone; both were safe on young almonds at the expected label rate in research orchards on campus. A non-chemical weed control project utilizing electrical weed control was initiated in a newly-planted block of organic almonds; thus far EWC appears safe on almond and effective for control of important weeds. A grower-scale (40 acre) demonstration of chemigated pendimethalin applied at 1 qt/A in spring provided impressive levels of supplemental control of glyphosate-resistant junglerice and other summer weeds in the frequently-wetted areas at each emitter at very low cost. Two evaluations of glyphosate and glufosinate fate almond residues were completed and a third project initiated; in this work, glyphosate, glufosinate and their primary metabolites were in soil as expected. When almonds were fractionated and analyzed, glyphosate detections were mostly in hull materials and little metabolite noted suggesting soil-bound glyphosate as the major contributor. Interestingly, the glufosinate detections were primarily a metabolite and were detected in hull, shell, and kernels in samples collected from various points in the process and also in nuts harvested directly from trees. This suggests a different management approach may be needed when considering residues of these herbicides in export markets with low MRL requirements.

These real world weed management and herbicide performance data form the backbone of an extension effort aimed at providing information to growers, Pest Control Advisors, Farm Advisors, the crop protection industry, and the regulatory community. This type of research which at times includes applied field research, basic lab and greenhouse science, is flexible, responsive, and relevant to the vast majority of almond producers.

# Region-Wide, Disease Risk Forecasting System

PROJECT NO: ENT035

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**Co-PIs:** Jordan Hazell

**Summary**

This project developed an area-wide disease risk prediction system for seven of the main almond-producing counties representing 13 regions in the California Central Valley. Counties included Butte, Colusa, Fresno, Kern, Madera, Merced, and Stanislaus. The forecasting system was based on in-canopy environmental data supplied by the Semios network of data loggers. By using the epidemiological models developed for anthracnose, Alternaria leaf spot, bacterial blast, bacterial spot, brown rot, and scab, we successfully scaled the use of the models to regional forecasts via aggregated site-level, in-canopy data. In 2024, the model for anthracnose was adjusted to higher rainfall amounts before reaching a critical value or action level. A new model for scab lesion sporulation on 1-year-old twigs was included on-line for 2024 forecasts. Weekly forecasts started on Feb. 26, 2024, and continued through June 24, 2024. The Almond Board website where forecasts were posted had a high number of visits for most of the spring and early summer seasons. Thus, this project effectively provided growers regional forecasts of the relative risk of these diseases for at-site-level management decisions. A farm advisor, an extension specialist, and the PI confirmed forecasts in selected regions by observing disease outbreaks for brown rot, scab, bacterial spot, and Alternaria leaf spot. Documentation in the field was also done for diseases forecasted but not posing a risk in any county or in specific regions of the state such as anthracnose and blast. This project was developed to ultimately contribute to the Almond Orchard 2025 goal of increasing adoption of environmental stewardship and successful disease management by reducing fungicide use by 25% to 50% based disease risk forecasts.

# Biology and Management of Almond Brown Rot Jacket Rot Shot Hole and Hull Rot

PROJECT NO: PATH4

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## Summary

In 2023, we continued to evaluate new fungicides and biological treatments for the management of brown rot (caused by *Monilinia* spp.), jacket rot (caused by *Botrytis*, *Monilinia*, and *Sclerotinia* spp.), shot hole (caused by *Wilsonomyces carpophilus*), and hull rot (caused by *Monilinia*, *Rhizopus*, and sometimes by *Aspergillus* spp.). Environmental conditions in the spring of 2023 were not favorable for disease development. Although high rainfall occurred at our trial sites, temperatures remained cool until late spring. Therefore, the incidence of brown rot was low, except when trees were inoculated, and shot hole also occurred at low severity. Gray mold blossom blight, however, developed at high incidence in a field study at KARE. Brown rot fungicides that resulted in statistically similar high efficacy in 2023 were the single-site Cevya (FRAC 3), Fontelis and Tesaris (both FRAC 7), Approach (FRAC 11), the pre-mixtures Elysis (Mibelya) and Miravis Duo (both FRAC 3/7), Merivon (FRAC 7/11), and Miravis Prime (FRAC 7/12), rotations of Luna Experience (FRAC 3/7) and Luna Sensaton (FRAC 7/11), as well as the experimental Axios Cion (FRAC 9/52), CX-10490 (FRAC 48), GF5249, and GF5003. The plant extract treatments BTS EXP100 and ProBlad Verde, the biocontrol Serifel, and the rotation of the plant extract Gargoil-Dart mixture with the biocontrol Botector showed intermediate efficacy in one study where trees were inoculated, and BTS EXP100 and CX-10490 were very effective in another trial. For the management of *Botrytis* blossom blight, Cevya, Miravis Duo, Miravis Prime, Elysis, Axios Cion, GF5003 and GF5249 numerically had the lowest incidence; ProBlad Verde was also very effective, but Botector was much less effective. The efficacy of field treatments was also evaluated on collected, field-treated flower petals in the UC Davis trial on cv. Drake, and Miravis Prime and Axios Cion resulted in the lowest incidence of gray mold. The *in vitro* toxicity of the new fungicide ipflufenquin (FRAC 52; a component of the experimental pre-mixture Axios Cion) was previously found to be very high against *M. laxa* (EC<sub>50</sub> value range 0.005 to 0.022 ppm) and *B. cinerea* (EC<sub>50</sub> value range 0.002 to 0.091 ppm). For shot hole on fruit and leaves, Cevya, Luna Sensaton, and CX-10490 resulted in the overall lowest disease ratings. Among three trials conducted for hull rot, the highest severity (i.e., 24.3 strikes/tree) on untreated trees occurred in a study with Monterey almond. Approach (FRAC 11) and Adamant (FRAC 3/11) reduced the disease to the lowest levels, but Cevya, Miravis Prime, Axios Cion, GF5003, and GF5249 were also very effective. In our previous studies, we found that the mode of action of the foliar fertilizer diKaP against hull rot caused by *R. stolonifer* was associated with reduction of fumaric acid that is produced by the pathogen and is involved in causing hull rot symptoms. When the fungus was grown in a minimal medium with the addition of diKaP, the production of fumaric acid was almost completely inhibited, while growth was increased. In our evaluations of natural host resistance to fungal diseases in new genotypes and cultivars in our almond block at UC Davis, brown rot developed at very low incidence in 2023. Evaluations were done for shot hole. Based on 2017-2020 and 2022-2023 data, cvs. Aldrich, Capitola, Jenete, Kester, Nonpareil, and Sterling, as well as genotypes 7-159 ucd, p13.019, p16.013, and UCD 8-160 had lower levels of disease on fruit and leaves in at least five of the evaluation years as compared to the other accessions, and therefore are less susceptible.

# Investigation of *Aspergillus Niger* Causing Hull Rot and Conditions Conducive to Disease Development in Kern County

PROJECT NO: PATH18

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## Summary

Hull rot is primarily caused by *Rhizopus stolonifer* and *Monilinia fructicola* causing the leaves, spurs, and parts of the shoot near the infected fruits to die. In Kern County, and the Southern San Joaquin Valley, *R. stolonifer* is more prevalent, and this fungus produces a toxin which moves from the infected fruit into the surrounding tissues killing the vascular tissues. Hull rot affects future yields by killing fruiting spurs and wood. In the past years, orchards affected with hull rot in Kern County and other Counties in the central valley showed presence of *A. niger* in symptomatic fruit in branches affected by hull rot. Many samples were processed by Dr. Michailides showed that hull rot samples from the San Joaquin and Sacramento Valleys were also infected with *A. niger* alone and/or *R. stolonifer*.

We successfully reproduced the symptoms of hull rot in the field by inoculating fruit with two different isolates of *A. niger*. We also successfully reproduced the symptoms using two different concentrations of an *A. niger* isolate used in 2018 and 2019 inoculations. This proves that the lower concentration 10,000 spores used in previous experiments was sufficient to reproduce hull rot symptoms and similar to using a higher concentration.

We determined the most susceptible developmental stage by inoculating fruit at the three developmental stages (unsplit, deep V (b2 stage), and split less than 1 cm (stage c) and found that inoculated fruits at stage (c) had the highest percentage of spurs developing hull rot symptoms. Also, population of *A. niger* on fruit were assessed, and results show that the highest population of *A. niger* was observed later in July and through early August and corresponded with fruit that already split with less than 1 cm.

Furthermore, July nitrogen leaf analyses were not significantly different between the two experimental plots showing differences in hull rot incidence for two years in a row and it was within the adequate range, and did not explain the difference in disease incidence in this orchard.

In 2019, preliminary work looking at sensitivity of *A. niger* to different groups of fungicides was tested In vitro and in the field. In vitro tests showed that *A. niger* was sensitive to fungicides in FRAC groups 3, 7+11, and 7. An experiment with three fungicides belonging to FRAC groups 3, 7+11, and 11 were conducted in a commercial orchard and all fungicides reduced the number of symptomatic spurs by approximately 39-54% compared to control.

Increase in hull rot can have a negative effect on yield. The data show that there can be a variation between sites or even within the same orchard, and we are going to continue with this work to collect more data points. It is important to understand disease incidence and severity and factors conducive to hull rot in the orchard to implement best management practices to control this disease. Our plans are to continue this effort and collect more data points in the coming seasons.

# Epidemiology and Management of Phytophthora Root and Crown Rot of Almond

PROJECT NO: PATH15

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## Summary

In 2023, 32 additional isolates of *Phytophthora* spp. from 16 orchards with declining trees in California's almond-growing areas were collected, bringing the total to 300. The new species *P. niederhauseri* and *P. mediterranea* (formerly *P. sp. ax*) were most commonly isolated, followed by species long endemic to California, such as *P. syringae*, members of the *P. citricola* complex (Clade 2c), and *P. cactorum*. Isolates of *P. niederhauseri* and *P. mediterranea* have an optimum growth temperature of around 30°C and are most active in late spring and summer, while the other species are active in cooler conditions. The emergence of the new high-temperature species may be linked to growers' shift to surface water, providing new inoculum sources. In spring 2023, with high rainfall and cool temperatures, *P. syringae* caused 'aerial *Phytophthora*' infections (pruning wound canker) of scaffold branches. Unlike previous reports, infections killed green shoots and progressed into scaffold and trunks, where summer cankers were not associated with pruning wounds. By late summer, the pathogen was no longer recoverable. A method was developed for detecting *Phytophthora* in cankers based on DNA amplification, allowing pathogen identification when isolations fail due to poor sample quality or loss of pathogen viability. Sensitivity studies showed that new fungicides—oxathiapiprolin (Orondis), mandipropamid (Revus), fluopicolide (Presidio), and ethaboxam (Elumin)—are highly toxic in vitro against all *Phytophthora* species. However, *P. mediterranea* isolates were insensitive to phosphites. Field studies compared systemic fungicide actions to Ridomil Gold (mefenoxam) using grafted trees. Orondis, Presidio, and Ridomil Gold were the most systemic, reducing cankers on rootstocks and scions, while Elumin and Revus only protected the rootstock. Orondis showed long-lasting efficacy in roots and trunks when applied to pre-wetted soil. Field studies on scaffold inoculations showed that ProPhyt significantly reduced cankers, even 16 weeks post-inoculation. Registrations for Elumin and Presidio are ongoing, with Syngenta seeking Revus approval for nurseries. Developing non-phytotoxic fungicides for managing *Phytophthora* root and crown rots is critical, given the need for alternatives to mefenoxam and phosphites due to resistance in *Phytophthora* species on other crops and regulatory restrictions on soil fumigants.

# Epidemiology and Management of Almond Band Canker Disease in Young Orchards

PROJECT NO: PATH16

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## **Summary**

Almond band canker has become more severe in California, especially in young orchards in recent years. Three objectives were involved in this project. In Objective #1, various materials were tested in an almond nursery by using culturing isolation in synthetic media and real-time quantitative PCR (qPCR). Latent infection levels of *Phomopsis* spp. and *Botryosphaeria dothidea* were high in most samples, while those caused by *Neofusicoccum* spp. and *Diplodia* spp. varied among various samples. *Botryosphaeria dothidea* is the predominant pathogen in the nursery, mother trees and new orchards occurring as latent infection. Various fungal species were isolated from canker-symptomatic trunk tissues of a mother tree and a 2nd –leaf orchard. In Objective #2, fungicide treatments on trunks and crotches of young trees were conducted in two young orchards. Topsin and BioMend+ significantly slowed the canker development compared to untreated control. In Objective #3, distribution of trees showing various levels of canker disease was investigated in an almond orchard, and various almond varieties were evaluated for their susceptibilities to canker disease. ‘Peerless’ and ‘Butte’ are most susceptible, ‘Nonpareil’ is less susceptible, and ‘Aldrich’ is most resistant among the four varieties.



# Improve the Detection and Risk Prediction of *Pseudomonas Syringae* Causing Bacterial Blast and Bacterial Canker of Almond in California

PROJECT NO: PATH20

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**Summary**

Bacterial blast and canker is a complex disease of almond and yet little information is available about the disease biology, epidemiology and bacterial pathogen diversity associated with almond in California. Furthermore, disease management is difficult, and tools are lacking to assess or predict disease risk in orchards. The present study investigated the diversity of pathogenic *Pseudomonas* species associated with almond and reexamined the disease biology and epidemiology. We also assessed the status of kasugamycin and copper resistance in bacterial populations from almond. Furthermore, we developed and validated a real-time PCR assay for the specific detection and quantification of pathogenic *P. syringae* strains directly from almond tissues. This assay was used in epidemiological studies to identify main inoculum reservoirs, monitor the seasonal population dynamics of *P. syringae* in almond orchards, and predict bacterial blast disease risk during bloom. The ultimate goal of this work is to provide decision support tools to growers to help them make informed decisions on disease management and bactericide applications.

Whole genome sequences of 120 fluorescent pseudomonads isolated from almond were obtained for this study. Phylogenomic analyses combined with genome mining and field pathogenicity tests identified *P. syringae* pv. *syringae*, *P. viridiflava*, and *P. cerasi* as the main pathogens associated with almond. *P. syringae* pv. *syringae* prevailed in orchards and caused both canker and blast symptoms. In contrast, *P. cerasi* and *P. viridiflava* appeared sporadic in orchards and only caused cankers.

Genome mining for resistant genes to kasugamycin and copper revealed variable combinations of antibiotic-resistance genes. Of interest was *ctpV*, a gene that encodes for a putative copper exporter that confers resistance to copper. *ctpV* was present in 44% of the isolate populations of *P. syringae* pv. *syringae* and in 50% of *P. viridiflava* isolates, but none of the *P. cerasi* isolates had the *ctpV* gene. The *ctpV* copper resistance genotype correlated with copper resistance phenotype in bioassays. As a result, we designed primers targeting the *ctpV* gene that can be used to identify copper resistant isolates in bacterial population from almond orchards. These findings also suggest that copper cannot be used reliably to manage bacterial blast and bacterial canker of almond. On the other hand, all isolates tested were susceptible to kasugamycin at the recommended rate of 100 ppm, suggesting that currently there is no resistance to kasugamycin in California populations of pathogenic *Pseudomonas* species. Thus, kasugamycin can be effectively used to manage bacterial blast and bacterial canker of almond.

We developed specific primers and a real-time PCR assay for the detection and quantification of pathogenic bacteria from almond tissues. Using this assay and following large surveys of orchards in the San Joaquin valley, we determined that almond buds were the main inoculum reservoir for *P. syringae* pv. *syringae*. Therefore, bud sampling and monitoring combined with PCR analyses were used to investigate the seasonal population dynamics of *P. syringae* pv. *syringae* in almond orchards. The detection of *P. syringae* pv. *syringae* populations from dormant buds sampled one month to two weeks prior to bloom time generally correlated with the detection of the bacterium on flowers during bloom. On the other hand, low bacterial levels in dormant buds usually correlated with low bacterial levels on flowers. Overall, results indicated that disease risk in orchards may be predicted from the sampling of buds in late winter and prior to bloom. Furthermore, bacterial level thresholds in buds prior to bloom that can predict disease risk were tentatively established. This information and tools may be used by growers for decision making on bactericide application to manage bacterial blast of almond.

# Assessing Almond Rootstock Susceptibility to *Ganoderma* Wood Decay and Crown Gall

PROJECT NO: PEST01

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## Summary

Previous work has found *Ganoderma* species to be associated with wood decay of dead or dying almond trees and failure of living almond trees. Pathogenicity tests were used to determine the pathogenicity of *Ganoderma adspersum* and *G. brownii* on young, healthy almond trees (rootstocks Nemaguard, Viking, and Lovell). Rootstocks of walnut and pistachio were also tested for comparison. Inoculations were made by coring into the sapwood of the crown and pipetting *Ganoderma* spore solutions into the wounds. In two greenhouse trials, 10 months post inoculation, trees had no visual, external symptoms of disease, though there were differences in the extent of internal wood decay and xylem discoloration. *Ganoderma adspersum* caused extensive wood decay and discoloration and was reisolated from Nemaguard and Viking. The data suggests that *Ganoderma adspersum* was capable of infecting healthy sapwood and has the potential to be a pathogen of young trees.

Wood decay caused by *Ganoderma* was often observed in orchards with extensive crown gall. The high coincidence of the fungal and bacterial pathogens suggested potential synergistic effects. However, coinoculation assays revealed that the pathogens do not significantly interact directly with each other nor promote increased initial disease development. The rise of multi-pathogen diseases in almonds likely results from the adoption of management practices that have created a system capable of sustaining multiple pathogens.

To gain an understanding of how and when *Ganoderma adspersum* was spreading in California almond orchards, disease progression and tree losses were studied in California almond orchards every eight months for two years. Substantial tree losses associated with wood decay were observed throughout the study. The seasonal abundance of *G. adspersum* spores was studied using mesh-covered buckets. Regression analysis resulted in a strong positive relationship between average temperature and *G. adspersum* spore abundance. The highest number of spores trapped in almond orchards during this study was during harvest from August to September. These findings warrant the need to assess management practices to reduce dust production and fungal spore transmission in California orchards. The key findings of this project and previous projects include: 1) *Ganoderma adspersum* appears to be an aggressive, emerging pathogen in California Almond orchards in California, especially in the San Joaquin valley, 2) Nemaguard and Viking cultivars appear to be especially susceptible to infection, 3) *G. adspersum* produces large amounts of spores in almond orchards during harvest from August to September. Continued monitoring of the distribution and impact of *G. adspersum* is warranted. Management of the pathogen once it is established in an orchard is very difficult. These findings suggest the need to assess management practices to reduce dust production and fungal spore transmission as well as consider the types of rootstocks that are planted in the future.

# Assessing Nematode Control Soil Health and Tree Vigor in a Commercial Almond Orchard Four Years After Soil Biosolarization

PROJECT NO: PATH19

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## Summary

Trunk diameter data was collected in February 2023, representing 5.6 years after the field was biosolarized and roughly 5 years after the orchard was established. For Monterey and Bennett Hickman varieties, no difference in trunk diameter were observed when comparing trees in biosolarized soils to untreated control soil. For Nonpareil trees, a significant increase in trunk diameter ( $P < 0.0001$ ) was observed for trees grown in soils that were biosolarized using Nonpareil hulls/shells as soil amendment (mean diameter of 16.3 cm) compared to trees grown in solarized soil (solar heated but lacking hull/shell amendment) (mean diameter of 15.4 cm) and those grown in untreated control soil (mean diameter of 15.1 cm). In the summer of 2023, the first yield measurements were performed for trees grown in biosolarized and untreated control plots. Although all varieties grown in biosolarized soils showed an increase in mean yield compared to trees in untreated control plots, only the Monterey variety exhibited a statistically significant increase in yield. However, when the global effect of biosolarization (regardless of biosolarization conditions such as amendment type) was compared against the control with variety as a blocking variable, a statistically significant increase in yield was detected ( $P = 0.05$ ). The mean increase in yield for trees in biosolarized soils was 4.8%. These are compelling results that represent the culmination of 7 years of field work. They complement data from prior years that have shown multiple benefits to soil and tree health associated with biosolarization, such as multi-year increases in soil nitrogen and potassium, multi-year suppression of phytoparasitic nematodes, and enhancement of trunk growth and canopy health.