YEAR-END PROGRESS REPORT

Agency: California Avocado Commission (CAC)
Fiscal Year: 2008-2009 **Project Completion Percentage:** 35%

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PROJECT TITLE: SALINITY CHLORIDE INTERACTIONS AND THEIR INFLUENCE ON AVOCADO YIELDS

PROGRESS REPORT (NOVEMBER 2008 – OCTOBER 2009)

PROJECT OBJECTIVES:

Avocado yields are significantly decreased by chloride toxicity and soil salinity throughout California avocado orchards. The use of salinity tolerant rootstocks offers one avenue for alleviating these problems, but actual differences in salinity tolerance among rootstocks and their corresponding fruit yields have not yet been evaluated across the range of soil, water, and climatic conditions where avocados are grown. The objectives of this research are to examine the effects of salinity and chloride toxicity on avocado yields across a transect of the avocado production areas in S. California, and the yield performances for the most common rootstocks that are now being used by the industry. The results of this study will lead to development of a predictive model to forecast differences in yields that can be expected in relation to soil properties, irrigation water quality, and management practices, and will identify the best rootstocks for saline soils.

PROGRESS:

Since the first annual report (Oct 2008), we have carried out all of the planned research activities that are outlined in our 2009 proposal. This has involved detailed monitoring of 14 orchards that are located in 5 coastal counties from San Luis Obsipo to San Diego. We have also completed the first iteration of a computer model that is able to separate out the effects of chloride and salinity on avocado leaf chloride content, root growth, and yield for avocado trees grown on different rootstocks and in different soil types. The main method that we are using for the statistical analysis relies on an artificial neural network (ANN) method, which uses machine learning to extract the mathematical relationships between all of the different variables that influence yield, root growth, and leaf chloride content (Peltarion, Synapse). Due to low crop yields this year across all areas, and crop failure in some areas, our analyses this year mainly considered the effects of salinity on leaf chloride content and root growth. Based on our observations for this coming year, the second iteration of our model that will be generated in 2010 will include the effects of salinity on avocado yields. Additional refinement of the model will include the impact of irrigation set duration and frequency as management variables. Details of the modeling procedures are provided in the midyear report for 2009.

Since the midyear report for 2009, our activities have centered on scheduled monitoring of the soil and water chemistry for the 14 experimental sites with 5 visits to each site per year between February and October. Our routine data collection measurements of EC values for soil water extracts and the irrigation water, and soil water monitoring data from the Watermark data loggers. In September and October of this year, we have been engaged in collection of plant tissue and soil samples that are currently being subjected to complete chemical analyses by Fruit Grower's Laboratory. After these analyses are completed this winter, the next full statistical analysis of the data set will be conducted, with the results available in May 2010.

In February 2009, we set up a new experimental site at Mud Creek Ranch in the Santa Paula area, which will bring in additional replication and soil variation data to increase the power of the statistical analyses. We also set up another location at the Bartleson orchard in the Nipomo area near San Luis Obispo in April 2009. This location adds an additional 45 trees to the trial, to include 15 each of Toro Canyon, Mexican, and Dusa. This location provides additional replication for nonclonal Mexican rootstocks that were previously underrepresented in the data set. This brings the total number of locations in our trial to 14.

Along with our field research activities, educational and outreach activities this year have included a presentation at the annual meeting of the California Avocado Society, and three grower seminars that were held in Temecula, Santa Barbara, and San Luis Obispo. I also wrote an article that has been published in the 2009 Avocado Yearbook, which provides practical guidance on salinity management, an overview of our project, and a summary of our results that we have obtained to date. The review further provides practical advice and procedures for grower monitoring of salinity using an EC pen, and guidelines for soil leaching practices to prevent salt accumulation.

Other relevant activities this year include preparation of two grant proposals for extramural funding agencies that will allow us to expand on our current project. One proposal that has since been funded (UC-MEXUS), will support Dr. Macario Bacilio, a scientist from CIBNOR in Baja, Mexico, to carry out research on water use efficiency in avocado under saline conditions and the utility of bacterial inoculants for improving salinity tolerance. Dr. Bacilio is an expert on the production and use of bacterial inoculants for improving root growth, water use efficiency, and salinity tolerance in field crops including maize and peppers. With these crops, bacterial inoculants significantly increase yields mainly by removing high concentrations of stress-ethylene from the plant rhizosphere, which is a potent inhibitor of root growth. In these crops, and in avocado, both water stress and salinity cause increases in ethylene production that inhibit root growth. This leads to a downward spiral, as poor root growth leads to decreased water and nutrient uptake, resulting in still further stress and more ethylene production. Certain bacteria that contain a gene called acdS, are able to break this cycle by producing an enzyme that degrades the precursor of ethylene, amino cyclopropane carboxylate, in the plant rhizosphere. These bacteria have been developed as soil inoculants for crop plants, but have not yet been tested with avocado or other tree crops. Nonetheless, they warrant complete testing. Dr. Bacilio has just arrived at UCR in October 2009, and will be here for one year to conduct field experiments on the use of these inoculants with avocado. This will also involve collaboration with Dr. Bob Heath, who will provide expertise in use of thermal pulse, sap flow measurements for monitoring water use efficiency. Our project will compare two bacterial formulations, including SOBEC, and BioSoil, both of which contain mixtures of plant growth promoting bacteria.

A second pending proposal was submitted to the Kearney Foundation of Soil Science in September 2009 and if selected for funding, will examine the population density of indigenous ethylene degrading bacteria in the avocado rhizosphere for trees in different soils, and over time to determine the extent to which this beneficial microbial activity is already present in soils, the effect of management practices on these bacteria, and the degree to which this can be further influenced by soil inoculation.

Lastly, we have been engaged in startup on the use of biochar for promoting increased microbial activity, increased root growth, and improved soil fertility in avocado orchards. All of these new activities will complement the salinity work at no additional cost to CAC.

Results for 2009 Project Year

<u>Root growth.</u> Soil salinity and chloride levels are known to directly affect plant root growth and their impacts have been measured using hydroponics. Comparisons of salinity effects on different rootstocks in the field, however, is much more complicated, as the distribution of the roots in the soil profile may vary at different depths and roots encounter constantly changing conditions with each irrigation and wet dry cycle. To examine the root distributions, the initial analysis used traditional statistical procedures in which we calculated averages of the fine root growth (g/kg soil) and distributions across all sites.

As shown in Figure 1, which presents the average for each rootstock across all sites and conditions, the fine root weights varied considerably depending on the particular rootstock. Mexican and Duke 7 produced the least weight of feeder roots, with an average of about 15 grams of roots per kg of soil. In contrast, Dusa and Toro Canyon had 50% more roots in the same volume of soil with mean weights of 22 g/kg soil. Among all the rootstocks tested, Thomas had the greatest mass of roots with approximately 47 g/kg soil, which corresponds to two to three times the root mass of the other rootstocks. The root growth distribution patterns in the soil profile further show that approximately 60% of the feeder roots are located in the top six inches of the soil, with another 15-20% occurring at 6 to 12 inches, and 15% in the lowest profile measured here 12 – 18 inches. The relative proportion of root distributed in the three zones was essentially identical for all five rootstocks (Figure 1b).

The abundant root production for Thomas has been observed before (John Menge, personal communication), and has been suggested as the reason for this particular rootstock's resistance to Phytophthora root rot, in which it appears to replace damaged roots rapidly by new root growth. While useful for root rot resistance, the extraordinary root production by this rootstock could also be a detrimental trait for saline soils and account for the greater problems with chloride toxicity that are commonly observed with Thomas. In this case, two times the root length would be expected to lead to twice the leaf Cl accumulation at equivalent Cl uptake rates by the roots. The abundant root growth also suggests that growth of Thomas roots may be less sensitive to Cl toxicity. To examine this question, an ANN model was generated using soil and water input variables to predict root growth in the 0-6 inch profile for each rootstock (data not shown). The resulting model confirmed our hypothesis and revealed that Toro Canyon, Dusa, Duke 7, and Mexican rootstocks sharply decrease root growth as irrigation water chloride concentrations increase from 20 to 100 mg/L. In contrast, Thomas proved to be completely insensitive to chloride and maintained the same root mass irrespective of the irrigation water chloride concentration. This strongly supports the idea that the sensitivity of Hass scions on this rootstock is due to the combined effects of a large root mass and the ability of this rootstock to resist the effects of chloride toxicity on new root development.

A.

B.

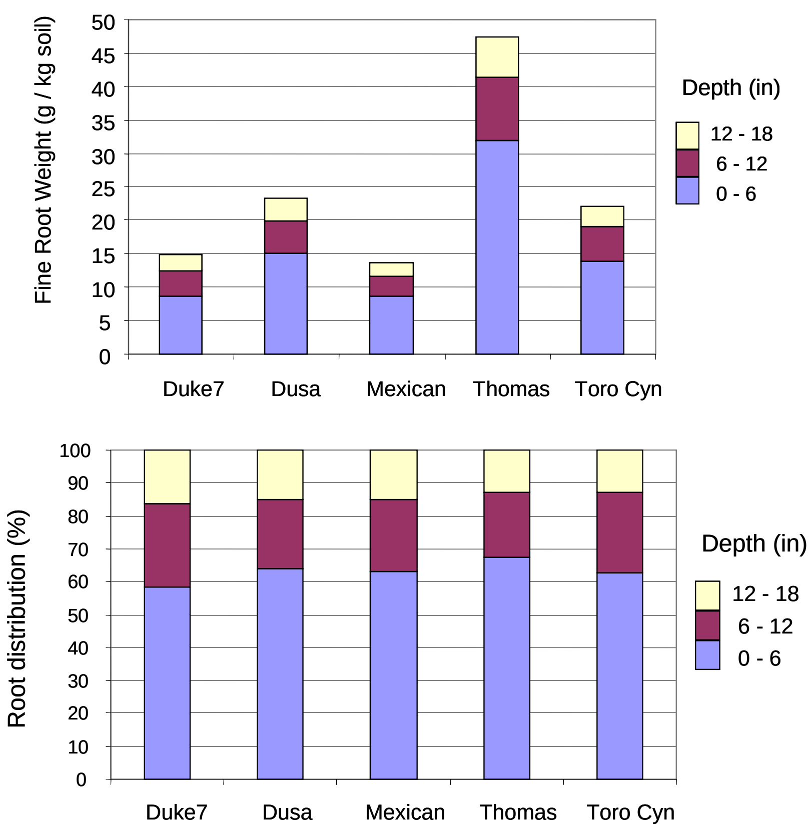

Figure 1. Root mass (A) and root distribution (B) in the soil profile for five avocado rootstocks grown across a 400 mile transect of the avocado production area in S. California from San Luis Obispo (north) to San Diego (south). Values shown are average for all soil and salinity conditions in the region that was surveyed.

Rootstock effects on leaf chloride accumulation

Sensitivity analysis for individual variables showed there were several important predictors of leaf chloride content including irrigation water chloride and EC values, soil chloride, rootstock, soil pH, and soil clay content. The relationships between soil chloride and leaf chloride contents are shown in Figure 2 for each of the 5 rootstocks.

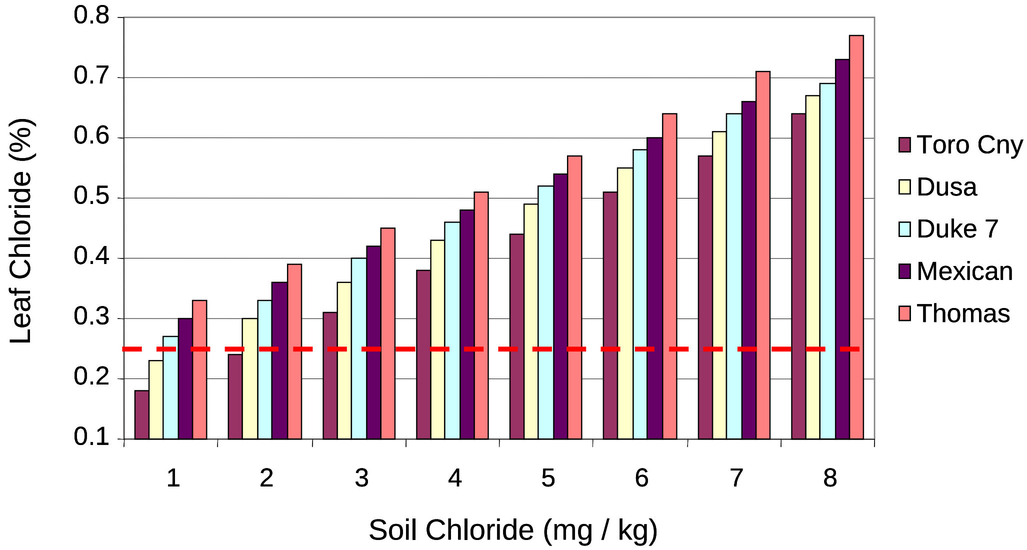

Figure 2. ANN model output showing predicted leaf chloride contents of Hass scions grafted on to five different rootstocks. The additional model parameters are fixed for soil ECe = 4.0 dS/m; water EC 0.8 dS/m; soil pH7; Clay 30%. The dashed bar indicates 0.25% leaf chloride content at which leaf burn symptoms appear.

Soil chloride concentrations exerted the greatest single effect on leaf chloride, which could be partially offset by differences in rootstock selection. At soil chloride concentrations of 1 to 2 mg/kg soil, the leaf values are below the 0.25% threshold for leaf burn for Toro Canyon, whereas Thomas is already above the threshold. As soil chloride levels increase to the maximum observed value of 8 mg/kg, the leaf chloride contents continue to increase in a linear fashion. Across all chloride concentrations, the lowest to highest chloride concentrations are observed for Toro Canyon <Dusa <Duke7 <Mexican <Thomas. The Hass scions on Thomas generally have twice as much chloride as Hass on Toro Canyon.

Surprisingly, soil pH also had a strong effect on leaf chloride contents (Figure 3). In this case, there is no direct mechanistic relationship that can be proposed to explain the data. However, pH affects many different variables and thereby could cause indirect effects that lead to chloride uptake. The pH range that was examined across the transect studied here ranged between 6.5 and 7.5, a difference of one pH unit. Increases in soil pH by one unit resulted in approximately 30% increases in leaf chloride levels. It is tempting to consider soil pH adjustment (decreases) as a management tool for reducing leaf chloride levels and leaf burn symptoms. Nevertheless, experimental validation of this hypothesis will be required before this practice can be recommended.

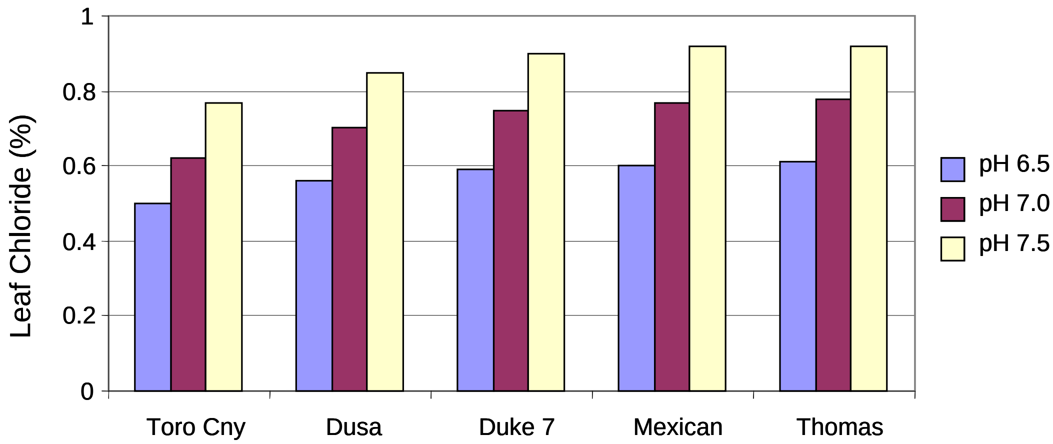

Figure 3. ANN model output showing predicted effect of changes in soil pH on leaf chloride content for five avocado rootstocks. Additional parameters were set under relatively harsh conditions that are associated with elevated chloride levels: soil ECe= 4.0 dS/m, soil Cl 8 mg/kg; irrigation water EC 0.8 dS/m; irrigation water chloride = 50 mg/L; soil clay 50%.

<u>Effect of soil clay content on leaf chloride.</u> Soil clay content also was found to be associated with increases in leaf chloride accumulation (data not shown). Across the transect of orchards, clay contents of the soils varied from 10% to as high as 50%. In all cases, increases in clay content between these two extremes were associated with approximately 20% increases in uptake of chloride. As with pH, there is no mechanistic reason for this effect. However, it can be speculated that heavy clay soils are more difficult to leach and retain greater amounts of water than better drained sandy or loam soils. Combined with short frequent irrigations, clay soils would thus be likely to accumulate chloride and total salts in the upper profile where the roots are most abundant. Hypotheses regarding the effect of soil clay content will be further examined in the remaining project years; for example, interrelationships with soil hydraulic conductivity and salt distribution in the soil profile over time.

<u>Interactions of irrigation water chloride concentration and irrigation water salinity.</u> Results of the first year's data provide an intriguing insight into possible interactions of chloride and salinity on leaf chloride content. Whereas high soil salinity appears to contribute to detrimental effects of high soil chloride leading to increased Cl uptake, the opposite appears to be true with respect to the chemistry of the irrigation water (Figure 4). High irrigation water salinity combined with low chloride concentrations in the water result in much lower uptake of chloride by avocado. As shown in Figure 6, these interactive effects are amplified as the water chloride concentration increases. The fixed variables for the data plotted in the graph are set at intermediate conditions with pH 7, 35% clay, soil ECe 2.0, and soil Cl at 4 mg/kg. Under these conditions, at water chloride concentrations of 70 mg/L, no leaf burn symptoms would be predicted to occur in Toro Canyon grafted trees when the accompanying water salinity has an EC of 1.2 dS/m. On the other hand, the same chloride concentration in

water with an EC of 0.2 is predicted to cause severe leaf burning with leaf chloride concentrations of 0.6%.

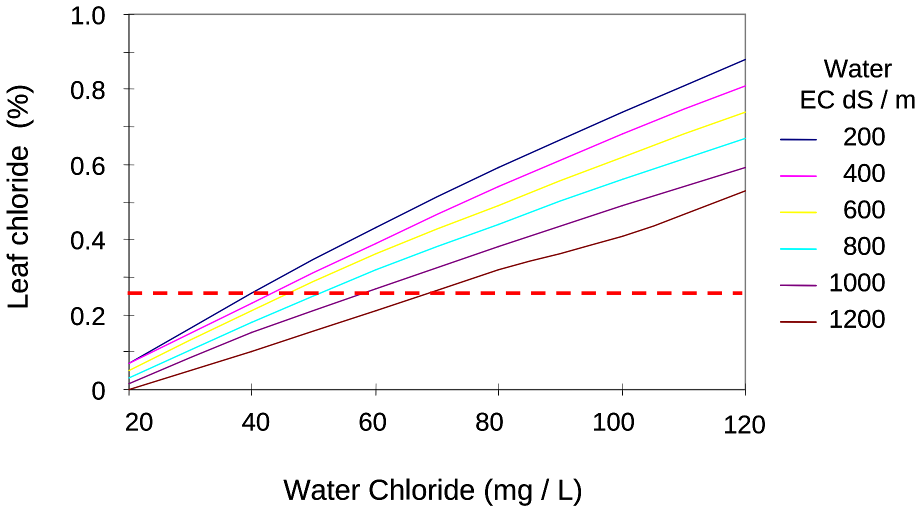

Figure 4. ANN model output illustrating the inverse relationship between irrigation water salinity and chloride concentrations on accumulation of chloride in leaves of Hass avocado grafted on to Toro Canyon rootstock. Fixed model values were set at pH 7, 35% clay, soil ECe 2.0, and soil Cl at 4 mg/kg

Benefits of the Research to the Industry

This research will provide several benefits to the avocado industry. These include:

Cost benefit analysis for irrigation water quality versus fruit yields over the range of salinity levels that occur in water supplies currently used by avocado growers.

Optimization of irrigation regimes for use of saline irrigation waters based on management of chloride versus total dissolved salts.

Basic information on mechanisms of salinity stress and tolerance in avocado rootstocks.

Recommendations for rootstock selections based on field performance data.

Improved guidance to growers for salinity management strategies.

Development of an artificial neural network ANN model, that can be deployed on an internet location for use by growers to examine the effects of salinity, chloride, soil properties, rootstocks and management practices on root growth and yields of avocado in California.

Achievements and Future Prospects

This research is providing a detailed analysis of salinity and chloride effects on avocado yields. Despite the lack of yield data for year one, results from this year's research have already revealed important differences in salinity tolerance between commercially used rootstocks, and the impacts of a wide number of environmental variables on root growth and chloride uptake. As the project proceeds and fruit yield data are obtained this coming year, we will be able to achieve our primary goal of relating salinity management to yield and economic returns. We will further be able to examine the independent and additive effects of chloride and total dissolved salts on avocado yields. With the extensive site characterization data, we will also be able to examine virtually all soil fertility and irrigation management variables on avocado yields. Our novel modeling approach using the powerful new methods with artificial neural networks will allow us to develop an online computer program that will enable growers to enter their own soil and water data to predict avocado yields via a user friendly interface. Through this combination of fundamental and applied knowledge, we may find new ways to manipulate chloride uptake, improve water use efficiency and reduce salinity problems that are currently causing significant yield reductions in avocado orchards.

CAC Salinity Project Activities in 2009

Tasks **Locations and Dates**

Spring grower consults, **Data logger setups** **Spring salinity** measurements, **Sample roots** for mycorrhizae **Soil sample** for PLFA microbial community analyses	**South Counties** Rancho Mission Viejo Woodworth Deardorff Carey	**Central Counties** McFadden Steve Smith Mud Creek Lyle Snow	**North Counties** Miller Abbot Van der Kar	**Morro Bay** Staller Tyson Davis
Harvest Yield Data Collection Data logger backup, site check Combined with May-June visits as feasible	Rancho Mission Viejo Woodworth Deardorff Carey	McFadden Steve Smith Mud Creek Lyle Snow	Miller Abbot Van der Kar	Bob Staller Tyson Davis
Early Summer Soil and Root Samples May-June 09	Rancho Mission Viejo Woodworth Deardorff Carey	McFadden Steve Smith Mud Creek Lyle Snow	Miller Abbot Van der Kar	Bob Staller Tyson Davis
Mid Summer Soil and Root Samples (begin mid July)	Rancho Mission Viejo Woodworth Deardorff Carey	McFadden Steve Smith Mud Creek Lyle Snow	Miller Abbot Van der Kar	Bob Staller Tyson Davis Bartleson
Late Summer Soil and Root Samples (end of August)	Rancho Mission Viejo Woodworth Deardorff Carey	McFadden Steve Smith Mud Creek Lyle Snow	Miller Abbot Van der Kar	Bob Staller Tyson Davis Bartleson
Harvest and Fall Leaf Tissue and Soil Sampling for Full Chemical Analysis	Rancho Mission Viejo Woodworth Deardorff Carey	McFadden Steve Smith Mud Creek Lyle Snow	Miller Abbot Van der Kar	Bob Staller Tyson Davis Bartleson

Additional Project Tasks Completed in 2009:

Jan – May 2009	Data summarization, Statistical Analyses, Model Development
Feb 2009	Grant proposal to UC-Mexus program for visiting researcher Marcario Bacillio to investigate bacterial inoculants for enhanced salinity tolerance
March 2009	Reinstall new data logger at Deardorf orchard
	Set up new site at Mud Creek Ranch, Santa Paula, organic grove TC, Thomas, DUSA
	Set up new installations at grove with Mexican rootstocks Nipomo, San Luis Obispo areas
April	PLFA analysis of salinity effects on microbial community structure and mycorrhizae
May 2009	Midyear Report
June 2009	Field Project Review: Cultural Management and Physiology Subcommittee (Tentative plan for RMV San Diego) Members: Gary Nichols, Ed McFadden, Lyn Francis, Darryl Nelson
June 12-14	Grower Meetings Temecula, Santa Paula, Santa Barbara
July 1	Salinity Article for Avocado Yearbook