

Zinc Fertilization of Avocado Trees

David E. Crowley¹ and Woody Smith²

Department of Soil and Environmental Sciences, University of California, Riverside, CA 92521

Ben Faber³

University of California, Cooperative Extension, 800 South Victoria Avenue, L#1280, Ventura, CA 93009

John A. Manthey⁴

Fruit and Vegetable Chemistry Laboratory, 263, South Chester Avenue, U.S. Department of Agriculture, Agricultural Research Service, Pasadena, CA 91106

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Abstract. Methods for Zn fertilization of 'Hass' avocado (*Persea americana* Mill.) trees were evaluated in a 2-year field experiment on a calcareous soil (pH 7.8) in Ventura County, Calif. The fertilization methods included soil- or irrigation-applied ZnSO₄; irrigation-applied Zn chelate (Zn-EDTA); trunk injection of Zn(NO₃)₂, and foliar applications of ZnSO₄, ZnO, or Zn metalosate. Other experiments evaluated the influence of various surfactants on the Zn contents of leaves treated with foliar-applied materials and on the retention and translocation of radiolabeled ⁶⁵ZnSO₄ and ⁶⁵Zn metalosate after application to the leaf surface. In the field experiment, tree responses to fertilization with soil-applied materials were affected significantly by their initial status, such that only trees having <50 µg·g⁻¹ had significant increases in foliar Zn contents after fertilization. Among the three soil and irrigation treatments, ZnSO₄ applied at 3.2 kg ZnSO₄ per tree either as a quarterly irrigation or annually as a soil application was the most effective and increased leaf tissue Zn concentrations to 75 and 90 µg·g⁻¹, respectively. Foliar-applied ZnSO₄, ZnO, and Zn metalosate with Zn at 5.4, 0.8, and 0.9 g·liter⁻¹, respectively, also resulted in increased leaf Zn concentrations. However, experiments with ⁶⁵Zn applied to leaves of greenhouse seedlings showed that <1% of Zn applied as ZnSO₄ or Zn metalosate was actually taken up by the leaf tissue and that there was little translocation of Zn into leaf parenchyma tissue adjacent to the application spots or into the leaves above or below the treated leaves. Given these problems with foliar Zn, fertilization using soil- or irrigation-applied ZnSO₄ may provide the most reliable method for correction of Zn deficiency in avocado on calcareous soils.

Zinc deficiencies are common in many subtropical areas where avocados are grown and are suspected to be an important limiting factor in fruit quality and tree health (Crowley, 1992; Piccone et al., 1985). Deficiencies of Zn typically are associated with calcareous soils in which availability of the metal is limited by its extremely low solubility at alkaline soil pH. Under these circumstances, applications of inorganic fertilizers, such as ZnSO₄, result in temporarily increased availability until the metal precipitates out of solution as the poorly soluble oxide. Zinc deficiencies also have

been reported to occur in acid, sandy soils that have low total Zn contents and in warm, semi-arid regions that typically have low soil organic matter. In these cases, the Zn is relatively soluble but is leached out of the surface soil occupied by the feeder roots. Thus, the strategies that should be used for correction of Zn deficiency depend largely on the soil pH and, perhaps, on the amount of organic matter available to form metal complexes (Labanauskas et al., 1959; Srivastava et al., 1981). Other factors that influence Zn deficiency are high levels of N and P fertilizers (Labanauskas et al., 1959) and seasonal fluctuations in leaf micronutrient content (Bingham, 1961; Labanauskas et al., 1961) that may be related to irrigation and climatic factors affecting root growth and nutrient uptake.

Foliar symptoms of Zn deficiency are manifested in the new leaf tissue by a reduction in leaf size and by development of "mottleleaf," which is characterized by interveinal leaf yellowing caused by impaired chlorophyll synthesis. Other symptoms include shortened internode length on the branches, reduced fruit size, and in 'Hass' avocado, the production of round misshapened fruit. The critical Zn level

in the leaf tissue has been established at 20 µg/g leaf dry weight (Goodall et al., 1979), with 30 to 150 µg·g⁻¹ considered to be normal. Using traditional sampling methods, Zn deficiencies are not always diagnosed easily, with bulk samples collected from the entire orchard, because affected trees frequently occur in clustered groups where the soil is calcareous, or on trees that have feeder roots damaged by phytophthora root rot (Whiley et al., 1987). In some cases, visual leaf yellowing symptoms attributed to Zn deficiency also may be confused with Fe deficiency, which produces somewhat similar leaf symptoms and can occur simultaneously on calcareous soils. As a result of this confusion and the historical problem with Zn deficiency, avocado growers have used a variety of methods with inconsistent success or they may fertilize an entire orchard to correct a deficiency that is apparent in only a few highly visible trees.

Several methods have been developed to correct Zn deficiency, including foliar applications of ZnSO₄ and Zn chelates (Goodall et al., 1979; Lee, 1973), trunk injections (Whiley et al., 1991), or soil applications of Zn fertilizers (Embleton et al., 1966; Wallihan et al., 1958). There is no consensus as to which method is the most effective, particularly for orchards on calcareous soils, or which materials are best used with the various application techniques. Foliar application of Zn fertilizers by helicopter is one of the more common methods used in southern California because of the difficulty of the terrain. However, there is concern as to whether Zn applied to the outer leaf canopy is translocated to the inner canopy, developing fruit, and roots (Kadman and Lahav, 1978). In some cases, avocado trees produced small, round fruit typical of Zn deficiency even after the foliage was treated with foliar Zn (Len Francis, personal communication).

Prior studies on foliar Zn fertilization of various plants have shown relatively little translocation of foliar-applied Zn when applied either as ZnSO₄ or after chelation with synthetic chelate, such as EDTA. For example in citrus, quantities of Zn and Mn translocated from sprayed to new leaves treated with ZnSO₄ increased foliar Zn concentrations by 2 to 5 mg·kg⁻¹, but only in some years and was considered a negligible effect (Swietlik and LaDuke, 1991). In pea (*Pisum sativum* L.), only 25% and 75% of Zn applied as Zn-EDTA or ZnSO₄, respectively, was recovered after removal of epicuticular waxes, of which ≈8% to 10% was translocated from the treated tissue (Ferrandon and Chamel, 1989). In the only available study on avocado (Kadman and Lahav, 1978), there was no translocation of ⁶⁵ZnCl₂ from spots applied to intact leaves, even to adjacent parenchyma tissue. Among the various foliar materials we tested, amino acid chelates (metalosates) have been taken up and translocated more effectively than inorganic metal salts or the synthetic chelate EDTA in a variety of crops and trees (Hsu, 1986; Shazly, 1986), but to our knowledge, there have been no comparisons of foliar-applied materials for avocado. Recognizing that there are a variety of problems associated with evalu-

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¹Assistant Professor; to whom reprint requests should be addressed.

²Staff Research Associate.

³Farm Advisor.

⁴U.S. Dept. of Agriculture Research Chemist.

Table 1. Schedule of application methods, Zn materials, and application rates for treatments used to correct Zn deficiency in 'Hass' avocado on a calcareous soil.

Application method	Zn		Application	
	material	% Zn	Rate	Timing
Control	NA ²		NA	
Foliar ³	Sulfate	36	15 g·liter ⁻¹	Once per year applied June 1993 and 1994
	Metalosate	6.8	11.7 ml·liter ⁻¹	Once per year applied June 1993 and 1994
	Zintrac 8	40	2.3 ml·liter ⁻¹	Once per year applied June 1993 and 1994
Trunk injection	Nitrate	34	10% Zn(NO ₃) ₂ at 15 ml·m ⁻¹ diameter	One time injection Oct. 1992
Simulated irrigation	Sulfate	36	794 g/tree	Quarterly, applied Oct. 1992; Jan., Apr., July, and Oct. 1993; and Feb., Apr., and July 1994
	Chelate	14	42 g/tree	Quarterly, applied Oct. 1992; Jan., Apr., July, and Oct. 1993; and Feb., Apr., and July 1994
Soil banding	Sulfate	36	3.2 kg/tree	Once per year applied Oct. 1992 and Feb. 1994

²NA = not applicable.

³Foliar materials applied at 900 liters·ha⁻¹.

ating avocado tree responses to foliar Zn fertilizers, this research combined field and laboratory studies to compare the efficacy of soil and foliar Zn fertilization methods for correction of Zn deficiency in avocado.

Materials and Methods

Zinc application in orchard. A mature orchard (>15 years old) planted with 'Hass' avocado on Mexican rootstocks was selected after a survey of Zn-deficient orchards throughout Ventura County, Calif. The orchard, under commercial management, was located on a moderately sloping (15% to 30%) hillside on the Las Posas Hills adjacent to the Santa Rosa Valley. Soil on this site was characterized as a Soper loam containing free CaCO₃, which was 60 to 150 cm deep over conglomerate rock. Surface soil (0 to 20 cm) pH values ranged from 7.8 to 8.0 and were buffered by 0.1% to 3% soil carbonate. Patchy areas containing CaCO₃ were associated with chlorotic trees that were subsequently determined to be Fe and Zn deficient. At the start of the experiment, all of the trees were permanently numbered and mapped, and baseline nutrient analyses were conducted using leaf samples from individual trees. To eliminate possible problems with Fe and Mn deficiency, the trees were fertilized with Fe-ethylenediamine di(o-hydroxyphenylacetic acid) (EDDHA) (Libfer 6% Fe; Allied Colloids, Brampton, Ont., Canada) and Mn-EDTA (12% Mn; W.R. Grace and Co., Lexington, Mass.) at 142 and 57 g/tree, respectively, using a banded application 0.3 to 1.5 m from the trunk.

The experiment involved a completely randomized design for the soil- and irrigation-applied materials and individual sets of similar trees for the foliar treatments. The Zn materials included soil- or irrigation-applied ZnSO₄ (36% Zn); irrigation-applied Zn-EDTA (14% Zn) (Librel Zn; Allied Colloids); trunk injection of Zn(NO₃)₂ (3.5% Zn); and foliar-applied ZnSO₄, ZnO (40% Zn) (Zintrac-8; Shield Brite Corp., Kirkland, Wash.), or Zn metalosate (6.8% Zn) (Albion Laboratories; Clearfield, Utah) using the timing and quantities described in Table 1. Application methods and timing were based on recommended procedures previously developed by farm advisors and commercial applicators or were recommended by the manufacturers of the vari-

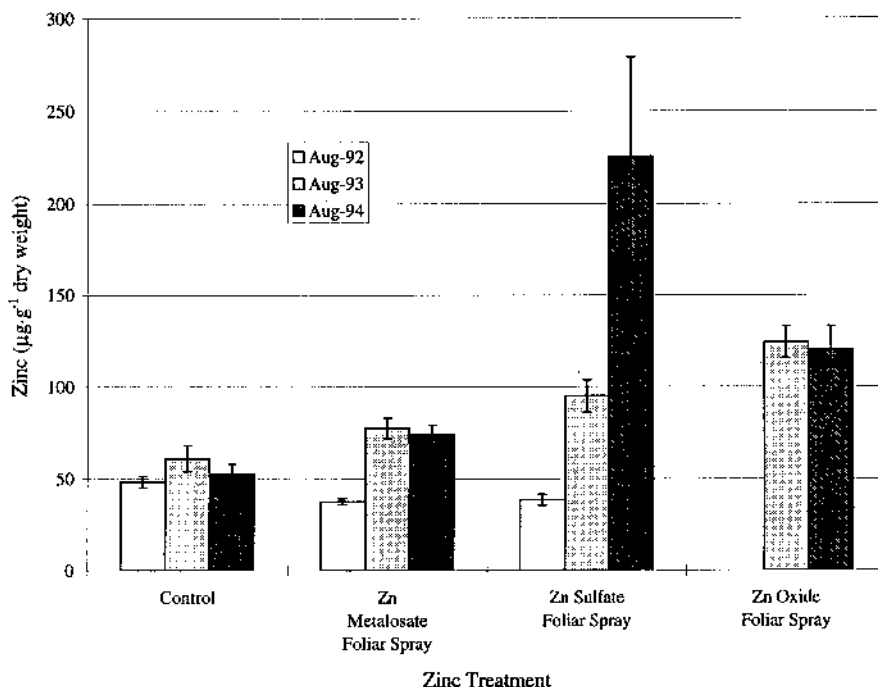


Fig. 1. Zinc concentrations in 'Hass' avocado leaves after foliar application of Zn metalosate (8×10^{-4} kg Zn/liter), ZnSO₄ (5.4×10^{-3} kg Zn/liter), and ZnO (9×10^{-4} kg Zn/liter). Vertical bars represent SES of the means within each treatment. All sprays were applied to the canopy to runoff using a commercial spray applicator from ground level. The surfactants, Sun-It II and Kinetic, were used in 1993 and 1994, respectively.

ous compounds. Soil-applied materials were placed in a 1-m-wide band in the minisprinkler irrigation-wetted zone 0.5 to 1.5 m from the base of the trunk. Irrigation-applied Zn fertilizer was dissolved in water and applied to the entire sprinkler-wetted soil zone using a watering can. Trunk injection was accomplished using a propane gas-powered gun inserted into a predrilled hole (6 mm in diameter) at the base of each of the main scaffold limbs (Phillips Gas Gun, N.J. Phillips PTY, Somersby, New South Wales, Australia). Injections were conducted early in the morning during the period of active leaf transpiration to reduce back pressure and facilitate rapid uptake into the limbs. Foliar fertilizers were applied to runoff using a gasoline-powered, ≈760-liter spray rig operated at ground level.

Eighteen replicate trees randomly were assigned to each treatment for the soil- and irrigation-applied materials. The number of replications was reduced to 14 to 16 trees for

some treatments following an orchard thinning in 1993. Foliar analyses on individual trees receiving either foliar- or soil-applied Zn were conducted yearly to monitor the efficacy of each treatment and the degree of variation that occurred among trees using the various fertilization methods. Yields of 'Hass' or 'Fuerte' avocado trees previously have been unaffected by correcting Zn deficiency, as documented by a field study conducted over 6 years (Kadman and Lahav, 1978). Thus, data reported in our research focused on changes in foliar Zn content to compare the efficacy of the Zn fertilizer materials when applied to 'Hass' avocado through the soil and the irrigation water or as a foliar spray. All data were subjected to analysis of variance (ANOVA) using a computer software program (Systat, Evanston, Ill.). When ANOVA generated a significant F value, mean separations were determined by pairwise *t* tests for the individual trees in each treatment. In the green-

house experiment with ⁶⁵Zn isotope-labeled fertilizers, the data were analyzed by Kruskal-Wallis ANOVA on Ranks, with mean separation by Student-Newman-Keuls all pairwise multiple comparison procedures (Sigma Stat; Jandel Scientific Software, San Rafael, Calif.).

A preliminary experiment examined the Zn, Fe, and Mn concentrations for 20 individual leaves collected from four trees to ascertain the appropriate sample size to minimize variation within bulk samples collected from individual trees. Using a sample-size statistical formula, a minimum of 25 leaves per tree was required for most trees to generate a mean value with a 10% error. Leaf washing procedures used for routine analyses of trees analyzed from Aug. 1992 to Aug. 1993 followed industry standard procedures (Chapman et al., 1978) that use a dilute soap-and-water solution, followed by a tap water and three deionized water rinses. In the foliar fertilization experiment conducted in 1994 to examine surfactants, more stringent procedures were tested in which leaves were washed in a dilute soap-and-water solution, followed by a tap water rinse, and a final 0.1 N hydrochloric acid wash for varying intervals ranging from 15 sec to 5 min to dissolve Zn adhering to the leaf surface. Hydrochloric acid washing previously has been highly effective for removing surface precipitated metals after foliar applications (Ferrandon and Chamel, 1989).

To examine how much Zn was associated with the epicuticular waxes, subsamples of acid-washed leaves were rinsed in deionized water and further treated with hexane. Subsequent procedures, developed from the leaf-washing experiments and used for the surfactant experiments and the main field experiment in 1994, used a dilute soap-and-water wash followed by a tap-water rinse, a deionized-water rinse, a 1-min acid rinse, and three deionized-water rinses.

Leaves processed for analyses after washing were dried in paper bags in a forced-air oven at 60C for 3 days, after which they were ground with a Wiley mill to pass a 1-mm mesh sieve screen. Weighed subsamples were transferred to Teflon vessels for microwave digestion in 5, 2, and 1 ml of concentrated HNO₃, 30% H₂O₂, and deionized water, respectively (Sah et al., 1992). The samples were heated under pressure for 15 min, after which the liquid digests were cooled to room temperature, transferred to 25-ml volumetric flasks, and diluted to a final volume of 25 ml with deionized water. Samples were analyzed using an atomic absorption flame spectrophotometer (model 5000; Perkin Elmer, Norwalk Conn.). Reagent blanks and apple (*Malus domestica* Borkh.) leaves (model SRM 1515; National Institute of Standards and Technology, Gaithersburg, Md.) were included with each analysis for quality assurance. Measured values for the standard reference materials were generally within 5% of the reported concentrations.

Evaluation of surfactants. A second experiment, conducted at the Univ. of California Agricultural Expt. Station in Riverside (UCR),

examined the effects of three surfactants used in combination with each of the foliar spray fertilizers. All of the materials were applied in Apr. 1994 to newly expanded 'Hass' avocado leaves. The surfactants included a methylated seed oil adjuvant, Sun-It II (American Cyanamid, Grand Forks, Nevada), the industry standard nonionic polyoxyethylenesorbitan detergent Tween 20 (Sigma Chemicals, St. Louis), and a nonionic silicone oil-based surfactant, Kinetic (Setre Chemical Co., Memphis). Twelve leaves per treatment were tagged and sprayed in early morning with each of the

three Zn fertilizers in combination with each surfactant or with no surfactant. Three of the leaves from each treatment were collected immediately after spraying and after 1, 2, and 4 weeks to assess changes in Zn content over time. Leaves were washed sequentially, subsampled at each step, and processed individually for analysis by flame atomic absorption spectroscopy.

Distribution of radiolabeled Zn. Uptake and translocation of foliar-applied ZnSO₄ and chelated Zn metallosate were evaluated using the radioisotope ⁶⁵Zn (Dupont, Wilmington,

Table 2. Foliar concentrations of Zn for trees with low and high Zn contents before fertilization with soil- and irrigation-applied Zn fertilizers in 1992 and after fertilization in 1993 and 1994.

Treatment	No. ^z	Foliar Zn concn (µg·g ⁻¹)			
		Initial tree status	1992 ^y	1993	1994
None (control)	8	< 50	38 (1.9)	40 (2.8)	37 (3.0)
	10	≥50	57 (1.9)	54 (6.7)	43** (2.1)
Zn-EDTA irrigation	6	< 50	41 (3.3)	73* (14.1)	57 (14.1)
	10	≥50	58 (1.6)	50 (3.9)	44 (3.1)
ZnSO ₄ irrigation	9	<50	37 (2.0)	75** (5.4)	86** (7.6)
	5	≥50	57 (2.1)	56 (5.8)	55 (4.7)
ZnSO ₄ soil banded	12	< 50	38 (1.8)	86** (8.9)	99** (9.4)
	4	≥50	58 (2.5)	103 (32.2)	61 (21.4)

^zNumber of replicate trees after random assignment to the various fertilizer treatments.

^yValues for trees having low and high Zn contents in 1992 are before fertilization with Zn. Values in parentheses are SE of the mean.

*,**Significant at P ≤ 0.05 or 0.01, respectively, between 1992 and 1993 and between 1992 and 1994 using a pairwise t test comparing changes in foliar Zn concentrations for individual trees.

Table 3. Two-way analysis of variance of avocado tree responses to soil fertilizer treatments and effect of initial tree status in 1992 before fertilization.

Source	Analysis of variance				
	Sum of squares	df	Mean square	F ratio	P
Treatment	13,097	3	4,365	8.963	0.000
Category ^z	4,750	1	4,750	9.753	0.003
Treatment × category	4,093	3	1,364	2.801	0.048
Error	26,790	55	487		

^zTrees were divided into two categories depending on their initial foliar Zn contents: trees with <50 µg·g⁻¹ and trees initially having >50 µg·g⁻¹.

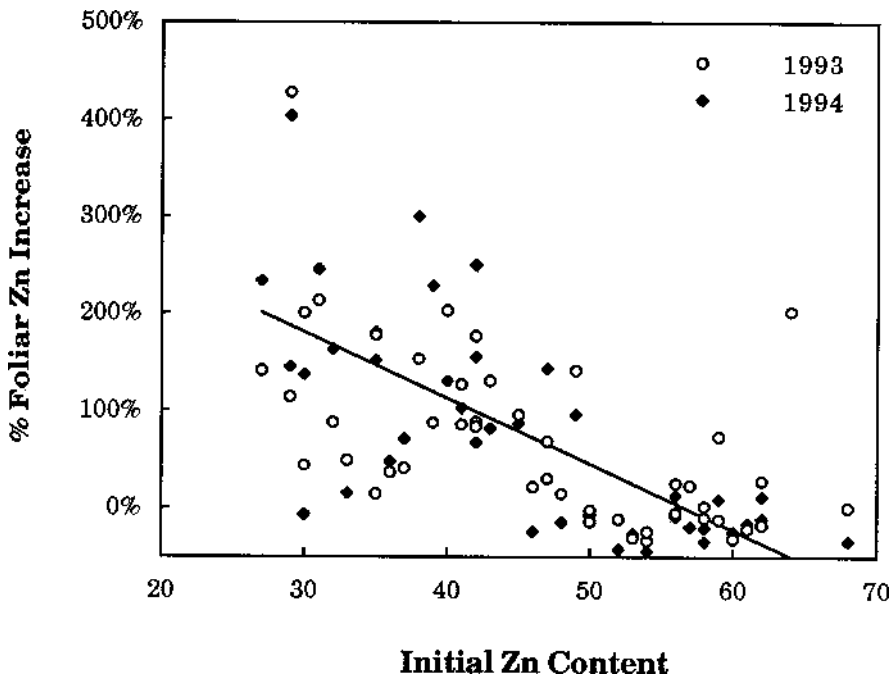


Fig. 2. 'Hass' avocado responses to soil applications of Zn fertilizers as affected by their initial Zn status. Line was fit to data points by regression analysis (r = 0.704).

Del.) with 6-month-old, container-grown avocado seedlings. The isotope was purchased as Zn chloride in 0.5 N HCl (specific activity $5.5 \times 10^8 \text{ Bq}\cdot\text{mg}^{-1}$). Zinc metalosate was prepared by complexation with a proprietary amino-acid hydrolysate by Albion Laboratories, who provided technical support and materials used for preparation of the radiolabeled chelate at UCR. Zinc metalosate contains 7% Zn by weight with a specific gravity of 1.25. The Zn complex, containing $3.7 \times 10^6 \text{ Bq}\cdot\text{ml}^{-1}$, had a specific activity of $43.7 \text{ Bq}\cdot\mu\text{g}^{-1}$ Zn. Radiolabeled ZnSO_4 was prepared at a 6.5% (v/v) concentration by adding 20 μl of $^{65}\text{ZnCl}_2$ to 180 μl of 1.1 M ZnSO_4 . Taking into account the concentration of HCl added with the isotope, the final solution was $\approx 95\%$ ZnSO_4 and 5% ZnCl_2 and had a specific activity of $57 \text{ Bq}\cdot\mu\text{g}^{-1}$ Zn.

To examine differential absorption into the upper or lower sides of the leaves, 1-cm² spots of the radiolabeled materials were applied to individual leaves of greenhouse, container-grown avocado seedlings using five replicate trees per treatment. Mean dry weight of the treated spots was $8 \text{ mg}\cdot\text{cm}^{-2}$. Leaves selected for treatment were all newly expanded leaves located three leaves down from the stem apex. After the leaves were treated in the morning, the materials were allowed to absorb into the leaf tissue for 3 days. The treated leaves then were analyzed individually by removing the leaf and excising the application spot with a razor blade. The excised tissue containing the application spots was treated by washing in vials containing 10 ml soap and water for 1 min, followed by a 10-ml water rinse; a 1-min, 10-ml acid wash; and a final water rinse, using separate vials for each sample at each step. Each of the wash solutions was analyzed for ^{65}Zn by liquid-scintillation counting of 1-ml samples to determine a mass balance for the isotope. Translocation into adjacent leaves was measured by sampling of the first leaf immediately above and below the treated leaf, the leaf tissue adjacent to the application spot, and the stem apex of each plant. Leaf content of ^{65}Zn was determined using standard procedures with a liquid-scintillation counter (model LS5000 TD; Beckman, Fullerton, Calif.).

Results and Discussion

Zn application in orchard. The mean foliar Zn content for all trees in this orchard before fertilization was $\approx 45 \mu\text{g}\cdot\text{g}^{-1}$, although several individual trees were marginally Zn deficient (normal Zn range = 30 to $150 \mu\text{g}\cdot\text{g}^{-1}$). In our experience, variability in foliar Zn concentrations in an orchard and within the canopy of individual trees is typical of Zn and Fe deficiency problems and appears to be associated with high CaCO_3 concentrations in calcareous soils. We report our leaf analysis data for 1992 (see Fig. 1 and Table 2) as the statistical means for each treatment before applying the fertilizer materials, whereas data for Aug. 1993 and 1994 are for the same trees after fertilization. An initial analysis of the data revealed that trees with $>50 \mu\text{g}\cdot\text{g}^{-1}$ did not respond significantly to any of the soil-applied fertilizer ma-

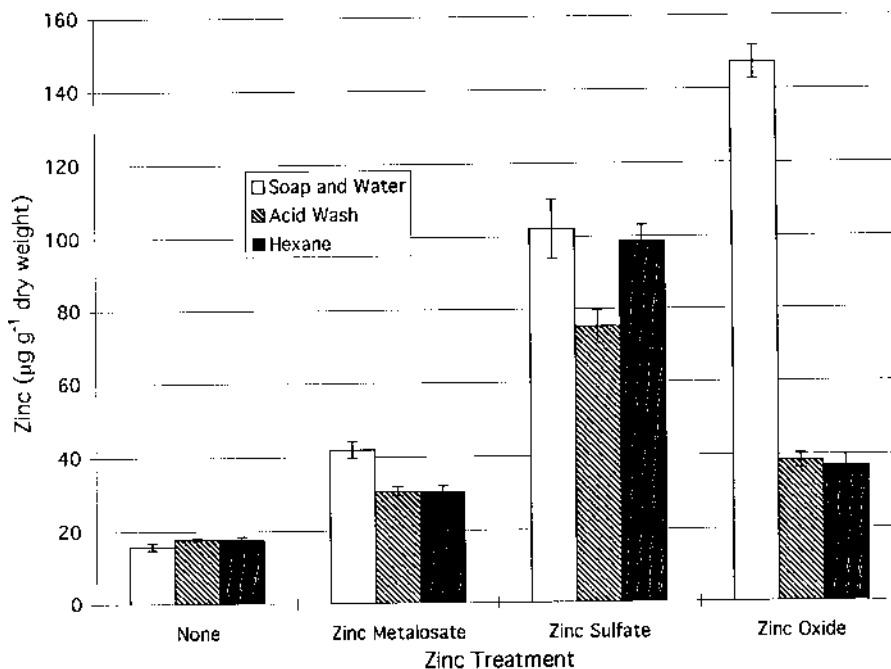


Fig. 3. Successive washing treatments for avocado leaves for removal of extracellular Zn applied as Zn metalosate, ZnSO_4 , or ZnO after foliar application in a water base with no surfactant. Vertical bars represent SEM of the means within each treatment. Leaves were processed 4 h after application.

Table 4. Wash removal and leaf retention (percent of that applied) of foliar-applied, radiolabeled ZnSO_4 and Zn metalosate by leaves of greenhouse-grown avocado seedlings 3 days after application to the adaxial or abaxial side of the leaf surface.

Removal method	ZnSO_4		Zn metalosate	
	Adaxial	Abaxial	Adaxial	Abaxial
Soap and water	96.5 a (2.5) ^a	96.8 a (0.9)	98.4 a (0.6)	92.8 b (2.8)
Water rinse	1.7 a (1.8)	1.3 a (0.5)	0.8 a (0.2)	1.3 a (0.4)
Acid wash	1.0 ab (0.5)	1.0 ab (0.2)	0.6 b (0.4)	1.6 a (0.8)
Retained by leaf	0.7 a (0.4)	0.9 a (0.1)	0.2 a (0.1)	4.2 b (2.5)

^aValues in parentheses are SE of the means. Mean separation within rows by Tukey's honestly significant difference at $P \leq 0.05$.

terials (Table 3) and that the relative response with respect to percent increase in foliar Zn was correlated ($r = 0.70$) with the initial Zn content of the trees before fertilization (Fig. 2). All subsequent comparisons of treatment means for soil- or irrigation-applied Zn were filtered for trees having low and high initial Zn concentrations (Table 2).

Almost all of the Zn fertilization methods resulted in increased foliar Zn concentrations, with the exception of irrigation-applied Zn-EDTA in 1994 and trunk injection of $\text{Zn}(\text{NO}_3)_2$. Trunk injection was tested only one time (Fall 1992) and gave a short-term response in which foliar Zn concentrations transiently increased from 40 to $65 \mu\text{g}\cdot\text{g}^{-1}$ in the spring but dropped to control levels by the following August. The 6-mm holes that were drilled into each of the major scaffold limbs healed slowly and continued to exude sap for several months. This treatment was judged to be potentially dangerous because it could allow introduction of *Phytophthora citricola* into the wounds, which is associated opportunistically with trunk wounding and desuckering in California avocado orchards (El-Hamalawi et al., 1994). Therefore, the treatment was discontinued.

Applications to soil. Among the three soil and irrigation treatments, the least effective

and most expensive was Zn-EDTA chelate applied at the relatively low rate (43 g Zn per tree) recommended by the manufacturer. Leaf Zn concentrations of trees initially having Zn at $<50 \mu\text{g}\cdot\text{g}^{-1}$ increased from a mean level of 41 to 73 and $57 \mu\text{g}\cdot\text{g}^{-1}$ in 1993 and 1994, respectively, after fertilization with Zn-EDTA, but this increase was highly variable and not significantly different ($P > 0.05$) from the Zn in the unfertilized control trees (Table 2). One reason for this result may be displacement of Zn from the chelate by Ca (Norvell et al., 1969), which was present in large quantities in the soil as CaCO_3 .

The most effective soil fertilizer was ZnSO_4 applied at an annual rate of 3.2 kg ZnSO_4 per tree, either quarterly as a simulated irrigation or annually as a single soil application, which increased foliar Zn concentrations in low-Zn-status trees to 75 and $86 \mu\text{g}\cdot\text{g}^{-1}$ in 1993 and 86 and $99 \mu\text{g}\cdot\text{g}^{-1}$ in 1994. In contrast, there were no significant increases in foliar Zn for trees initially having Zn at $>50 \mu\text{g}\cdot\text{g}^{-1}$ (Table 2). One possible exception was noted for trees receiving ZnSO_4 by soil banding in which Zn levels increased from 58 to $103 \mu\text{g}\cdot\text{g}^{-1}$ in 1993. These levels decreased in the following year to $61 \mu\text{g}\cdot\text{g}^{-1}$. Due to the few trees in the high-Zn category—treatment combination ($n = 4$), the

transient increase of 1993 was not statistically significant.

The lack of response of trees in the initially high-Zn category and the lack of further increase in foliar Zn after the second fertilizer applications suggests that avocado roots may regulate Zn uptake to maintain levels between 50 and 100 $\mu\text{g}\cdot\text{g}^{-1}$ in the foliage. The longevity of the soil fertilizer treatments needs to be monitored to determine the frequency of fertilizer application that is necessary to maintain adequate foliar Zn contents. An important consideration with long-term use of the soil treatments is the total quantity of Zn that is being applied. After two successive applications, 2.4 kg of actual Zn had been applied per tree, which, in a 10-m² area, is equivalent to 144 $\mu\text{g}\cdot\text{g}^{-1}$ in the upper 10 cm of soil. Continued annual applications may exceed the legally allowable quantities of Zn, which at high levels becomes a soil pollutant. Current Environmental Protection Agency (EPA) guidelines for Zn are 2800 $\text{g}\cdot\text{g}^{-1}$ soil as a pollutant concentration limit and an annual pollutant loading at 140 $\text{kg}\cdot\text{ha}^{-1}\cdot\text{yr}^{-1}$, or 63 $\mu\text{g}\cdot\text{g}^{-1}$ when applied as an organic soil amendment (EPA, 1994). Once the Zn has been applied to the soil, other options would be to lower the surface soil pH by applying granular S or using acidic N or P fertilizers supplied with the irrigation water.

Foliar treatments. By weighing representative leaves before and after spraying, we determined that as much as 1 ml of spray may be retained on the surface of an individual leaf. If all of this material dried on the leaf surface and is not removed by washing, this wetness would result in more than several thousand micrograms Zn per gram associated with the leaf tissue. Thus, there is an inherent problem in evaluating the efficacy of foliar-applied materials because any residual extracellular Zn may be artifactually interpreted as an increase in leaf tissue Zn content. During this research, various leaf washing methods were assessed to determine how much of the foliar-applied Zn could be removed from the leaf surface using industry standard procedures: 1) a soap-and-water wash, 2) more stringent washing with hydrochloric acid, or 3) removal of the cuticle by treatment with hexane. We used highly controlled conditions in which individual leaves of trees at the UCR field station were treated with fertilizers sprayed on to the upper leaf surfaces. Preliminary experiments showed that soap-and-water washing of the tissue resulted in the removal of >95% of foliar-applied Zn but left residual Zn that could be removed by a subsequent acid-wash of the leaves (Fig. 3). This aspect was particularly evident for leaves treated with ZnO, which contained Zn at 145 $\mu\text{g}\cdot\text{g}^{-1}$ after soap and water but only 40 $\mu\text{g}\cdot\text{g}^{-1}$ after a subsequent acid wash. No further decreases in Zn content occurred with subsequent washing steps or after removal of the cuticle by treatment with hexane. Results were similar with respect to leaf washing in the experiment with ⁶⁵ZnSO₄ and ⁶⁵Zn metalosate (Table 4). The small amount of foliar Zn that remained on the leaf represented from 10 to 80 $\mu\text{g}\cdot\text{g}^{-1}$, of which an

unknown proportion would be metabolically available. Thus, analysis of sprayed leaf tissue does not indicate physiological effectiveness of the spray.

Results of the field experiment at the Ventura County field site in 1993 and 1994 showed that all three of the foliar Zn fertilizers resulted in an increase in foliar Zn concentrations at the time of leaf analysis in August, 4 months after the materials had been applied (Fig. 1). When applied with a methylated, seed-oil adjuvant (Sun-It II), the mean leaf Zn concentrations in 1993 were 75, 100, and 125 $\mu\text{g}\cdot\text{g}^{-1}$ for Zn metalosate, ZnSO₄, and ZnO, respectively. In 1994, a different surfactant, (Kinetic) with better dispersion properties was used but resulted in similar Zn contents to those measured the previous year, except for ZnSO₄, which increased leaf Zn concentrations to 220 $\mu\text{g}\cdot\text{g}^{-1}$.

Surfactant effects on foliar Zn absorption. Among the three surfactants we tested, the organosilicone-based material Kinetic gave superior leaf coverage, which is one of the most important properties of a good surfactant. However, with one exception, the individual surfactants had no statistically significant effects on foliar Zn contents immediately after application or at any of the subsequent

leaf sampling dates. Because there were no statistically significant changes over time, the cumulative data for all sampling dates were reduced to a single mean for each of the surfactant-fertilizer treatments (Fig. 4). Zinc in control leaves was at $\approx 32 \mu\text{g}\cdot\text{g}^{-1}$, whereas leaves treated with ZnSO₄, ZnO, and Zn metalosate had mean Zn concentrations of 94, 43, and 47 $\mu\text{g}\cdot\text{g}^{-1}$, respectively, when applied in water with no surfactant. With Zn metalosate, the surfactants Tween 20 and Sun-It II actually appeared to inhibit absorption of the fertilizer ($P = 0.12$). The only positive benefit of a surfactant with any of the fertilizer materials was Sun-It II with ZnO. Here, foliar Zn concentrations increased to 97 $\mu\text{g}\cdot\text{g}^{-1}$, which was significantly different ($P > 0.01$) from all other surfactants with this fertilizer. One possible reason for this effect might be that the insoluble ZnO was partially solubilized to form a water-soluble complex with components of the surfactant.

To study leaf retention and translocation of Zn provided as foliar-applied ZnSO₄ and Zn metalosate, experiments were conducted with ⁶⁵Zn-radiolabeled materials applied as spots to either the top or bottom of the leaf surfaces of greenhouse, container-grown trees. Results of these experiments showed that generally <1%

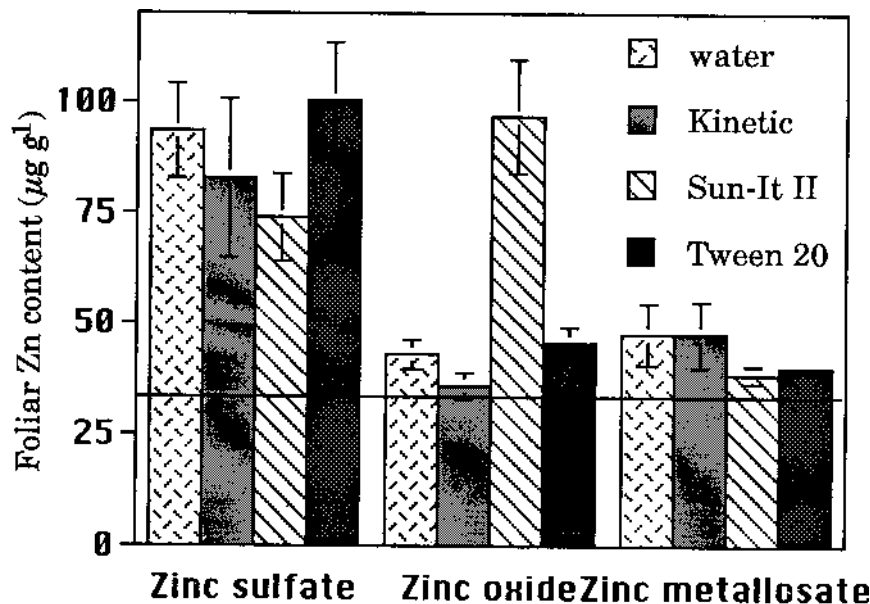


Fig. 4. Zinc contents of avocado leaves treated with ZnSO₄, ZnO, or Zn metalosate as affected by various surfactants. Vertical bars represent SEs of the means within each treatment. Horizontal line indicates the Zn content of nontreated control leaves that had a mean Zn content of 32 $\mu\text{g}\cdot\text{g}^{-1}$ and a SE of 1.3. There were no statistically significant effects of the surfactants, except for the treatment with Sun-It II when used with ZnO ($P \leq 0.05$ by Tukey's honestly significant difference).

Table 5. Translocation and quantities of Zn (in nanograms) retained by avocado leaves treated with radiolabeled ZnSO₄ or Zn metalosate after application to the adaxial or abaxial leaf surfaces of greenhouse, container-grown seedlings.

Removal method	ZnSO ₄		Zn metalosate	
	Adaxial	Abaxial	Adaxial	Abaxial
Stem apex	50 a (20) ²	18 a (5)	11 a (4)	37 a (26)
Leaf, adaxial	47 a (23)	37 a (8)	19 a (4)	22 a (5)
Leaf, abaxial	30 a (9)	26 a (7)	24 a (5)	38 a (6)
Adjacent tissue	223 a (55)	95 a (54)	16 b (5)	394 a (118)
Application spot	276 a (51)	1,700 b (306)	586 a (105)	13,995 c (3057)

²Values in parentheses are SEs of the means. Mean separation within rows by Student-Newman-Keuls all pairwise multiple comparison procedures at $P \leq 0.05$.

of the applied Zn was retained by the leaf surface after the treated spots were washed in soap and water followed by an acid wash (Table 4). One exception was with Zn metalosate, in which 3.8% of the total material applied was retained when the fertilizer was applied to the bottom side of the leaf. After determining leaf weight of the treated spots, the amounts retained by the leaf tissue were equivalent to increases in Zn content of 35 and 72 $\mu\text{g}\cdot\text{g}^{-1}$ for ZnSO_4 and Zn metalosate, respectively, when the fertilizers were applied to the upper leaf surface. These values are within the same range observed for foliar fertilizer-treated trees in the field experiments. There was no statistically significant difference between ZnSO_4 and Zn metalosate when applied to the upper leaf surface. However, retention of ZnSO_4 and Zn metalosate was significantly increased (6- and 24-fold, respectively) when applied to the abaxial leaf surface compared to the amounts retained when applied to the adaxial surface (Table 5).

Of the small amounts of radiolabel retained by the leaves treated with $^{65}\text{ZnSO}_4$ on the adaxial leaf surfaces, about half was translocated into the adjacent leaf parenchyma tissue, and 5% to 8% was translocated into the leaf apex and leaves above and below the treated leaf. Although much higher levels of Zn were associated with the Zn application spots on the abaxial sides of the leaves, there were no differences in the actual quantities translocated to the other leaves or adjacent parenchyma tissue. Similar trends were observed with Zn metalosate, but much smaller percentages of the total Zn retained by the plant tissue were translocated out of the application spots. With Zn metalosate, only 2% to 4% of the total Zn retained was located in the apex tissue or leaves above and below the treated leaves. These data confirm that there is relatively little translocation of Zn from leaves treated with a foliar spray and suggests over the short term that there is no significant difference in the efficacy of Zn metalosate vs. ZnSO_4 for treatment of Zn deficiency in avocado.

Although foliar feeding is used to supply trace metals to a variety of ornamentals and tree fruit crops, different plant species vary with respect to their cuticle thickness and physiological characteristics that may influence the absorption and translocation of fertilizers. In trees, such as citrus, which have been studied much more extensively than avocado, absorption of trace metals also has been influenced by the formulation of the spray mixture and the type of surfactant (Alva et al., 1992). Paradoxically, neither avocado (Kadman and Lahav, 1978) nor citrus (Embleton et al., 1965; Labanauskas and Puffer, 1964; Swietlik and LaDuke, 1991) have responded to Zn applications with respect to yield increases. This research confirmed the poor mobility of foliar-applied Zn, whether it is applied as an inorganic salt or synthetic chelate, and is in agreement with earlier research on avocado and citrus (Ferrandon and Chamel, 1989; Kadman and Lahav, 1978). Thus, foliar applications are primarily useful for correcting Zn

deficiency in the treated foliage, and their efficacy cannot be evaluated by monitoring changes in adjacent nontreated leaves or by increases in fruit yield. Under field conditions, the amount of liquid spray applied to the leaves during foliar applications, and the time it will remain soluble before drying on the leaf surface, will depend on the application method and a variety of factors, such as location within the canopy, wind drift, air temperature, and humidity. As shown in our research, there also may be differential absorption of the materials by the upper and lower leaf surfaces, which have different cuticle thickness and stomatal densities.

Conclusion

This research revealed that there are critical problems in evaluating the efficacy of foliar sprays due to potential artifacts associated with the retention of extracellular Zn precipitates on avocado leaves. Results of experiments with radioisotope-labeled fertilizers suggested that, in some cases, foliar fertilization may supply small amounts of Zn. The efficacy of the Zn application will depend on the percentage of the tree leaf area that receives the Zn spray. If all leaves receive Zn spray, then the 5% translocated Zn may be adequate to supply new growth and buds with Zn. Some calculations based on leaf area, total Zn applied, total translocatable Zn, and dry-matter production over 1 year might give some clues as to potential efficacy. However, given the uncertainty as to whether foliar-applied Zn fertilizers can satisfy the Zn requirement of the entire tree and the expense of helicopter applications of Zn fertilizers, soil application of Zn appears to be a more reliable and simpler fertilization method. Zinc fertilization may be accomplished by using ZnSO_4 in a band applied under the irrigated sprinkler zone of the canopies of Zn-deficient trees or by adding solubilized ZnSO_4 to the irrigation water.

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