Mechanical transmission of the Texas ringspot was not reported, and it first appeared that the mechanical transmissibility of CRSV-SR might be a significant difference between these viruses. However, subsequent tests by L. W. Timmer in Texas (personal communication), plus results of a comparative host-range study now in progress in the Division of Plant Industry's quarantine greenhouse at Gainesville (Garnsey, Timmer and McRitchie, unpublished results), have shown that CRSV-NS is also mechanically transmissible to herbaceous plants and produces symptoms generally similar to those described here for CRSV-SR.

The symptoms produced by CRSV-SR are also generally similar to those produced by a ringspot virus previously isolated from a 'Zatima' navel orange tree in Florida (2) and an undescribed mechanically transmitted virus from commercial navel orange trees with severe psorosis symptoms (Garnsey, unpublished). Minor differences in symptom expression appear to exist in 1 or more hosts among all 4 of these virus isolates, but these results are inconclusive, and other properties of these isolates will have to be compared.

Regardless of its relationship to other ringspot virus isolates, CRSV-SR can cause serious effects in citrus plants and fruit and is a hazard to Florida citrus. The degree of danger from this virus depends on the extent of its spread. Apparent natural spread has occurred in Texas (5) by unknown means. Natural spread has not yet been observed in Florida, but its occurrence may not become apparent for awhile. The chances for natural spread increase with the number of infected trees that can serve as inoculum sources. Budwood was cut from the CRSV-SR-infected 'Star Ruby' in Florida before its destruction in June, and spread of CRSV-SR by propagation has apparently occurred. Detection of infected trees is difficult, because symptoms are erratic and irregular distribution of the virus within infected plants hampers indexing.

We strongly urge all nurserymen and growers to avoid propagation of any source of 'Star Ruby' grapefruit introduced without permit. Further spread of CRSV-SR into Florida citrus groves will occur if this propagation continues, and it will increase the hazard already created.

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# REMOVAL OF SURFACE IRON DEPOSITS FROM CITRUS LEAVES

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Additional index words. spray residues.

Abstract. Several Fe formulations are now being used to increase the availability of Fe or to correct Fe deficiency in citrus. Washing citrus leaves with different washing procedures, immediately following foliar application of Fe, did not remove or leach Fe. Methods for sampling leaves receiving foliar sprays of Fe are discussed. It was concluded that, under Florida conditions, rainfall 3 hr following foliar application of Fe would not totally leach this element from citrus leaves.

In the past 4 decades, the increased availability of soluble fertilizer formulations and engineering advancement in spray equipment have increased the use of foliar sprays as means of plant nutrition. Foliar sprays are now commonly used to furnish fruit trees with most of the micro- and some of the macro-nutrients which can not be supplied effectively through soil applications. Absorption of foliarly-applied minerals takes place through a chain of events beginning with the entry of an externally applied substance into the leaf and continuing with its movement within and, in some cases, transport from the leaf. Early studies dealing with the time course of ion uptake into plant tissues showed that an initial, relatively brief, period of uptake is normally followed by a slower but more prolonged period of absorption (3, 4, 5). The initial rapid phase occurs as a consequence of the free diffusion of ions into the apparent free space (AFS) of the plant tissue (4).

Some investigators (11, 12) have reported considerable losses of some nutrient elements from foliage by washing. Losses of nutrients from mineral deficient plants were greater than losses from non-deficient or healthy ones (8).

Complete removal of leaf surface contamination of foliarly-applied Fe is of utmost importance for precise quantitative measurements of the tissue level of this element. Inadequately cleaned or unwashed leaves were found to contain 17 times more Fe than carefully washed ones (6). The present study was undertaken to test the relative effectiveness of different washing procedures in removing Fe from citrus leaves following a foliar application of Fe, and also to determine the time course of Fe penetration into citrus leaves.

#### Materials and Methods

Leaves for this study were obtained from 20-year-old trees of 'Pineapple' orange (*Citrus sinensis* (L.) Osbeck) growing on a calcareous soil (pH 8.0) of the Florida East Coast. The trees were showing mild to moderate symptoms of Fe deficiency. A number of attached twigs from comparable growth were selected and dipped for 1 min in Fe formulation containing 60 g Rayplex (a lignin-free poly-flavonoid type chelate, Rayonier Incorporated, New York), 20 g MnSO<sub>4</sub>, 75 g urea and 15 ml Vatsol-OT (surfactant) per 3.785 liters. Twigs were harvested after 1, 2, or 3 hr and 3 replications of 30 leaves each of comparable samples were washed with 5 different washing procedures. Leaves were

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soaked and agitated for 3 min in the washing solution, after which each sample was thoroughly rinsed 3 times in tap water and in 3 separate distilled water baths for 5 min.

In order to determine the time course of Fe penetration into citrus leaves, 18 uniform, 5-month-old sweet orange seedlings were used. The seedlings were planted in waterwashed quartz sand in a greenhouse. After mild symptoms of Fe deficiency had been induced, the foliage was dipped in the above Fe formulation for 1 min. Leaves were sampled 6, 12, 24, 48 and 96 hr following Fe treatment. The leaves were washed by agitation in 0.3% solution (w/v) Dreft, a commercial detergent containing sodium lauryl sulfate, followed by scrubbing both leaf surfaces with cheesecloth. Samples were then prepared for analysis with precaution to avoid contamination. Fe and chlorophyll determinations were made as described in a previous report (2).

### Results

Effects of washing on Fe content and total chlorophyll of citrus leaves. Except for the acid wash procedure, there was significant uptake of Fe by citrus leaves, particularly 3 hr after treatment, as indicated by general increase in leaf Fe content (Table 1). The different washing procedures resulted in the removal of various amounts of leaf surface Fe which reflected the different degrees of effectiveness of these procedures. Agitating citrus leaves in 0.3% Dreft solution, followed by scrubbing both leaf surfaces with cheesecloth, seemed to remove a larger proportion of surface Fe than any other procedure. Washing leaves with 1 M NaEDTA (sodium salt of ethylenediaminetetraacetic acid) induced visible damage and was ineffective in removing surface Fe from citrus leaves. Leaves washed with NaEDTA showed significantly higher Fe levels than any other washing. It was possible that EDTA chelated the Fe on the leaf surfaces, and passage of Fe complex through leaf cuticles and cell membranes was enhanced with the urea in the dipping formulation. It has been reported earlier that wiping leaves with damp cheesecloth, washing in 3% HCl or in Alconox (a laboratory detergent) were satisfactory methods in removing Fe deposits from leaves of different fruit crops (9, 10, 11). However, the data indicated that none of these methods were entirely effective in this respect, and complete removal of surface Fe from citrus leaves by these methods was not obtained.

None of the washing procedures had any effect on the total leaf chlorophyll (Table 1). The apparent variations in chlorophyll content among samples of similarly chlorotic leaves which received different washing procedures, were mainly due to the randomization method used in securing comparable leaf discs for chlorophyll extraction. Contrary to previous report (9), no visual evidence of regreening of citrus leaves was observed even 3 hr after treatment with the Fe formulation.

Time course of Fe penetration into citrus leaves. Citrus leaves progressively absorbed various amounts of Fe following treatment (Fig. 1). There was 57% initial increase in Fe uptake 6 hr after treatment and washing with Dreft solution, followed by 65, 82, 87 and 90% increases after 12, 24, 48 and 96 hr respectively. This demonstrated that absorption of Fe in citrus leaf tissue did occur and proceeded steadily over 96 hr period. However, accumulation of Fe was significant only in the first 24 hr after treatment (Fig. 1, Table 1), while the difference between leaves sampled after 48 and 96 hr was insignificant. This suggested that, under the conditions of this study, there was no further uptake of Fe 48 hr following foliar application, while Fe surface residue was observed on treated leaves after the termination of the experiment. It is important to mention that slight regreening of some chlorotic leaves was observed 96 hr



Fig. 1. Time course of Fe penetration into citrus leaves.

Table 1. Effects of different washing procedures on Fe content and total chlorophyll of citrus leaves.<sup>z</sup>

Treatment	Fe content (ppm) <sup>y</sup>			Total chlorophyll (mg/g fr. wt.) <sup>y</sup>		
	1	2	3	1	2	3
Wiping with damp cheesecloth 0.3 % Dreft solution Alconox solution 3% HCl solution 1 M NaEDTA	70.1 a 52.3 b 64.4 c 62.2 c 76.6 d	73.4 a 55.1 b 65.1 c 65.7 c 79.5 d	75.2 a 57.4 b 68.9 c 60.1 d 81.1 a	0.473 0.452 0.456 0.461 0.447	0.452 0.469 0.448 0.432 0.478	0.456 0.444 0.459 0.453 0.455
Green leaves <sup>x</sup> Chlorotic leaves <sup>x</sup> Chlorotic leaves (Treated & unwashed)	115.3 042.1 196.9					

\*Values in column followed by the same letter are not significantly different at 5% level as determined by Duncan's multiple range test. Each value is the mean of 3 replications.

Samples collected 1, 2 and 3 hr after treatment.

following treatment with Fe. This further indicated that a portion of the foliarly-applied Fe was taken up by citrus leaves.

## Discussion

The data presented in this report showed that regardless of the cleaning method, washing following Fe application considerably reduced surface Fe from citrus leaves. Leaves sampled 1 hr after dipping in Fe formulation contained less Fe than leaves sampled at longer intervals. This indicated that Fe was accumulated in the AFS of citrus leaves and was removed by washing; therefore, it was 'physiologically inactive'. On the other hand, both cuticles, covering the outer walls of the epidermal cells, and ectodesmata, microchannels on the epidermal cells of citrus leaves, contain hydrophilic COOH groups. These groups have a tendency to chelate Fe before it crosses cell membranes to the cytoplasm. This portion of Fe, although it is physically bound to the cell organelles and will eventually reach the metabolic sites, is considered "physiologically inactive" and contributes substantially to the increase in Fe content of the leaves. Higher levels of Fe were accumulated in cirus leaves 3 hr following treatment with Fe formulation. Fe accumulation proceeded at a steady rate over a 96 hr period. It was assumed that a larger portion of this accumulated Fe might have moved symplastically and been taken up by the cytoplasm in an active uptake process requiring metabolically derived energy. However, it was difficult to conclude that the increase in Fe content at longer intervals was totally due to a 'physiologically active' Fe within the cell cytoplasm. Additional work is needed in this area.

Washing leaves of different crops with Dreft solution has been tested by many investigators (1, 7, 9). Despite the fact that this procedure removed more Fe from citrus leaves than the others, there was 24% more Fe in the chlorotic leaves washed with Dreft solution than untreated, but washed, chlorotic ones. Undoubtedly, a portion of this Fe was a surface deposit, yet it was not leached out from the leaves by washing. This could be interpreted to mean that rainfall or sprinkler irrigation, following Fe spray of citrus trees, would not have an appreciable effect on leaching out or totally removing Fe from citrus leaves. On the other hand, citrus leaves previously sprayed with Fe should not be recommended for analysis for diagnostic purposes. Residues from foliarly-sprayed Fe may remain for many weeks; hence, it is appropriae that the new leaves may be sampled since they reflect the status of Fe in the sprayed ones. Also, control leaves could be obtained on the same foliarly-sprayed branch or tree by covering several comparable, uniform twigs showing symptoms of Fe deficiency with plastic bags before spraying with Fe formulation and removing these bags shortly after spraying.

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# SURFACE TEMPERATURE VARIATION WITH RESPECT TO ELEVATION UNDER FREEZING CONDITIONS

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Abstract. Citrus growers have long observed large variations in temperature with respect to elevation on cold calm nights. The term "cold pockets" or "cold holes" is used to describe an area that traps cold air. These "pockets" can be considerably colder on calm nights than the surrounding area. A study was made using thermal film, topographical maps, and field inspection to demonstrate the effect of air drainage on surface temperature. Temperature data were collected by NASA personnel using a thermal scanner at an altitude of 7000 ft. A number of sites located by the thermal film study were visited and one was selected for a detailed study, where relationships among elevation, air drainage, and temperature were plotted. Cold pockets, which did not have adequate air drainage, were at least 10°F cooler than surrounding locations at the same elevation on calm nights.

Citrus growers continually cope with cultural problems, most of which can be controlled fairly successfully. Cold weather is the problem they find most difficult to deal with. Some growers continue to try protecting their groves with heat but they are becoming fewer in number as the price of fuel increases, burning controls become more stringent, and workers become harder to find. Various surveys indicate that over 11% of the groves were protected by heaters in 1971 but this acreage was reduced to less than 8% (2) by 1975.

Growers have long known that the best type of cold protection was to plant on "warm land" (3). Lawrence (3) states "the first consideration [when selecting a grove site] is topography. Rolling lands with gentle slopes and no depressions are preferred because of air drainage". Depressed