SPECIFIC EFFECTS OF ZINC APPLICATIONS ON LEAVES AND TWIGS OF ORANGE TREES AFFECTED WITH MOTTLE-LEAF[1]

By H. S. REED, *plant physiologist*, and E. R. PARKER, *assistant horticulturist*, *California Agricultural Experiment Station*

INTRODUCTION

This paper presents data on the effects of zinc salts on the growth of leaves and twigs of orange trees affected with mottle-leaf.[2] Trees in an orchard near Riverside, Calif., were selected for observation. They were about 30 years old at the time of treatment and had been severely affected with mottle-leaf for many years. Although the trees had produced a certain amount of new growth each year, the leaves were mottled and dwarfed. Many of the shoots had died back, and the quantity of fruit produced was subnormal.

In March 1934, a short time before the appearance of the first cycle of growth, the trees were sprayed with a mixture containing 10 pounds of zinc sulphate (commercial) and 5 pounds of hydrated lime, in 100 gallons of water. A few weeks after the application of the spray material the trees began to show improvement in the amount and character of the spring growth. This improvement has been maintained during the two following seasons, while untreated trees in adjacent rows have remained unthrifty and have borne typically mottled foliage. Other trees were similarly sprayed in October 1934 and showed normal growth the following spring.

The old, dwarfed leaves on the treated trees did not grow to normal size or shape after treatment, but they produced chloroplasts and assumed the green color of normal leaves. Apparently the leaf cells had lost their meristematic character insofar as multiplication was concerned, but the mitochondria proceeded to develop into plastids which synthesized chlorophyll.[3]

On treated trees it was possible to find in May 1935 many representative twigs bearing the small, elliptic-lanceolate leaves (fig. 1) characteristic of unsprayed mottled trees and also bearing normal green leaves of the 1934 cycles of growth. No such contrasts were observable on the untreated trees. The marked transition from dwarfed to normal leaves was coincident with the production of the first cycle of growth after the application of the spray mixture containing zinc sulphate and lime. This phenomenon indicates that zinc was quickly absorbed by the tree and promptly affected the metabolism of the organs which suffered from the systemic disease mentioned. The application of zinc sulphate to the soil has been

[1] Received for publication Mar. 26, 1936; issued October 1936. Paper no. 350, University of California Citrus Experiment Station and Graduate School of Tropical Agriculture, Riverside, Calif.

For detailed descriptions of the symptoms of affected trees, see the following: FAWCETT, H. S., and LEE, H. A. CITRUS DISEASES AND THEIR CONTROL. 582 pp., illus. New York and London. 1926.

[3] REED, H. S., and DUFRÉNOY, J. THE EFFECTS OF ZINC AND IRON SALTS ON THE CELL STRUCTURE OF MOTTLED ORANGE LEAVES. Hilgardia 9: 113–141, illus. 1935.

followed by similar improvements in growth, though the time required for expression has been longer, and injury has resulted in some cases.[4]

OBSERVATIONS ON TWIGS AND LEAVES

The twigs shown in figure 1 were photographed in May 1935. The differences between the leaves produced before and after the

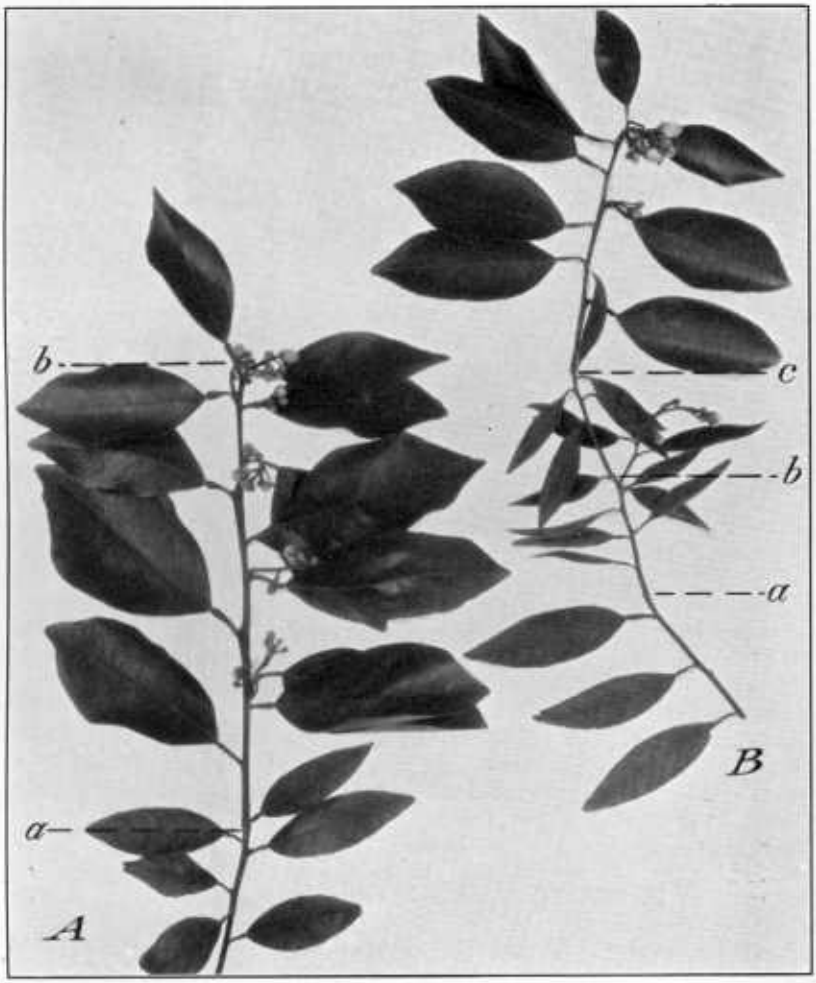

FIGURE 1.—Valencia orange twigs showing the effects of zinc-lime spray on trees affected with mottle-leaf: *A*, Cycle of growth below *a* produced small leaves before the tree was sprayed in October 1934; the cycle *a-b* was produced in the autumn of 1934 and bore large leaves; *B*, *a-b* and *b-c* show the cycles of growth and the small leaves borne before the tree was sprayed; the cycle above *c*, bearing large leaves, was produced after the tree was sprayed in May 1934. Both samples were collected in May 1935.

zinc sulphate-lime mixture was applied are striking. The shoot *a-b-c* in figure 1, *B*, was cut from a tree which had been sprayed in May 1934. During the remainder of the summer it produced one

[4] PARKER, E. R. EXPERIMENTS ON THE TREATMENT OF MOTTLE-LEAF OF CITRUS TREES. Amer. Soc. Hort. Sci. Proc. 31 (sup. vol.): 98-107. 1934.

cycle of growth, starting at the point *c;* this cycle was longer than earlier ones and bore healthy leaves. The greater length of the internodes of the healthy cycle is characteristic of normal growth. The other shoot shown in figure 1, *A*, produced one cycle of growth, *a-b*, after it was sprayed with the zinc sulphate-lime mixture in October 1934. It also bore healthy, oval leaves, which were much larger than those of the preceding cycle. The healthy twigs produced flowers at many of the nodes, in contrast to the few flowers on twigs of the preceding cycles.

The average area of leaves from affected trees gives a more precise indication of the effects of the spray solution containing zinc sulphate. Data were obtained from leaves collected April 15, 1935, from trees which had been sprayed in May 1934, and from comparable unsprayed trees. Each sample consisted of 100 mature leaves selected from the four sides of the trees. The outlines of the leaves were traced on paper and measured with a planimeter. The mean areas of the small leaves on the cycle of growth immediately preceding the treatments were very similar, 0.74 ± 0.01 and 0.75 ± 0.01 square inch for the untreated and for the sprayed trees, respectively. The leaves produced on the late summer or fall cycle of growth were larger on both the control and the treated trees, but the increase in size on the sprayed trees was striking, the figures being 1.61 ± 0.02 and 3.67 ± 0.02 square inches, respectively. The statistical reliability of the means is evident by comparison with their probable errors.

The width of phloem and xylem tissues in twigs may also serve as an index of their growth and hence express the effect of zinc sulphate on cellular activity, at least in respect to the cambium.

On May 7, 1935, twigs were selected from sprayed and unsprayed trees. As in the collection of the leaves, care was taken to obtain representative samples. Only twigs of an ascertainable age were chosen. These were of three ages: (1) Twigs of the 1935 spring cycle; that is, new growth; (2) twigs of the 1934 spring cycle, which had grown approximately 14 months; and (3) twigs of the 1933 spring cycle, which had grown approximately 26 months.

Twenty to thirty twigs were cut to convenient lengths and placed in a 1 : 1 mixture of water and glycerin, where they remained for several weeks. Transverse sections were then cut on a sliding microtome, stained with safranine, washed, and mounted on slides for measurement.

TABLE 1.—*Effect of zinc sulphate-lime spray on the average width of phloem and xylem tissues of orange twigs*

Designations of the twigs (see text)	Average width (microns) of—			
	Phloem in—		Xylem in—	
	Unsprayed twigs	Twigs sprayed May 1934	Unsprayed twigs	Twigs sprayed May 1934
1, 1935 spring cycle (March to May 1935)	127.8±2.3	113.8±2.0	119.6± 5.7	124.3± 3.8
2, 1934 spring cycle (March 1934 to May 1935)	199.7±5.4	187.3±4.9	381.3±14.5	518.5±14.8
3. 1933 spring cycle (March 1933 to May 1935)	249.0±8.0	218.9±5.0	630.8±18.1	716.4±17.3

The data in table 1 show that there was no very significant difference in the thickness of the phloem cylinder in twigs from unsprayed and sprayed trees. The phloem of the older twigs was thicker than that of the younger ones. However, the xylem cylinder was distinctly thicker in the twigs from sprayed than from unsprayed trees. The averages for the young twigs showed no significant difference, but in the case of the two lots of older twigs the differences are statistically significant.

The 26-month-old twigs were nearly a year old before zinc sulphate was applied to the trees on which they grew. Hence, it is not surprising that the difference in size between sprayed and unsprayed twigs in this lot was somewhat less than in the 14-month-old lot.

These measurements may be interpreted to mean that the improvement in general growth activities already described extended to the activity of the cambium and resulted in the production of more wood in these twigs. Although too few data are available to permit a discussion of the specific action of the zinc on the cambium cells, it may be pointed out that the increased cambial activity would logically follow the great increase in chlorophyll-bearing area previously described and the general improvement in the condition of the trees elsewhere reported.[5]

SUMMARY

Orange trees affected with mottle-leaf produced healthy foliage and increased growth after they had been sprayed with a zinc-lime mixture. The specific effects observed were: (1) Larger leaves, (2) longer internodes, and (3) more xylem tissue.

[5] PARKER, E. R. See footnote 4.

U. S. GOVERNMENT PRINTING OFFICE: 1936