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ZINC ACCUMULATION IN THE WOOD OF CITRUS TREES AFFECTED WITH BLIGHT

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Abstract. In Florida, blight is primarily a wilt disease of citrus with an unknown cause. Zinc deficiency symptoms are frequently found in the foliage of trees affected with blight. Accumulation of Zn in the wood of the trunk, large limbs, and roots indicates selective absorption from the transport stream. Zn concentrations in the outer layers of wood were about 8 times higher than in healthy trees. There was a slight copper accumulation also, but 9 other minerals were not affected. Interference in Zn transport is assumed to be a result of the affliction and not the cause, since foliar applications of the mineral cured the deficiency but did not alleviate the blight.

The term "Blight" has long been applied to a specific chronic wilt and decline of citrus trees in Florida (1, 3, 7, 8, 9). The cause is still unknown. Various names have been applied to describe this, or similar, diseases. Two of the most common are "young-tree decline" and "rough-lemon decline". Blight has precedence in usage and is therefore used here.

One of the earliest symptoms is induced zinc (Zn) deficiency. Patterns of Zn deficiency often show up in the foliage of one sector of the tree before or at the same time as the symptoms of wilt. The symptoms of Zn deficiency and of drought

spread radially until the entire canopy is affected. Foliar sprays of Zn cause the chlorosis to disappear, thereby often masking a relation between the deficiency and the disease.

In 1966, blight started on 9-year-old orange trees in one of my Zn fertilization experiments (5, 6). These trees had been specifically propagated for the experiment from nucellar 'Valencia' (*Citrus sinensis* (L.) Osbeck) budwood and planted on virgin soil in Lake County. The experiment had been underway for several years and there was detailed knowledge available about the status of Zn and other minerals from frequent leaf analysis.

Unexpected patterns of Zn deficiency on a few scattered trees attracted my attention in the summer of 1966. In the spring of 1967, symptoms of wilt and delayed flush were obvious. The chlorosis symptoms disappeared rapidly following a Zn spray, which left no doubt as to the diagnosis of Zn deficiency on the declined trees. In the 8 years between 1966 and 1974, about 15% of 800 trees on rough lemon (*C. limon* (L.) Burm. f.) rootstock became blighted. However, none of over 200 trees on 'Cleopatra' mandarin (*C. reticulata* Blanco) rootstock were affected.

The incidence of decline was not related to the soil Zn status. High and low rates of Zn had previously been incorporated in the soil in many plots of 12 trees each. Once a tree started to decline, chronic Zn deficiency symptoms developed on all subsequent flushes of growth. Application of several Zn sprays per year for 6 years prevented foliar Zn deficiency symptoms, but the declined trees did not recover.

The present study was made to find out why Zn deficiency develops on trees affected with blight.

Experimental Procedure

Because Zn evidently was either not being absorbed or not being conducted through the tree, the internal distribution of Zn was compared in healthy and declined trees. Trees in an early stage of decline were selected, because they still had an extensive and apparently healthy system of feeder roots. Most of the samples taken for analysis were wood with bark removed. Leaves and rootlets were washed in detergent, but the wood samples were unwashed. All samples were dried at 65° C and ground to a powder for mineral analysis. Most of the samples were analyzed spectrographically for P, K, Ca, Mg, Cu, Zn, Mn, Fe, B, Al, and Na. The others were analyzed for Cu, Zn, and Mn by atomic absorption.

Vertical distribution of Zn. A declined and an adjoining healthy tree, in the experimental block of nucellar 'Valencia' orange trees, were selected for this comparison. Both trees were 15 years old when sampled in April 1972. From each tree, 100 leaves of the previous year's growth were picked at random, and 25 small branches were cut. Sec-

tions about 2 inches long were cut from these branches to provide wood samples that were 1/8 and 1/4 inches in diameter. The bark was peeled off and discarded. Four branches were sawed from each tree to provide 7/8-inch diameter wood. Cores of wood were taken by auger from the larger branches, trunk, and large roots. For small and fibrous roots, 8 cores at the drip line of the canopy of each tree were taken with a 3-inch aluminum soil tube to a depth of 6 inches.

Radial distribution of Zn in wood. Sections about 1/2-inch thick were cut from the above-ground woody parts of another blighted tree with a chain saw. Discs were taken from the trunk and from limbs 1 to 5 inches in diameter. After the bark was peeled off, each disc was cut into concentric rings with a band saw. Two outer rings were cut at 1/4 inch in radial thickness on all sections. Other rings were cut 1/2-inch wide, until the center of the disc was reached. Each ring of wood was analyzed separately.

Zn distribution in healthy vs. declined tree trunks. Sawed sections from trunks or large limbs were prepared, as just described, for 16 trees. Eight of these showed severe symptoms of blight, and 8 appeared healthy. Because the trees were sacrificed in the process of getting trunk samples,

Table 1. A comparison of the vertical distribution of Zn in a healthy and a declined nucellar Valencia orange tree on rough lemon stock.²

Portion of tree	ppm Zn in dry matter		Percent diff.
	Healthy	Declined	
Leaf blades	33	13	-61
Petioles	14	9	-36
Twig wood (1/8 in.)	7	8	+14
Twig wood (1/4 in.)	4	5	+20
Branch wood (7/8 in.)	3	4	+25
Limb wood (3 in.)	4	12	+200
Trunk wood (12 in.)	4	19	+375
Root wood (3 in.)	5	18	+260
Root wood (5/8 in.)	6	16	+167
Root wood (1/4 in.)	10	22	+120
Fibrous roots	760	738	-3

²Adjoining trees were sampled April 6, 1972. All wood samples had bark removed.

the healthy trees selected were ones that were being destroyed in land-clearing operations. Thus, random ages and varieties were involved.

Zn staining of wood sections. Thin tangential sections were cut freehand from 1/4-inch roots of blighted trees. These were placed on a microscope slide, stained with a drop of an alcoholic solution of a-pyridylazol b-naphthol, and observed immediately under 100X magnification. This dye changes from yellow to orange-red in the presence of Zn (4).

Results

Of the 11 mineral elements, Zn was the only element that showed an enormous difference in relation to blight.

Vertical distribution of Zn. Table 1 shows the Zn concentrations found in various parts of healthy and diseased trees. In both cases, fibrous roots contained similar but high amounts of Zn. In declined trees, the woody parts showed a higher and leaves and petioles showed a lower Zn content than in healthy trees. In declined trees, the greatest accumulation of Zn was in the trunk and root wood.

Radial distribution of Zn in wood. The data in Table 2 show that Zn accumulated most in the outer inch of wood in the trunk and main limbs. The deeper wood, inside about 1-1/2 inches, contained Zn in a near-normal range. Small limb wood showed little or no gradient in Zn concentration.

Zn in healthy vs. declined tree trunks. The data

in Table 3 show that the Zn concentration in the large wood of healthy trees is low and there is virtually no gradient from outer to inner wood. However, each declined tree had a relatively high Zn concentration in the outer layers of the wood and a rather sharp gradient in concentration across the outer 2 inches of trunk wood. In the outer 1/2 inch of the trunk, Zn was about 8 times as high in declined trees as in healthy.

The mean values of Cu and Mn in the same 16 trees are shown in Table 4. Copper concentration was also somewhat higher in declined trees, but the difference was not nearly so great as with Zn. Manganese concentrations were alike in both diseased and healthy trees.

Zn staining of wood sections. Many plugs were seen in the vessels of the wood. These stained a deep orange-red color, thus indicating that Zn had accumulated in the material that formed the plugs. Staining also took place in the border pits of some vessels, indicating that a Zn-containing substance was located there. Elsewhere there was little or no staining.

Discussion

These results overwhelmingly show that there is an interference in the translocation of Zn through trees affected with blight. Failure of the roots to absorb Zn from the soil does not appear to be a factor.

The pattern of Zn distribution indicates ab-

Table 2. Radial distribution of Zn (ppm in dry matter) in cross sections of wood from aboveground parts of a declined Valencia orange/rough lemon tree.²

Diameter and tree part	Distance from cambium toward core (in.)						
	0-.25	.25-.5	.5-1	1-1.5	1.5-2	2-2.5	2.5-3
1-in. limb	3	4					
2-in. limb	5	5	5				
2.5-in. limb	11	8	7	3			
4-in. limb	16	13	8	5	3		
5-in. limb	27	18	16	10	5	3	
8-in. trunk	29	11	5	2	2	2	2
10-in. union	31	30	14	6	3	2	2

²Wood cross-sections approximately 0.5 in. thick were cut into concentric samples. The two outermost samples were 1/4 in. in radial thickness; the others, 1/2 in. Not all of the inner wood samples are shown for the trunk samples, because they had levelled off.

Table 3. A comparison of the Zn accumulation in the trunks or large limbs of healthy and declined citrus trees.^z

Citrus variety/stock	Tree age yr	Trunk or limb	ppm Zn in dried wood				
			Distance inward from cambium (inches)				
			0-.25	.25-.50	.50-1.0	1.0-1.5	1.5-2.0
Healthy trees							
Valencia/RL	15	L	3	3	3	4	3
Valencia/RL	15	T	4	3	2	3	2
Valencia/RL	10	T	6	2	2	2	3
Pineapple/Tri	7	T	2	3	1	2	-
Pineapple/Tri	7	T	2	1	1	1	-
Orlando/RL	7	T	2	1	2	1	1
Orlando/RL	7	T	1	1	2	1	1
Grapefruit/RL	10	L	5	2	3	3	2
Mean Zn concentration			3.1	2.0	2.0	2.1	2.0
Declined trees							
Valencia/RL	15	T	30	14	7	6	4
Valencia/RL	15	T	23	13	11	9	5
Valencia/RL	15	T	17	16	17	12	5
Valencia/RL	15	T	36	20	15	9	5
Valencia/RL	15	T	18	26	7	4	2
Valencia/RL	15	T	31	30	14	6	3
Pineapple/RL	18	T	29	11	5	2	1
Queen/Sweet	12	T	14	7	6	2	2
Mean Zn concentration			24.8	17.1	10.2	6.2	3.4

^zSee Table 2 footnote concerning sample preparation. Scion varieties are all sweet orange except Orlando tangelo (*C. paradisi* Macf. X *C. reticulata* Blanco) and Marsh grapefruit (*C. paradisi*). Rootstocks listed other than rough lemon were *Poncirus trifoliata* (L.) Raf. (Tri) and sweet orange.

Table 4. Mean concentrations of Cu and Mn in the same wood samples shown in Table 3.

Tree condition	Distance inward from cambium (inches)				
	0-.25	.25-.50	.50-1.0	1.0-1.5	1.5-2.0
ppm Cu in dried wood					
Healthy	2.6	2.0	2.0	2.1	1.8
Declined	4.0	3.4	2.4	3.0	2.9
ppm Mn in dried wood					
Healthy	2.2	2.0	1.7	1.6	1.3
Declined	1.9	1.4	1.3	1.3	1.4

normal function in the large skeletal wood, both above and belowground. The gradients of Zn accumulation in the outer layers of trunk wood seem to show that absorption of Zn from the transpirational water stream started at the onset of decline, probably before visible symptoms could be seen.

The plugging of vessels in blighted trees was described by Childs in 1953 (1). The nature and cause of plugs is not yet fully understood. The present observations show that they have an affinity for Zn. Thus, deficiency symptoms develop in the leaves because Zn is strained out of the water stream. The failure of Zn to reach the foliage in adequate amounts can scarcely be considered a cause of the disease, because there has been no real benefit from supplying the foliage with spray Zn.

The selective absorption of Zn instead of some other heavy metal, is probably relevant to an explanation of the disease. Copper is generally bonded preferentially by proteins, lignins, phenols, and chelation materials. It would be helpful to know what the bonding material is that holds Zn while letting Cu, Mn, and Fe pass.

The primary physiological effect of blight is the desiccation that results from inadequate movement of water to the foliage. This would appear to indicate extensive blockage of vessels. Childs (1) suggested that plugged vessels might be the physical cause of blight. Cohen (2) verified the presence of xylem plugs, but thought they were not extensive enough to cause wilt. More information is needed about the occurrence of plugging in both healthy and blighted trees and on its cause. It would also be of interest to see if Zn accumulation is related to plug formation, or occurs afterward.

A complete explanation of the disease must account for the accumulation of Zn, even if it is only an incidental event.

The Zn values found in fibrous roots may seem high in comparison to those in other parts of the tree. However, the cortical tissues of both living and dead roots have a strong affinity for heavy metals (4).

A Zn determination of trunk wood might provide a method of pre-visual detection of trees that are destined to become blighted. Such prior knowledge would greatly help other studies on the disease. For instance, cytological examination and studies on chemotherapy could be concentrated on the incipient victims instead of being wasted on the high proportion of trees that do not become blighted in the immediate future. Studies are underway to see if such a method can detect blight in its formative stages.

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