

## Relationship of Age of Plants and Resistance to a Severe Isolate of the Beet Curly Top Virus

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### ABSTRACT

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Strains of the curly top virus capable of causing appreciable damage to resistant cultivars of sugar beet are found throughout the western United States. However, little knowledge of the extent of the damage induced by these isolates was known. Field trials on sugar beets have indicated that current strains of the curly top virus caused serious losses, even when inoculated as late as 10 wk after seeding or

after more than 40% of the growing period had elapsed. Both root yield and sucrose content were significantly reduced. Disease resistance appears to be associated with differences in incubation period rather than differences in the abilities of resistant cultivars to recover from the effects of virus infection.

Beet curly top virtually destroyed the sugar beet industry in the western United States before the introduction of resistant cultivars in 1934. The disease was the principal limiting factor to sugar beet production west of the Rocky Mountains from the early 1900's until World War II.

Seeding early in the growing season (5, 11, 12), the use of resistant cultivars (3, 4), and insecticide application (2, 6, 10) may reduce these extremely heavy losses to less than catastrophic proportions.

There is much evidence to indicate that beet curly top virus (BCTV) is a complex of strains that differ in virulence, symptoms induced, and host range (1). During the last several years, strains capable of causing marked damage on resistant cultivars of sugar beet have increased in number and distribution (1). However, the extent and the implications of the actual damage and yield losses due to infection with the more severe isolates on sugar beet cultivars previously have not been determined.

Field inoculation experiments with BCTV have been difficult to conduct and usually resulted in low infection rates in the inoculated plants and high contamination rates in the control plots, resulting in inaccurate estimates of the effects of BCTV on sugar beet yield. The most recent paper on this subject (7) implied that little damage occurs on resistant cultivars when the plants are infected 4-5 wk after planting.

The lack of information on the damage induced on resistant cultivars at different stages of plant development by the widespread virulent isolates of BCTV prompted this study and preliminary studies during 1970, 1973, and 1974 (Skoyen and Duffus, *unpublished*). The earlier studies established a reliable inoculation technique that resulted in high levels of curly top infection, while noninoculated plots remained relatively free of BCTV.

### MATERIALS AND METHODS

The sugar beet cultivars US 75 and US 15 were sown 30 April 1975, in a split-plot design with five replications at the U. S. Agricultural Research Station, Salinas, California. The main plots were cultivars and the sub-plots were five dates of inoculation, approximately 4, 6, 8, 10, and 12 wk after sowing. Dates of inoculation were randomized over each main plot. Sub-plots were four rows wide and 4 m long. Stands in each plot were reduced to 48 plants before the first inoculation. Cultivar US 15, which was introduced in 1938, was the first cultivar to meet the requirements for winter sowing (bolting and curly top resistance) and was used in California for about 10 yr (8). It has what would be considered today low curly top resistance. Cultivar US 75 has what was considered a high level of curly top resistance and good bolting resistance when it was introduced in 1952 (9).

A severe isolate (Logan) of BCTV collected from Utah and which is as virulent as any isolate previously tested in California, was used in the inoculation. Inoculations were made by attaching two small leaf cages containing three leafhoppers previously reared on virus-infected plants, to the youngest leaves of the plants. The cages, made from 25-mm diameter acrylic tubing 2 cm long, were covered at each end by nylon material and held tightly to the leaf surface with bent hair clips. Cages remained on the inoculated plants for 1 wk. Insect survival was over 90% during these inoculation periods.

The plants were examined at weekly intervals for curly top symptoms until harvest, 15 October 1975, when the plants were 24 wk old.

### RESULTS

**Percentage infection.**—Percentage of infection obtained at different intervals after seeding (Table 1) substantiated results obtained in 1970, 1973, and 1974

(Skoyen and Duffus, *unpublished*) in that the inoculation technique resulted in high levels of BCTV infection in the field. Infection percentages ranged from over 90% on the susceptible cultivar even after 8 wk from seeding to 77.1% after 12 wk. On the resistant cultivar, more than 95% infection was obtained even after 6 wk, but infection percentages dropped to 38% for plants inoculated 12 wk after seeding. These results indicate a significant increase in resistance to infection as the plants increased in size and/or age. There was no statistical difference in resistance to infection between the two cultivars for the inoculations on the 4th and 6th wk after planting, but all inoculations after that time resulted in significantly lower infection rates for the resistant cultivar. The control plots at harvest had 2% and 8% curly top infection for resistant and susceptible (respectively) cultivars. This contamination probably resulted from beet leafhoppers escaping during the inoculation procedure and, perhaps, a few naturally occurring leafhoppers.

**Yield.**—Yield observations and analysis of the field plot data in 1970, 1973, 1974, and 1975, and analysis of the 1975 yield data (Table 1) indicated that catastrophic losses could result when susceptible cultivars were inoculated with severe isolates of the BCTV as late as 8 wk after seeding. The resistant cultivar had yield losses of 47.3% even with inoculations as late as 6 wk after seeding.

It is important to note that significant yield losses (approximately 11%) resulted when the resistant cultivar was inoculated as late as 10 wk after seeding, when more than 40% of the growing period had elapsed.

**Sucrose.**—Beet curly top virus had a severe impact on the sugar content of infected sugar beets (Table 1). Sucrose losses in the susceptible cultivar ranged from approximately 55% at the earliest inoculation date to approximately 3% at 12 wk after seeding, when 50% of the growing period had elapsed. Sucrose losses in the resistant cultivar ranged from approximately 25% to about 3%.

**Gross sugar.**—Root yield and sugar percentage data indicate that both yield components played a significant role in the reductions caused by curly top. Early inoculations (through the 4th week from seeding) drastically reduced sugar production of both susceptible and resistant cultivars. Resistance to yield and sucrose losses increased rapidly in the resistant cultivar from the 6th wk after planting; however, inoculation at 10 wk still resulted in a significant reduction of over 13% in sugar yield (Table 1).

**Incubation period.**—A comparison of the incubation period of BCTV in susceptible and resistant sugar beet cultivars showed several significant differences (Table 2). Both the average minimum incubation period (the time in

TABLE 1. Effects of a severe isolate of the beet curly top virus on resistant and susceptible sugar beet cultivars inoculated at different intervals after seeding

Time of inoculation (weeks after seeding)	US 15 (susceptible)				US 75 (resistant)			
	Infection <sup>y</sup> (%)	Yield of beets/ha (metric tons)	Sucrose (%)	Gross sugar/ha (kg)	Infection (%)	Yield of beets/ha (metric tons)	Sucrose (%)	Gross sugar/ha (kg)
4	98.3 a <sup>z</sup>	0.16 a	6.71 a	11 a	95.4 a	3.27 a	11.42 a	377 a
6	99.7 a	11.34 b	10.78 b	1,222 b	96.5 a	35.99 b	13.35 b	4,818 b
8	94.3 a	34.04 c	12.91 c	4,415 c	77.2 b	58.31 c	14.45 c	8,440 c
10	84.6 b	52.49 d	14.08 d	7,386 d	72.1 b	60.90 c	14.90 cd	9,076 c
12	77.1 b	54.04 d	14.75 de	7,993 de	38.4 c	68.09 d	14.88 cd	10,135 d
No inoculation (control)	8.3 c	57.01 d	15.18 e	8,663 e	2.1 d	68.34 d	15.35 d	10,496 d

<sup>y</sup>Least significant difference,  $P = 0.05$ , between cultivars for percent infection at different times of inoculation = 8.03.

<sup>z</sup>Means within columns followed by the same letter are not significantly different from each other,  $P = 0.05$ , by Duncan's multiple range test.

TABLE 2. Incubation periods of a severe isolate of the beet curly top virus on resistant and susceptible sugar beet cultivars inoculated at different intervals after seeding

Time of inoculation (weeks after seeding)	US 15 (susceptible)		US 75 (resistant)	
	Minimum <sup>y</sup> incubation period (weeks)	Time for maximum % infection (weeks)	Minimum incubation period (weeks)	Time for maximum % infection (weeks)
4	2.0 a <sup>z</sup>	2.0 a	2.0 a	2.8 a
6	2.0 a	5.5 b	3.0 b	8.6 b
8	2.4 ab	7.4 c	4.6 c	11.1 c
10	3.2 bc	8.7 cd	4.3 c	10.7 c
12	3.6 c	9.3 d	5.9 d	9.5 bc

<sup>y</sup>Least significant difference,  $P = 0.05$ , between cultivars for minimum incubation period = 0.86, and for maximum percent infection = 1.65 at different times of inoculation.

<sup>z</sup>Means within columns followed by the same letter are not significantly different from each other,  $P = 0.05$ , by Duncan's multiple range test.

weeks for the plants at each inoculation date to show initial curly top symptoms) and the time in weeks for the plants at each inoculation date to show the maximum percentage infection were significantly different at the different stages of inoculation for both the susceptible and the resistant cultivars. Incubation periods tended to increase with successively later inoculation dates. The cultivars also differed significantly in incubation period. Both the average minimum incubation period and the average time for maximum percentage infection were longer for the resistant cultivar.

**Disease index.**—Regression analysis showed highly significant correlations between the incubation periods and root yield, sucrose percentage, and gross sugar. A highly significant negative correlation was found between the length of time the plants showed symptoms and the final yield of sucrose per hectare. A disease index,

computed by multiplying the percentage of plants showing symptoms by the number of weeks until harvest, was calculated for each replicate and each inoculation date for both cultivars. Regression coefficients of the loss of sucrose yields on the disease index are shown in Fig. 1. The coefficients indicate that for each increase of one unit of the disease index, there was a 7.75% decrease of sugar per hectare for US 15, and a 6.01% decrease in sugar per hectare for US 75. The regression coefficients are highly significant and the slopes are significantly different from each other.

DISCUSSION

The avoidance of infection during the early part of the growing season has long been recognized as an important factor in preventing excessive losses from BCTV in regions where the disease is prevalent. With the development of resistant cultivars, the prevention of early infection was assumed to be less urgent. During the last 20 yr, BCTV isolates have increased in severity to the point that isolates considered severe in the 1950's are now considered mild. The conclusions based on age-at-infection studies conducted during these early periods are no longer valid and could seriously mislead sugar beet growers and processors. The implication that little damage occurs on resistant cultivars when the plants are infected 4-5 wk after planting is no longer true for the new, more virulent isolates of BCTV. The ability of modern curly top-resistant cultivars to withstand infection and injury from curly top and to outgrow distinct evidence of injury, even when infected at a young stage of growth, as was reported in the 1940's, is not true for the curly top isolates of today. Although the curly top virus has been studied extensively since it was first reported in 1888, and has caused severe and extensive losses, little has been known about actual losses induced by the virus.

The, still not infrequent, destructive attacks of the curly top virus are probably not as important economically as the generally accepted curly top infections at later stages of plant development that have been assumed to cause little damage. The fact that infection of a resistant cultivar with severe BCTV isolates, even 10 weeks after seeding, can cause losses of over 13%, indicates the need for more effective control measures. Such losses were the result of only 72.1% of the plants showing symptoms. In areas of high leafhopper incidence, the percentage of infected plants undoubtedly could approach 100%.

Disease loss from infection at later stages in plant development probably has been underestimated or overlooked because of the lack of contrast with disease-free plants.

The lack of a satisfactory field inoculation technique has led to the assumption that although roots of diseased plants tend to be woody, sucrose percentages were unaffected (1). It was clearly evident in our preliminary experiments in 1970, 1973, and 1974 (Skoyen and Duffus, unpublished) and confirmed in the present work, that the current virulent isolates of the BCTV have a serious impact on the sugar content of both susceptible and resistant sugar beet cultivars.

Plotting the incubation periods resulting from the different inoculation dates (Fig. 2) showed clearly that

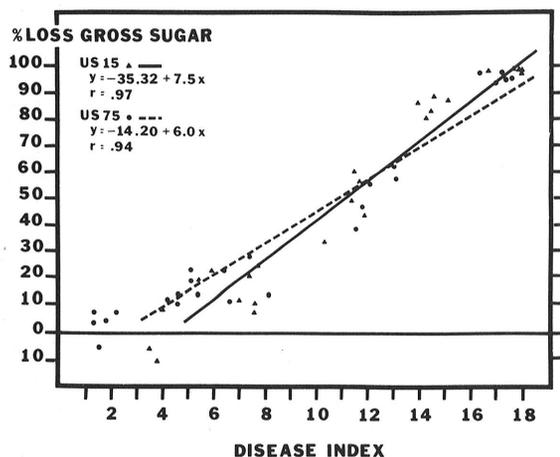


Fig. 1. Relationship between curly top disease index (based on percentage infected plants and the time the plants showed symptoms until harvest) and loss of gross sugar in susceptible (US 15) and resistant (US 75) sugar beet cultivars.

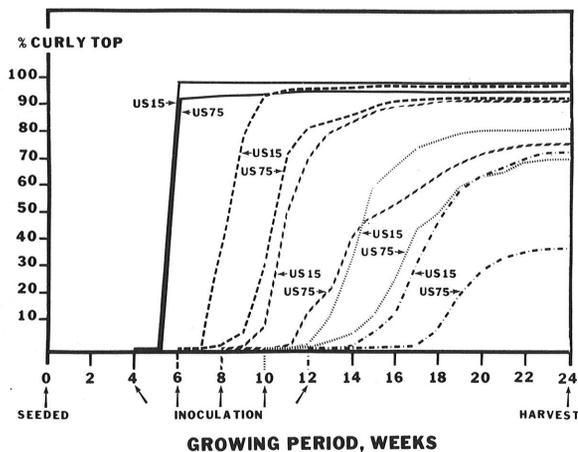


Fig. 2. Curly top development in sugar beet as expressed by percentage plants showing symptoms following five inoculation dates during the growing season.

increased incubation periods at the later inoculation dates were probably the most significant factor in disease resistance. A disease index, based on the percentage of infected plants and the time from when the plants showed symptoms until harvest, was highly correlated with the percentage loss in gross sugar yield. Interestingly, although the regressions for the susceptible and resistant cultivars were statistically different, they are remarkably similar for cultivars differing so greatly in resistance. There is little evidence that resistant sugar beet cultivars show any tendency toward recovery, as far as yield factors are concerned.

A reasonable estimate of sugar beet yield losses due to effects of severe isolates of BCTV possibly could be derived from regression coefficients for essentially the range of resistance now found in sugar beet cultivars.

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