

Temperature Effects on Curly Top Resistance in *Phaseolus vulgaris*

M. J. Silbernagel and A. M. Jafri

Research Plant Pathologists, respectively, ARS, USDA, Western Region, Irrigated Agriculture Research and Extension Center, Prosser, Washington 99350; present address of second author: Department of Plant Pathology, Washington State University, Pullman 99163.

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ABSTRACT

Greenhouse and growth-chamber studies indicate that resistance to curly top virus (CTV) in certain snap bean (*Phaseolus vulgaris*) cultivars may break down at high temp, but is not influenced by the stage of plant development at time of inoculation. The virus was not recovered from inoculated symptomless plants. Plants of the resistant variety in which resistance was broken

showed considerable tolerance, since symptoms do not become as severe as on a susceptible variety. In a susceptible variety, higher incubation-period temp increased % CTV incidence and symptom severity and reduced the number of days from inoculation to symptom expression.

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The curly top virus (CTV) disease of beans (*Phaseolus vulgaris* L.) is transmitted by the sugarbeet leafhopper *Circulifer tenellus* Baker. Two types of resistance to CTV have been described in beans: the tendency to escape infection by vector nonpreference (2, 6, 10, 14); and physiologic resistance, which depends on factors operative after introduction of the virus into the plant (2). Schultz and Dean (13) studied the inheritance of resistance to CTV in a number of bean hybrids and concluded resistance was based on two epistatic factors, one dominant and one recessive. Dana (5) and Murphy (11) summarized the sources of curly-top resistance in beans used in the respective USDA and University of Idaho bean-breeding programs.

Carter (3) showed that sugarbeets developed more severe curly top symptoms at high temperatures than at low temperatures and light intensities. Giddings (9) and later Schneider et al. (12) found that symptom severity in the sugarbeet decreased as the age of the inoculated plant increased. Similarly, Dana (5) reported that CTV-susceptible beans infected later in

the season showed milder symptoms.

Several CTV-resistant snap bean varieties have been developed by the USDA breeding program at Prosser, Washington. However, even among the most highly resistant of these varieties; Apollo, Goldcrop, and Yakima, 2-5% of the plants develop symptoms of curly top in the field during seasons when the disease is severe. The purpose of the investigations reported here was to determine whether high temperatures after inoculation, or stage of growth at inoculation, might account for this disease incidence in resistant varieties.

MATERIALS AND METHODS.—The curly top-resistant varieties Goldcrop and Yakima, used in these experiments, derived their resistance from Burtner's Blightless, a white pea-bean type used in the early 1930's by a grower in Wasco County, Oregon, after whom it was named (4). The varieties Bountiful and Tendercrop are known to be highly susceptible to CTV (14).

Bean plants were inoculated by means of leafhoppers carrying a mixture of wild strains of the

virus. Leafhoppers were reared uncaged in an insectary, on infected sugarbeets dug from fields near Prosser, Washington, or were collected from infected susceptible beets in the field.

Growth-chamber studies were conducted using 16, 136 lux for a 16-h photoperiod.

A disease-severity index (SI) was developed to quantitate (on a 0 to 100 scale) the host-population reaction to curly top. At a given time all plants are rated into six disease categories as follows: 0 – no symptoms; 1 – barely discernible symptoms; 2 – clearly positive symptoms, no serious damage; 3 – severe symptoms and plant damage; 4 – severe symptoms and damage, plus tip kill; 5 – dead. The following formula is then applied:

$$SI = \frac{[On_0 + 1n_1 + 2n_2 + 3n_3 + 4n_4 + 5n_5] \times 100}{n_t \times (n_c - 1)}$$

Where n_1 = the number of plants in disease category 1 etc., n_t = the total number of plants, and n_c = the total number of disease categories.

The SI takes into account the total number of plants as well as the number in each disease category, and gives a better indication of the total disease effect on a population than does the percent curly-top incidence.

RESULTS.—Initial greenhouse tests were conducted with the resistant variety Yakima. Plants growing individually in 10 cm clay pots were covered with 20 cm lamp chimneys with gauze-covered tops. Five viruliferous leafhoppers were placed under each chimney with the plant. These experiments were conducted in late spring and early summer, when the unshaded greenhouse temp ranged from 20-31 C. Presumably temp inside the lamp chimneys on hot sunny days were much higher than greenhouse temp. Under these conditions, after 2 wk 40% (16/40) of the Yakima plants showed mild CTV symptoms, while 100% of the susceptible variety Bountiful developed severe symptoms. The virus was readily recovered from plants with symptoms by indexing with nonviruliferous leafhoppers to a curly top-susceptible variety. These results suggested that high temp during the inoculation-feeding period or

the post-inoculation incubation period may be responsible for the breakdown of resistance. After the chimneys were removed, Bountiful plants eventually died, while Yakima plants remained quite tolerant showing only mild symptoms.

Temperature effects on a resistant and a susceptible variety.—A growth-chamber study was conducted with constant light and photoperiod to determine the effects of two temp ranges on curly-top incidence, severity, and rate of symptom expression on a resistant (Goldcrop) and a susceptible (Tendercrop) variety. The two temp ranges compared were 21-24 C and 31-34 C. Each plant was exposed to four viruliferous leafhoppers for 48 h by attaching a small clip cage (7) containing two leafhoppers on each primary leaf. Results were recorded 15 days after inoculation (Table 1).

In tests with the susceptible variety, the higher temp range increased the total curly-top incidence from 71% to 86%, increased the severity index from 39 to 59, and decreased the average number of days to symptom expression from 10 to 8. No disease was observed in the resistant variety at 21-24 C; but 4% of the plants showed mild symptoms at 31-34 C, supporting the indications from the tests of Yakima that high temp may induce a breakdown of resistance. We were not able to recover the virus with nonviruliferous leafhoppers from the inoculated symptomless Goldcrop plants after 15 days at 31-34 C.

Further studies were conducted to determine whether the increased incidence, severity, and rate of symptom expression at the higher temperatures in susceptible Tendercrop were the effects of high temperature during the inoculation-feeding period (i.e., increased leafhopper activity) or during the post-feeding incubation period. Thirty Tendercrop (susceptible) plants were exposed to an inoculation-feeding at 21-24 C. After a 48-h inoculation-feeding period, 15 plants were maintained at 21-24 C, while 15 were moved to the 31-34 C growth chamber. After 15 days at 21-24 C, 11/15 plants were infected, the severity index was 39 and it took an average of 10 days for symptom development. At 31-34 C, 14/15 plants were infected, the severity index was 60 and it took 7 days for symptom expression. These results were nearly identical with Table 1 data for Tendercrop at those respective temp. This suggests that temp has little if any effect on factors involved in inoculation or the early infection process, but has a profound effect on subsequent disease development during the 2-wk incubation period.

Stage of plant development.—Symptoms on susceptible varieties are usually mildest when plants are inoculated at later stages of growth (5). By analogy it seemed possible that the curly-top resistance in Goldcrop might not be expressed in seedlings at the very early crookneck stage. To test this possibility, 20 Goldcrop seedlings were inoculated in the Crookneck stage, and 20 control plants (Goldcrop) were inoculated in the primary-leaf stage. Each plant was exposed to four viruliferous

TABLE 1. Effect of temp on curly-top incidence, severity, and rate of symptom development in a resistant and a susceptible bean variety

Variety	21-24 C			31-34 C		
	Total infected	Avg days to symptoms	SI ^a	Total infected	Avg days to symptoms	SI
Goldcrop (Resistant)	0/66 ^b	3/66	11	1
Tendercrop (Susceptible)	41/58	10	39	50/58	8	59

^aSeverity Index based upon 0-100 scale 15 days after inoculation, see text.

^bNumber of infected plants out of total tested.

leafhoppers under a lamp chimney for 48 h at 31-34 C. No symptoms were observed after 15 days at 31-34 C in either group, indicating that the stage of plant development at the time of inoculation has no effect on curly-top resistance of Goldcrop. Our preliminary work (Silbernagel and Jafri, *unpublished*) indicates susceptible varieties are equally susceptible during these two early stages of growth.

DISCUSSION.—High temp can induce a breakdown of resistance to curly top in snap beans. The effect of temp on breakdown of curly-top resistance in Goldcrop is consistent with field results (2-5%) and temp maxima observed early in the growing season. Stage of growth at inoculation in resistant Goldcrop apparently is not a critical factor.

In susceptible Tendercrop, the increased disease severity, incidence and rate of expression is apparently due to high temp during the 13 day incubation period, rather than the two day inoculation period.

The existence of new strains of the virus may be a plausible explanation for the 2-5% CTV incidence observed in resistant cultivars, since strains of the virus differing in virulence and pathogenicity on several hosts (1, 15), including beans (6, 8), have been reported. This factor is being investigated in garden beans and they will be reported later. However, in 45 yr of exposure of Burtner's Blightless resistance in snap bean-breeding lines to curly top, one might expect a new strain, once established, to reach a higher level of virulence than an occasional 2-5% under severe exposure conditions; unless Giddings was correct in suggesting that highly virulent strains of CTV tend to be self-limiting (6). He reported two new strains that induced severe injury in a curly top-resistant Red Mexican variety (8); however, curly top has never become an economic factor in varieties carrying resistance from Red Mexican.

Recovery of the virus from infected Yakima plants showing mild curly-top symptoms indicates some tolerance mechanism still operates, even after initial resistance to establishment of the virus is overcome, because symptoms were not as severe as on susceptible varieties. Therefore, resistance from Burtner's Blightless must be physiologic and more than simply resistance to initial infection.

Lack of virus recovery from inoculated, but symptomless, Goldcrop (resistant) plants maintained at 31-34 C is interesting. The virus was not recovered by leafhopper assay from new growth. So unless the virus can overcome the initial resistance to its

establishment, the inoculum apparently is either inactivated or remains at an ineffective titer and/or location.

Why only 2-5% of a resistant variety lose resistance under field conditions is not known, unless the 31-34 C range represents the breakdown threshold. This possibility is indicated by the greenhouse results with Yakima, where 40% of the plants showed symptoms after very high incubation temp inside the lamp chimneys.

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