

Bacterial Vascular Necrosis and Rot of Sugar Beet: Effect on Cultivars and Quality

E. D. Whitney and R. T. Lewellen

Plant Pathologist and Geneticist, respectively, Agricultural Research Service, U.S. Department of Agriculture, U.S. Agricultural Research Station, Salinas, CA 93901.

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ABSTRACT

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An epiphytotic caused by an *Erwinia* sp. on sugar beet in the San Joaquin Valley of California in part was caused by a greater susceptibility of newly introduced hybrid cultivars. The greater susceptibility of the hybrid cultivars resulted predominantly from the use of highly susceptible pollen parents, but also was influenced by the F₁ moderately susceptible seed parent. The *Erwinia*-susceptible pollen parents were selected for virus yellows resistance from a moderately *Erwinia*-resistant parent. This suggested that an

association might exist between yellows resistance and bacterial rot susceptibility. Our results did not support this hypothesis. All cultivars tested were susceptible to some degree to bacterial rot, but there was variability among selections, suggesting that the pathogen probably has been present for many years in soil and became obvious after the introduction of more susceptible cultivars. A disease index (mean percentage of rot per beet) was found to be a reliable means of estimating cultivar susceptibility.

Additional key words: genetic vulnerability, *Erwinia*.

In 1971, in the Lost Hills area of the San Joaquin Valley of California, a root rot of sugar beet occurred which reached epiphytotic levels (7, 8). Not only was the yield of the beets reduced, but the quality of the beets also was impaired, causing problems in sugar factories because of low sugar extraction. In 1972, it was discovered that the incitant of the rot was a bacterium of the genus *Erwinia* (7). New virus yellows-resistant hybrids, US H9A and US H9B (3), were released in 1968 to California growers. The possibility existed that there was an association between the introduction of these cultivars and the epiphytotic that had developed in 1971. Studies were conducted between 1972 and 1975 to determine the susceptibility of cultivars and the effect of bacterial infection on beet quality. Preliminary reports of some of these data have been published (7, 8, 9).

MATERIALS AND METHODS

Tests in 1972.—Three sugar beet hybrids, US H9B, US H10B (3, 4), and S301H8, and the pollinator line of US H9B, C13 (5), were used in these tests. Descriptions of these entries are given in Table 1. Seed of each of the four entries (two adjacent rows per entry) was placed randomly in an eight-row planter and two strips, each 800 m long, were planted at four locations near Lost Hills in

late April and early May. At 3- to 4-wk intervals from 25 July to 16 October, random plots two rows wide and 6 m long were dug from each strip and individual roots were evaluated for percentage of rot and a Disease Index $DI = [(\sum \% \text{ rot per beet}) / (\text{No. of beets per plot})]$ was calculated for each plot. A five-increment scale was used in 1972: 0, 25, 50, 75, and 100% rot. On 21 November, six randomly selected plots (12 m long) were weighed, infected beets were counted, and beets were randomly selected from each plot for sucrose yield and purity analysis.

Tests in 1973.—Experiments at three locations (Dos Palos, Woodland, and Salinas) were conducted to evaluate the susceptibility of virus yellows-resistant and -susceptible selections or breeding lines, components of hybrids, and hybrids (Table 1). Twelve entries were planted at Dos Palos and Woodland and seven entries were planted at Salinas (Table 2). The experiments were arranged in split-plot designs with inoculation treatments as whole plots and entries as subplots. The tests at Dos Palos and Salinas had four replications and the test at Woodland had six. The sugar beets were inoculated about 10 wk after planting. Five *Erwinia* isolates, UR-7, SB-4, SB-6, SB-7, and SB-13, were grown on medium B of King et al. (2) for 40 hr at 26 C. Suspensions of each isolate in tap water were standardized to a concentration of cells equivalent to an absorbency of 200 with a Klett-Summerson colorimeter and equal proportions of each were mixed. The composite stock solution was diluted 1:7 (approximately 10^7 cells/ml) with tap water and was applied into the crowns of noninjured beet plants at the

rate of 10 ml/30 cm of row (6.7^{10} cells/plant) with a pressurized sprayer. Plots were harvested 5.5 mo after planting and yield and sucrose percentages were determined. Each root was sliced to estimate the percentage of rot. A six-part increment scale was used in 1973 to calculate the DI: 0, 1, 15, 50, 85, and 100 percent rot. The beets from the plot at Salinas also were evaluated for beet quality.

Two approaches were used to evaluate beet quality. In the first, known weights of rotted and healthy beets of US H9B were mixed and run through a beet rasp to collect the

brei (one test, five replications). In the second, paired adjacent beets, one completely rotted and one healthy, were selected in the field, run individually through a beet rasp to collect the brei and amounts (w/w) of the brei from the healthy and rotted beets then were mixed (three tests, 57 paired samples). The percentage of rotted beet brei in the samples was classified as 0, 10, 20, 40, and 100. The brei samples were analyzed for percent sucrose and concentrations of amino nitrogen, sodium, and potassium. From these data, an impurity index was estimated (6).

TABLE 1. Hybrids, breeding lines, cultivars, and selections of sugar beet that were tested for reaction to *Erwinia* infection during 1972-1975 at Salinas, Lost Hills, Dos Palos, and Woodland, California

| Sugar beet entry | Description | Characteristic |
|------------------|---|---|
| US H9B | 546H3 × C13 | commercial hybrid, yellows-resistant |
| US H10B | 546H3 × C17 | commercial hybrid, yellows-resistant |
| US H7A | 546H3 × C64 | commercial hybrid, yellows-susceptible |
| S301H8 | 569H3 × S3 | commercial hybrid, yellows-susceptible |
| HH 23 | Holly hybrid | commercial hybrid, yellows-resistant |
| US 75 | Parent of C13 and C17 | commercial open- pollinated cultivar, yellows-susceptible |
| C13 | Pollen parent of US H9B | yellows-resistant |
| C17 | Pollen parent of US H10B | yellows-resistant |
| Maris Vanguard | English cultivar | yellows-resistant |
| Y03 | Selected from accession from The Netherlands | yellows-resistant |
| C64 | Pollen parent of US H7A | yellows-susceptible |
| 546H3 | C562CMS × C546 seed parent of US H9B and US H10B | F ₁ hybrid, yellows- susceptible |
| 718H54 | C705CMS × C718 | F ₁ hybrid, yellows- resistant |

TABLE 2. Effect of inoculation and sugar beet cultivar on severity of disease incited by an *Erwinia* sp. at three California test locations

| Sugar beet entry | Disease index ^c observed at test location: | | | | | | |
|--------------------|---|-------|-----------------------|-------|----------------------|-------|------------------------|
| | Dos Palos ^a | | Woodland ^b | | Salinas ^a | | |
| | Noninoc. | Inoc. | Noninoc. | Inoc. | Noninoc. | Inoc. | Na(mg/kg) ^d |
| C17 | 15.1 | 37.5 | 22.1 | 53.6 | 15.9 | 33.3 | 219.4 |
| Y03 | 4.8 | 9.4 | 6.9 | 15.4 | 8.3 | 10.0 | 641.5 |
| Maris Vanguard | 1.0 | 10.3 | 9.9 | 23.1 | 6.5 | 12.7 | 757.6 |
| US 75 | 3.7 | 9.7 | 9.2 | 31.7 | 5.2 | 8.4 | 672.0 |
| 546H3 | 4.2 | 8.8 | 14.0 | 32.5 | 3.0 | 10.6 | 425.0 |
| US H7A | 5.0 | 5.7 | 17.6 | 27.3 | 5.1 | 7.3 | 387.5 |
| US H10B | 9.6 | 20.1 | 12.7 | 52.0 | 13.5 | 23.6 | 483.4 |
| C64 | 2.4 | 2.9 | 7.5 | 14.9 | | | |
| 718H54 | 4.1 | 17.5 | 21.8 | 45.6 | | | |
| US H9B | 5.6 | 25.2 | 7.6 | 39.6 | | | |
| S301H8 | 2.4 | 10.9 | 12.1 | 27.7 | | | |
| HH 23 | 1.2 | 8.5 | 10.1 | 24.8 | | | |
| LSD ($P = 0.05$) | 13.3 | 13.3 | 10.8 | 10.8 | 9.6 | 9.6 | 267.0 |

^aMean of four replications.

^bMean of six replications.

^cDisease Index (DI) = (Σ % rot per beet)/(No. of beets per plot).

^dFresh weight.

The 1974 and 1975 tests.—The yellows-resistant pollinator of US H9B, C13, and selections made from C13 for *Erwinia* resistance were tested in split-plot designs to determine whether selecting for *Erwinia* resistance affected virus yellows resistance. Virus inoculation treatments were whole plots and selections were subplots. Viruliferous aphid-infested beet leaf pieces were used for virus yellows inoculations as outlined by Bennett et al. (1). Yield and sucrose percentage were determined at harvest time.

RESULTS

Cultivar susceptibility.—Two of the four field tests conducted in 1972 developed sufficient natural infection to permit evaluation of the entries. The pollen parent of US H9B, C13, was more susceptible than the other entries tested (Table 3). The DI's of the four entries were similar early in the season but increased more rapidly in C13 as the season progressed. The yellows-resistant hybrids appeared to be more susceptible than the yellows-susceptible hybrid, S301H8, but not significantly so. The mean percentages of beets in each of the five incremental categories for the four cultivars in 1972 was 0 = 76.6, 25 = 1.4, 50 = 2.8, 75 = 1.4, and 100 = 18.0. Sucrose determinations and beet quality did not differ among the four entries.

Of the variables tested in 1973 (root yield, DI, percent sucrose, and gross sugar yield), DI was the best indicator of the susceptibility. The DI ranged from 7.3 to 33.3 at Salinas, 14.9 to 53.6 at Woodland, and 2.9 to 37.5 at Dos Palos. Parental line C17, the pollen parent of US H10,

presently the most popular hybrid cultivar in California, was the most susceptible entry at all locations. The parent of C17, US 75, was more resistant than C17 at all locations; DI = 16.6 vs. 41.5. In all tests, bacterial rot developed in the noninoculated blocks of beets. Root yield at Dos Palos and percent sucrose at Woodland were not reduced in the inoculated plots. A cultivar by inoculation interaction, as measured by DI, was significant at all locations but the interaction was not significant for sucrose at any location or for root yield at Salinas and Dos Palos. Maris Vanguard and Y03, European cultivars resistant to virus yellows, were more *Erwinia*-resistant than C17, a USA virus yellows-resistant cultivar. The mean DI's for inoculated plants of Y03, Maris Vanguard, and C17 from all three locations were 116.6, 15.4, and 41.5, respectively. The F₁ hybrid, 546H3, the seed parent of US H9B, and US H10B, was more resistant than the pollen parent C17, with a mean DI of 17.3 vs. 41.5. Hybrid US H7A, 546H3 × C64, a predecessor of US H9B and US H10B, was resistant to bacterial rot in all tests. The pollen parent, C64, was the most resistant selection tested.

Effect on beet quality.—Two types of rot occurred in the field: a wet rot and a dry rot. Dry rot causes almost complete disintegration of the affected beet or beet part. However, beet roots with wet rot do not disintegrate and may be harvested and taken to the sugar factory. A beet with dry rot that has partially disintegrated may be harvested, and adds little diseased material to affect the sugar extraction process.

The two methods of collecting and mixing the brei from wet rot beets gave essentially the same results. There was

TABLE 3. Evaluation of the performance of four sugar beet cultivars when tested under moderately severe natural infection by an *Erwinia* sp.

| | Cultivars | | | | LSD (<i>P</i> = 0.05) |
|--------------------|-------------------|---------|--------|-------------------|---------------------------|
| | US H9B | US H10B | S301H8 | C13 | |
| Infected beets (%) | 13.7 ^a | 13.4 | 9.6 | 32.7 | 8.0 |
| Yield (tonnes/ha) | 81.2 | 78.0 | 81.4 | 59.6 ^b | 7.2 |
| Sucrose (%) | 12.3 | 13.0 | 12.4 | 12.8 | NS ^c |
| Na(mg/kg) fresh wt | 1647 | 1484 | 1757 | 1368 | NS ^c |

^aMean of two tests with six replications per test.

^bSugar beet line C13 consistently yields about 95% of US H9B and US H10B.

^cNS = not significant.

TABLE 4. The effect of percentage rot on beet quality criteria

| Beet quality criterion | Percentage rot ^a | | | | |
|------------------------------------|-----------------------------|--------|--------|--------|---------------------|
| | 0% | 10% | 20% | 40% | 100% |
| Sucrose (%) | 12.0 ^b | 11.0 | 10.1 | 8.0 | 1.8 |
| NH ₂ -N(mg/kg) fresh wt | 776.7 | 776.8 | 787.9 | 883.1 | 1043.5 ^c |
| Na (mg/kg) fresh wt | 1942.7 | 1951.5 | 1962.4 | 2020.0 | 2196.7 |
| Impurity index ^d | 1703 | 1882 | 2067 | 2759 | 13510 |

^aBrei from healthy beets and beet with wet rot were mixed (1:1, w/w).

^bMean of four tests (62 samples) of US H9B.

^cAn increase in two of four tests at *P* = 0.10.

^dThe impurity index was calculated as follows: $[10 \times \text{NH}_2\text{-N(mg/kg)} + 3.5 \times \text{Na(mg/kg)} + 2.5 \text{K(mg/kg)}]$
Sucrose percentage

an inverse linear relationship between percent rot and percent sucrose (Table 4). In two of the four tests, amino nitrogen was increased in beets that were 100% rotted. Neither sodium nor potassium concentrations were affected. However, the sodium concentration of the beets from the Lost Hills area was three to four times that of beets grown in the Salinas Valley (Tables 2 and 3).

Virus yellows tests of *Erwinia*-resistant selections.—None of the lines from C13 selected for *Erwinia* root rot resistance was more susceptible to virus yellows. Yield and percent sucrose losses from virus yellows were not significantly higher for the selections than for the parent (Table 5).

DISCUSSION

An important factor contributing to the epiphytotic of the *Erwinia* sp. on sugar beets in California was the introduction of new hybrid cultivars US H9B and US H10B which are more susceptible than formerly grown hybrids; i.e., US H7A and S301H8. This increased susceptibility resulted from the use of highly susceptible pollen parents, C13 and C17, introduced into hybrids grown in California because of their superior production under conditions of virus yellows infection. Because US 75, the parent of C13 and C17, is less susceptible to *Erwinia* sp. than the yellows-resistant selections, the possibility existed that virus yellows resistance may be associated with bacterial rot susceptibility. However, this does not seem to be the case. Maris Vanguard and Y03, two European virus yellows-resistant cultivars, were among the most bacterial rot-resistant entries tested, suggesting that an association does not exist between yellows resistance and bacterial rot susceptibility. However, the nature of resistance to virus yellows may be different in the USA and European selections.

Most of the infected beets in the 1972 test had developed dry rot and were not removed from the field at harvest time; therefore, percent sucrose and beet quality

of the samples were not affected by rot, but yield was drastically reduced in cultivar C13. Therefore, if infection occurs early and no subsequent infection occurs, most of the rotted beets will have disintegrated and will not be harvested. However, late-infected beets with wet rot will be harvested and will affect sucrose content and beet quality.

The efficiency of sugar extraction is a function of the sucrose content of the beets and the concentrations of sodium, potassium, amino nitrogen, and other plant constituents which are referred to as impurities. Therefore, with the higher than normal impurities and lower sucrose in beets with wet rot, the difficulty of sucrose extraction is compounded.

Our data show that resistance to *Erwinia* sp. is not associated with virus yellows resistance because resistance was maintained during selection for bacterial rot resistance without simultaneously testing for yellows resistance. Because yellows resistance apparently is multigenically inherited, an association was not likely; however, it was necessary to evaluate this possibility.

All of the cultivars and selections tested possessed plants that were resistant or susceptible to bacterial rot, suggesting that bacterial rot has been a disease of sugar beet for many years but became highly vulnerable only when more susceptible lines of beet were introduced.

Virus yellows resistance, curly top resistance, and nonbolting tendency are the general requirements needed for the culture of sugar beet in the California-Arizona Region, and each of these should be present in a commercial cultivar. Therefore, it was not possible to replace the pollen parent, C17 or C13, with the European virus yellows-resistant selections which do not have curly top resistance.

Why the selected cultivars, C13 and C17, are more susceptible than the parent is not known.

We hypothesize that with the great differences in susceptibility of the cultivars, a cultivar by inoculation interaction (as measured by yield) would be evident or that the yield of resistant cultivars would be less affected by inoculation. However, in our tests, the spread of the pathogen from inoculated to noninoculated plots, natural infection of beets in noninoculated plots, and the compensatory effect of adjacent healthy beets in inoculated plots are probably reasons for masking of the interaction.

TABLE 5. Effect of yellows infection on loss of yield and sucrose of *Erwinia*-resistant selected offspring and their common parent, C13

| Cultivar | Root yield loss (%) | Sucrose loss (%) | DI ^c |
|-------------------|---------------------|------------------|-----------------|
| E301 ^a | 24.5 | 8.8 | 18.3 |
| E307 | 18.4 | 6.2 | 20.1 |
| C13 | 30.8 | 6.0 | 40.0 |
| LSD ($P=0.05$) | 9.3 | NS ^d | 6.4 |
| E402 ^b | 16.3 | 7.5 | 1.3 |
| E406 | 15.4 | 10.3 | 0.8 |
| E434 | 20.2 | 8.0 | 1.7 |
| E435 | 13.4 | 7.3 | 1.7 |
| C13 | 18.3 | 9.8 | 20.4 |
| LSD ($P=0.05$) | NS ^d | NS ^d | 5.1 |

^aCultivar designation E3- - identifies breeding lines after one cycle of selection for resistance to *Erwinia* from C13.

^bCultivar designation E4- - identifies breeding lines after two cycles of selection for resistance to *Erwinia* from C13.

^cDisease index (DI) = ($\Sigma\%$ rot per beet/No. beets per plot) data from separate *Erwinia*-inoculated tests.

^dNS = not significant.

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