Sugar and Root Yield of Sugar Beets as Affected by Bacterial Vascular Necrosis and Rot, Nitrogen Fertilization, and Plant Spacing

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ABSTRACT

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In field experiments at Davis, CA, nitrogen (N) fertilization increased the incidence of rot caused by *Erwinia carotovora* subspecies *betavasculorum*. When plants were inoculated, rot per beet root increased from 11% with no fertilizer N to 36% when 336 kg N/ha was applied. The sucrose concentration of roots declined at a faster rate with increasing rates of fertilizer N when plants were inoculated than when not inoculated, and root yield increased at a slower rate with N fertilization when plants were diseased. Sugar yield estimations from regressions increased for healthy plants but decreased for diseased plants as fertilizer N increased. As the in-row spacing between plants was increased from 10 to 46 cm, the percent of diseased roots increased more rapidly when plants were inoculated than when they were not inoculated. Root sucrose concentration decreased with increasing in-row spacing but the rate of decline was more rapid for inoculated than for uninoculated plants. For uninoculated plants, root and sugar yields were maximized at in-row spacings of 15–30 cm but, when plants were inoculated, these yields declined linearly as in-row spacing increased from 15 cm. In field tests at Salinas, CA, inoculated plants of *Erwinia*-susceptible cultivar C17 showed more rot per root with increased in-row spacing than did inoculated plants of moderately susceptible cultivar 546H3.

Additional key words: cultural control, soft rot, disease assessment, crop loss.

Bacterial vascular necrosis and rot of sugar beet (*Beta vulgaris* L.) was first reported in 1973 in the San Joaquin Valley of California (7). Subsequently the disease was found in Washington, Arizona, and Idaho (4,5,8) and in many cases has caused significant yield losses. The pathogen is similar to other species in the *Erwinia carotovora* group and has a proposed name of *Erwinia carotovora* subsp. *betavasculorum* (6).

Losses from the disease will probably be reduced in the future because of the identification of genetic sources that contributed to susceptibility of sugar beet cultivars U.S. H9 and U.S. H10 and the development of resistant cultivars (3,10,11).

In lieu of resistant cultivars, sugar beet growers have asked if a change in cultural practices would influence incidence and severity of the disease. The effects of spacing and nitrogen (N) nutrition were of primary concern since these factors appeared to influence the amount of disease; spacing and N fertilization are variable throughout California. Growers also wanted to know how much loss of sugar yield was caused by the disease. The research reported here was designed to answer those questions.

MATERIALS AND METHODS

Nitrogen nutrition. The effect of nitrogen nutrition on the susceptibility of sugar beet to *E. carotovora* subsp. *betavasculorum* was evaluated by using cultivar U.S. H10 in field plot experiments. The experimental area near Davis, CA, was planted 6 May 1974, with a row spacing of 76 cm.

The design was a split plot with main plots of uninoculated plants or plants inoculated with *E. carotovora* subsp. *betavasculorum* and subplots of six rates of N fertilization. Each treatment was replicated four times for a total of 48 plots. Subplots were six rows wide and 15.2 m long. Ammonium nitrate was side-dressed on 21 May about 25 cm from both sides of each row. Six N rates were established in increments of 67 kg from 0 to 336 kg N/ha.

The center two rows of appropriate subplots were inoculated without wounding on 12 July. *E. carotovora* subsp. *betavasculorum* inoculum (Strains UCBPP 173, UCBPP 175, UCBPP 176, and UCBPP 193) was prepared by washing bacteria from 24-hr-old cultures on King's medium B (2) with tap water and adjusting the final concentration to 4×10^8 colony-forming units (cfu) per milliliter. Approximately equal numbers of each strain were present in the inoculum. One-tenth milliliter (about 4×10^7 cfu) was applied to the crown of each inoculated plant with a CO_2-pressurized spray tank.

Beets from 7.6 m of the center two rows of each plot were harvested on 1 October, 21 wk after planting and 11 wk after inoculation. Only roots that would be delivered to a processing factory were evaluated. Plants were topped and weighed, and two random samples of 10 roots were taken for sugar and tare analysis. The remaining roots were cut and rated for the percentage of rotted beet by using a disease rating scale—0, 7, 25, 50, 75, 93, and 100%.

Spacing. The effect of in-row spacing on yield loss and incidence of disease was investigated to determine the best spacing for optimum yield of sugar beets in the presence of bacterial vascular necrosis. Sugar beet cultivar U.S. H9 was planted 6 May 1974 in field plots in rows spaced 76 cm apart at Davis, CA. Beets were thinned by hand 3–8 June to 10, 15, 30, and 46 cm in-row spacing. Treatments were arranged in a split-plot design with inoculated and uninoculated areas as main plots in six replications. In-row spacings were subplot treatments four rows wide and 15.2 m long. The center two rows of appropriate subplots were inoculated without wounding on 26 July with strain UCBPP 193 as previously outlined. Inoculum suspensions containing approximately 1×10^7 cfu were sprayed on the crown of each inoculated plant.

Beets from 7.6 m of the center two rows of each plot were harvested 9 October, 22 wk after planting and 11 wk after inoculation. Plants were topped and weighed, and two random samples of 10 roots were cut and rated for percentage of rot as previously described.

Spacing × cultivar. Spacing × cultivar tests were conducted at Salinas and Spence, CA. Three cultivars, C17, 546H3, and U.S. H10, were seeded at the two locations in a split-plot design with spacings as whole plots and cultivars as subplots. Each treatment had four replications. Stands were thinned about 4 wk after seeding

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to the appropriate in-row spacings of 10, 20, 30, or 40 cm. The bacteria were cultured and inoculations were performed as previously described (3,10) when the plants were 10 wk of age at each location. Five months after seeding, plots were harvested and plants were weighed, analyzed for percentage sucrose, and the percentage of rot was estimated as before by slicing each beet with a knife. A combined analysis of the data was performed for the tests at Salinas and Spence since there was not a location effect.

RESULTS

Nitrogen nutrition. Inoculation with *E. carotovora* subsp. *betavasculorum* decreased the number of harvestable beets at all levels of N fertilization. Inoculation resulted in 17% fewer plants at harvest than when plants were not inoculated (Table 1). Due to very few infected roots and the low percentage of rot per beet in uninoculated plots, these plots were not included in the analysis of variance for these variables. N fertilization resulted in linear increases in the percentage of infected roots and the percent rot per root, both increasing about 0.4%/kg fertilizer N. Linear regressions for percent infected roots and percent rot per root on N rate were, respectively: $\hat{Y} = 25.9 + 0.04$ N, $r^2 = .752$ and $\hat{Y} = 13.4 + 0.04$ N, $r^2 = .878$.

As previously reported by Whitney and Lewellen (10), there was a significant decrease in the sucrose concentration of infected roots. However, their results were based on a technique that involved the mixing of certain quantities of rotted beets with healthy beets. In our study the sucrose concentration also declined linearly with increasing fertilizer N (9). The rate of decline was essentially the same for both uninoculated and inoculated plants with a lower sucrose concentration in diseased roots at all levels of fertilization. Regressions of percent root sucrose on kilograms of fertilizer N per hectare for uninoculated and inoculated plants, respectively, are: $\hat{Y} = 13.1 - 0.0063$ N, $r^2 = 0.979$ and $\hat{Y} = 12.7 - 0.0079$ N, $r^2 = 0.985$.

With respect to root yield, there was a significant linear interaction of nitrogen rate with inoculation. The means presented in Table 1 show that the response to fertilizer N leveled off at 202 kg/ha. Regressing root yield on fertilizer N up to 202 kg/ha for uninoculated beets gives $\hat{Y} = 44.6 + 0.04$ N, $r^2 = 0.855$ and for inoculated plants gives $\hat{Y} = 37.0 + 0.02$ N, $r^2 = 0.736$, a rate increase only one half that of the uninoculated plants (ie, 0.04 vs 0.02). From these regression equations and those for the effect of N rate on percent sucrose for uninoculated and inoculated plants, the estimated sugar yields, respectively, for 0, 67, 137, and 202 kg N/ha are 5.84, 6.01, 6.15, and 6.22×10^3 kg/ha for uninoculated beets and 4.70, 4.67, 4.61, and 4.55×10^3 kg/ha for inoculated plants. Thus, total sugar yield per hectare increased with N fertilization when plants were not inoculated but decreased with N fertilization when plants were diseased.

Spacing. When healthy plants were closely spaced, a loss in plants from early to late season was observed, but few plants were lost when roots were spaced 30 cm or more (1). Table 2 shows a similar effect in our study but, when plants were inoculated with *E. carotovora* subsp. *betavasculorum*, greater losses occurred at all spacings. The percentage of diseased roots increased from about 5 to 18% in uninoculated plots as in-row spacing was increased from 10 to 46 cm; in inoculated plots the increase was from about 14 to 39%. The predicted percent of diseased roots for a given spacing in inoculated plots is: $\hat{Y} = 7.8 + 0.71$ cm, $r^2 = 0.928$. The amount of rot per root also increased as spacing between plants increased. For

TABLE 1. Effect of nitrogen (N) fertilization on sugar beets inoculated and not inoculated with *Erwinia carotovora* subsp. *betavasculorum*

Fertilizer nitrogen (kg/ha)	Marketable roots per plot			Infected roots (%)	Rot per root (%)	Root sucrose concentration (%)			Root yield (t/ha)		
	NI[a]	I[a]	$\bar{N}$[b]	I	I	NI	I	$\bar{N}$	NI	I	$\bar{N}$
0	64	53	58	22	11	13.0	12.8	12.9	44.1	36.6	40.3
67	62	49	55	31	18	12.9	12.1	12.5	49.3	39.4	44.4
134	58	52	55	33	19	12.2	11.5	11.8	48.6	38.4	43.5
202	64	52	58	36	24	11.8	11.0	11.4	54.1	41.1	47.6
269	60	50	55	39	23	11.4	10.7	11.0	54.5	40.8	47.6
336	60	50	55	36	26	11.0	10.0	10.5	53.9	40.3	47.1
$\bar{I}$[c]	61	51				12.0	11.4		50.7	39.4	
Statistical significance[d]	I**[e]	N_{ns}	$N \times I_{ns}$	NL*[e]	NL**	I*	NL**	$N \times I_{ns}$	I**	NL**	NL × I*

[a]Not inoculated and inoculated, respectively.
[b]Nitrogen means.
[c]Inoculation means.
[d]NL refers to a linear effect of N rate, ns is not statistically significant ($P > 0.05$).
[e]*Significant at $P = 0.05$, **significant at $P = 0.01$.

TABLE 2. Effects of in-row sugar beet spacing and inoculation with *Erwinia carotovora* subsp. *betavasculorum* on several variables affecting root and sugar yield

In-row spacing (cm)	Dead plants per plot			Roots diseased (%)			Rot per root (%)			Root sucrose concentration (%)			Nitrate rating on root pulp (1–4)[c]		
	NI[a]	I[a]	$\bar{S}$[b]	NI	I	$\bar{S}$	NI	I	$\bar{S}$	NI	I	$\bar{S}$	NI	I	$\bar{S}$
10	33	61	47	4.7	13.7	9.2	1.0	6.2	3.6	13.2	13.3	13.3	1.0	1.2	1.1
15	8	31	20	4.6	17.6	11.1	2.0	7.9	5.0	13.2	12.9	13.1	1.2	1.1	1.1
30	1	24	12	8.8	34.3	21.6	5.4	19.2	12.3	12.8	12.3	12.6	2.0	2.1	2.0
46	1	20	11	18.0	39.1	28.5	12.2	26.0	19.1	12.4	11.3	11.9	2.4	2.5	2.5
$\bar{I}$[d]	11	34		9.0	26.2		5.2	14.8		12.9	12.4		1.6	1.7	
Statistical significance[e]	I**[f]	S**	I × SL*[f]	I**	S**	I × SL*	I**	S**	I × SL*	I**	S**	I × SL**	I_{ns}	SL**	$I \times S_{ns}$

[a]Not inoculated and inoculated, respectively.
[b]Spacing means.
[c]Blue color development rating (1–4) on exposure to 2,000 ppm diphenylamine in concentrated sulfuric acid (9).
[d]Inoculation means.
[e]SL refers to a linear effect of spacing, ns means not statistically significant ($P = 0.05$).
[f]*Significant at $P > 0.05$, **significant at $P > 0.01$.

inoculated plants the predicted percent rot per root is: $\hat{Y} = 0.29 + 0.57$ cm, $r^2 = 0.984$.

With increased plant spacing root sucrose concentration declined more rapidly in the inoculated plants than in the uninoculated plants (Table 2). The percent sucrose in inoculated roots for a given spacing is estimated by $\hat{Y} = 13.8 - 0.05$ cm, $r^2 = 0.990$ and for uninoculated roots, $\hat{Y} = 13.5 - 0.02$ cm, $r^2 = 0.984$. Nitrate ratings showed comparable increases in nitrate in beet roots for both uninoculated and inoculated plants as spacing increased. The negative relationship between N in beet plants and sugar concentration is well known (9) as is the effect of Erwinia rot on sucrose concentration (10). Thus, the observed interaction of inoculation × spacing is a logical effect of higher concentrations of nitrogen in plants as spacing was increased and more rot occurred in the inoculated plants.

With respect to both root and gross sugar yield, there was a highly significant nonlinear effect of spacing on root yield and a highly significant interaction of inoculation × spacing, indicating a differential root yield response to spacing depending on whether plants were inoculated or not (Fig. 1). These results indicate that in-row spacings between 15 and 30 cm produced maximum root and sugar yields for healthy plants as others have shown (1), but when Erwinia rot is present, yield decreases almost linearly as spacing increases from 15 cm. This decrease in yield is associated with changes in number of roots and the consequences related to increases in spacing: greater loss of roots, increase in the percent of roots with rot, more rot per root, and more nitrate per root. While sucrose yield changed very little for uninoculated plants spaced from 15 to 46 cm, the sucrose yield for inoculated plants declined and is estimated by $\hat{Y} = 61.7 - 0.61$ cm, $r^2 = 0.983$, where $\hat{Y}$ is 100 × kg sugar per hectare.

Spacing × cultivar. The percent rot of each cultivar (U.S. H10, 546H3, and C17) increased as spacing was increased (Fig. 2). Less rot developed with closer spacings of beets and compared favorably with the above study on spacing. A spacing × cultivar interaction was shown for the combined analysis at Salinas and Spence. The susceptible cultivar, C17, had a significantly higher percent rot at the wider spacings than the intermediately susceptible cultivar, U.S. H10 and the moderately resistant cultivar, 546H3 (10) (Fig. 2). None of the other factors measured (root yield, sucrose yield, or percent sucrose) showed a spacing × cultivar interaction. Yields of roots and sugar were highest at spacings of 10–20 cm between beets. Sucrose percent was also highest at the narrower spacings.

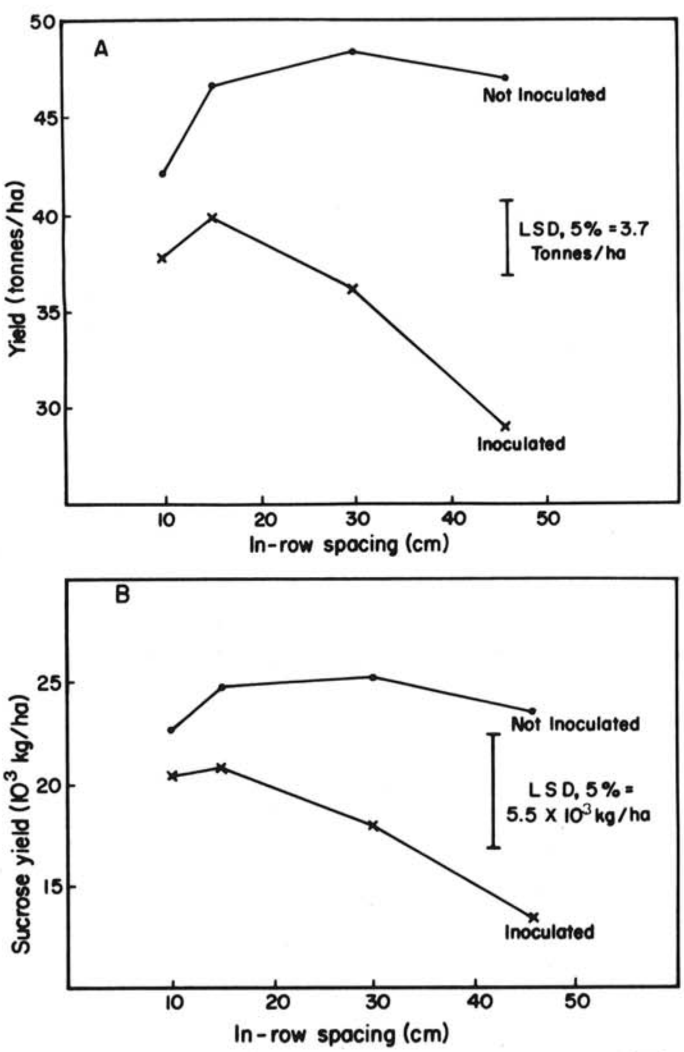

Fig. 1. Effect of in-row spacing on **A,** sugar beet yield **B,** sucrose yield in plants inoculated with *Erwinia carotovora* subsp. *betavasculorum*.

DISCUSSION

The incidence of bacterial vascular necrosis and rot of sugar beet may be reduced by cultural practices that reduce the occurrence of cracks and injuries that serve as portals of infection for the bacteria (8). Growers consistently report that the susceptible sugar beet cultivars U.S. H9 and U.S. H10 readily crack, especially when growing rapidly with high N fertilizer rates. Our field studies showed that increased N fertilizer in inoculated plots substantially increased the incidence of disease. However, there was no corresponding increase in root or sugar yield in contrast to the significant increases that were obtained with higher N fertilizer rates in uninoculated plots. Thus, the benefits of higher N fertilizer realized in healthy beets may not be obtained in beets diseased with *E. carotovora* subsp. *betavasculorum*.

A greater yield loss resulting from rot occurred with widely spaced beets in both inoculated and uninoculated plots, despite the high mortality rate in the closer spacings. Compensation probably resulted from the ability of those beets adjacent to dead ones in the closer spacings to grow larger because of less competition for water, nutrients, and sunlight. Beets planted at wider spacings were already growing near their maximum rates, and additional space or nutrients made available by the death of a plant apparently had little effect on adjacent healthy beets. This phenomenon is exemplified in the cultivar trial in which the susceptible cultivar (C17) was affected to a greater extent than the resistant cultivar (546H3) at the wide spacing (40 cm).

Similar to the effects of higher N rates in the fertilization experiment, the more widely spaced beets were larger and had more

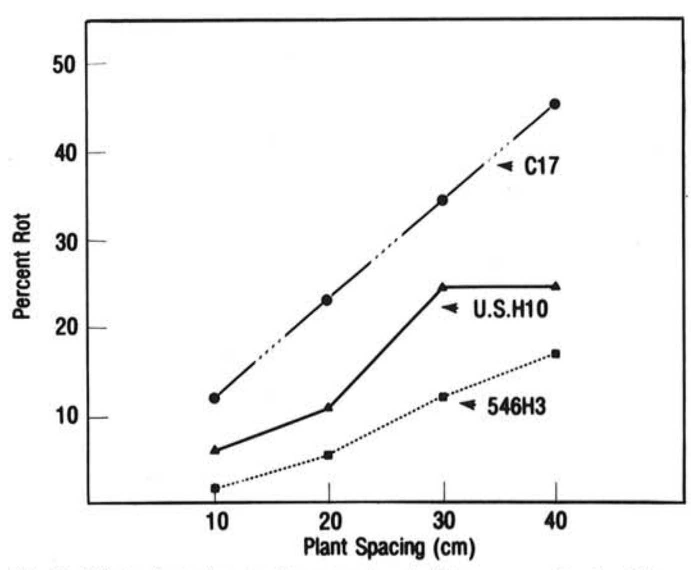

Fig. 2. Effect of spacing on the percent rot of three sugar beet cultivars inoculated with *Erwinia carotovora* subsp. *betavasculorum*. Tests were performed at two locations, Salinas and Spence, CA. Each datum represents the overall mean of four replications for each location. The slope of the C17 line is significantly different from the other two, which indicates a spacing × cultivar interaction. $LSD_{0.05} = 6.0$.

growth cracks and split petioles, which are entry portals for the pathogen. Greater availability of nitrogen appears to be partly responsible for increased disease in the spacing trials, and this is substantiated by the higher amounts of nitrates in the wider spaced beets (Table 2). Our results indicate that in-row spacing of 15–30 cm and optimum N rates for each location are practices that should help reduce losses when planting susceptible sugar beet cultivars. These cultural practices coupled with the use of resistant cultivars should result in satisfactory control of bacterial vascular necrosis of sugar beets.

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