

## Comparison of Thiabendazole and Genetic Resistance for Control of Sugar Beet Storage Rot

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### ABSTRACT

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The fungicide 2-(4-thiazolyl)benzimidazole (thiabendazole) protected roots of two susceptible sugar beet cultivars from storage rot caused by *Phoma betae*, *Botrytis cinerea*, and *Penicillium claviforme* when roots were stored at 5 C and 100% relative humidity for 100–150 days. A similar reduction in storage rot was measured for roots of two breeding lines with resistance to those storage pathogens. Susceptible roots that were inoculated with a mixture of the three pathogens suffered 12–13% rot (fresh weight basis) in 1976 and 2% in 1977. In roots treated with thiabendazole, rot was 3–7% in 1976 and 0.02–0.1% in 1977 and inoculated roots of the

resistant breeding lines suffered 3–8% rot in 1976 and 0.6–1.2% in 1977. Statistically significant losses in sucrose, clear juice purity, and recoverable white sugar per ton occurred during the 1977–1978 storage season in susceptible inoculated roots that averaged 2% rot by weight. Loss in juice quality was less in roots of commercial sugar beet cultivar American Crystal 2 hybrid B treated with thiabendazole than in those of untreated roots. There was no change in juice quality of the inoculated resistant roots. The genetic storage rot resistance in the two breeding lines used here may afford protection comparable to that provided by thiabendazole.

The use of 2-(4-thiazolyl)benzimidazole (thiabendazole) to control storage rot of sugar beet (*Beta vulgaris* L.) is increasing. The Environmental Protection Agency has granted specific exemption to processors in Washington, Idaho, and Michigan to treat over 400,000 tons of roots. Germplasm resistant to storage rot is available (1), but control with genetic resistance will not be possible until that resistance has been incorporated into commercially acceptable cultivars. However, an estimate of the relative benefits of genetic resistance and chemical control would be valuable and was the purpose of this study. Such a comparison was not possible until recently when enough seed of resistant germplasms became available.

### MATERIALS AND METHODS

Two storage rot-susceptible cultivars were used: American Crystal 2 hybrid B (2B) and Great Western 'Mono Hy D-2' (D2). Two storage rot-resistant selections were used for comparison: 75P1 and 7326. The breeding line 7326 was developed by three generations of selection for resistance to *P. betae*. The original population was developed for *Rhizoctonia* crown rot resistance. The breeding line 75P1 was a product of a single cycle of selection at Fargo for resistance to *P. betae* and was an introduction from the USSR, where it had been selected for resistance to *B. cinerea*.

Two plots of each susceptible cultivar and one plot of each breeding line were planted in a randomized complete block design with seven blocks. There were 42 plots in the field design. Roots from the two middle rows of four-row plots 9.1 m long were harvested and washed. Roots from one plot of each susceptible cultivar in each block were dipped 10–15 sec in a flowable formulation of thiabendazole at 1,500 µg/ml. The roots from each plot were then divided into four lots. Lot 1 was wounded and inoculated; lot 2 was only wounded; lot 3 was neither inoculated nor wounded; and lot 4 was processed at harvest to determine quality.

Roots were inoculated by wounding at two sites on opposite sides of each root. Wounds were made by inserting with a twisting motion an 11-mm-diameter cork borer to a depth of 8–10 mm. The end of the cork borer had a serrated edge and was dipped in

inoculum before it was used to wound each root. The inoculum consisted of a mixture of conidia of *Phoma betae* Frank, *Penicillium claviforme* Banier, and *Botrytis cinerea* L. suspended in 0.1% water agar. Roots in lot 2 were wounded with a cork borer that had been dipped in ethanol and flamed. The roots were incubated in perforated plastic bags at 4–6 C and near 100% relative humidity.

The roots were stored after inoculation for 150 days in 1976–1977 and 100 days in 1977–1978. The amount of rot was determined for each bag of roots by excising the rotted tissue, weighing it, and expressing rot as a percent of the total root fresh weight.

The quality parameters measured were sucrose content, clear juice purity (CJP) and recoverable white sugar per ton of roots (RWST). Sucrose was measured with a polarimeter following juice preparation by the cold digestion method (3). The CJP was determined by the method described by Dexter et al (4). Purity was calculated by the formula  $100 \times \text{sucrose content (by refractometry) of root extract after clarification with CaO and filtration}$ . The RWST was calculated with an assumed factory loss of 0.3% and a molasses purity of 62.5% (4). The data were corrected for root shrinkage during storage.

### RESULTS

Yields of roots per hectare were greater in 1977 than in 1976 (Table 1). The commercial cultivars D2 and 2B yielded about the same in the dry year of 1976 but D2 yielded more than 2B in 1977 when moisture was not limiting. The breeding line 75P1 yielded about the same as 2B but significantly less than D2 when yields from both years were averaged. Line 7326 yielded significantly less than the others, because of low vigor and poor stands. It had been selected for three generations, whereas 75P1 had been selected only once.

The roots produced and stored 100 days in 1977 developed less rot than those produced and stored 150 days in 1976 (Tables 2 and 3). This difference in rot cannot be attributed entirely to the 50-day difference in the storage period. The roots in the wounded but noninoculated treatment developed rot during the 1976–1977 storage season, whereas rot in the 1977–1978 storage season was insignificant in that treatment group. Rot developed even in the nonwounded roots in 1976–1977, which indicated that pathogens entered through injuries inflicted during the harvest and washing

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procedures.

In the 1976–1977 storage season, the inoculated and the noninoculated but wounded roots of cultivars 2B and D2 that had not been dipped in thiabendazole developed similar amounts of rot, although both had considerably more rot than the inoculated and the noninoculated but wounded roots of the two resistant breeding lines. There was no noninoculated but wounded treatment for line

TABLE 1. Yields of roots of two storage rot-susceptible cultivars (D2 and 2B) and two storage rot-resistant breeding lines (75P1 and 7326) of sugar beet

Cultivar or breeding line	Yield (t/ha)		
	1976	1977	Mean <sup>z</sup>
D2	41.0	63.0	52.0 a
2B	42.3	50.0	46.1 ab
75P1	29.9	47.7	38.7 b
7326	16.2	11.2	13.7 c

<sup>z</sup>Means followed by the same letter are not significantly different,  $P = 0.05$ , according to Duncan's multiple range test.

The breeding line 75P1 was a product of a single cycle of selection at Fargo for resistance to storage rot caused by *Phoma betae*. The original line was introduced from the USSR where it was selected for resistance to storage rot caused by *Botrytis cinerea*. The breeding line 7326 was developed by three cycles of selection for resistance to storage rot caused by *P. betae*. The original population was developed for resistance to crown rot caused by *Rhizoctonia solani*. Mono Hy D2 (D2) and American Crystal 2 hybrid B (2B) are commercial cultivars.

TABLE 2. Rotted tissue and sucrose content in two commercial sugar beet cultivars (2B and D2) and in two storage rot-resistant breeding lines (7326 and 75P1) during the 1976–1977 storage season<sup>y</sup>

Cultivar or breeding line	Treatment <sup>z</sup>	Rot (% w/w)	Sucrose content (% w/w)	
2B	No thiabendazole:			
	Inoculated	11.6 b	12.3 gh	
	Wounded only	11.6 b	11.8 h	
	None	5.9 d	12.8 d–g	
	Dipped in thiabendazole:			
	Inoculated	7.3 c	12.7 efg	
	Wounded only	6.2 cd	12.7 efg	
	None	5.4 de	13.1 b–g	
	D2	No thiabendazole:		
		Inoculated	13.2 a	12.8 d–g
Wounded only		11.9 b	12.4 fgh	
None		3.6 e	14.1 a	
Dipped in thiabendazole:				
Inoculated		3.2 ef	13.6 a–d	
Wounded only		2.5 f	13.4 a–e	
None		2.5 f	13.8 ab	
7326		No thiabendazole:		
		Inoculated	8.5 c	12.6 e–h
	Wounded only	...	...	
	None	3.5 e	13.7 abc	
75P1	No thiabendazole:			
	Inoculated	3.3 ef	13.2 b–f	
	Wounded only	3.5 e	12.9 c–g	
	None	1.1 f	13.0 b–g	

<sup>y</sup>After ~ 150 days of storage at 5 C, the rotted portions were excised and weighed, and sucrose content was measured by polarimeter. Within each column means followed by the same letter are not significantly different,  $P = 0.05$ , according to Duncan's multiple range test.

<sup>z</sup>Where indicated, roots were dipped in thiabendazole (1,500 µg/ml) and allowed to dry for 1 day before inoculation. Where indicated, roots were inoculated with *Phoma betae*, *Botrytis cinerea*, and *Penicillium claviforme*. Breeding lines as described in Table 1.

7326 because there were too few roots. The greatest amount of rot was accompanied by a decrease of the sucrose content in D2 and 7326, but not in 75P1 or 2B. The benefit of using thiabendazole was evident in the reduced rot and sucrose loss during storage. There was less rot and less sucrose loss in roots of D2 than in those of 2B when both were treated with thiabendazole.

The control of storage rot in roots of cultivar D2 with thiabendazole was comparable to the control obtained with the genetic resistance of 75P1 during the 1976–1977 storage period (Table 2). During the same period, the control of storage rot in 2B with thiabendazole was similar to that obtained with the genetic resistance of 7326. Control of storage rot with thiabendazole was slightly better than with genetic resistance during the 1977–1978 storage period, when the incidence of storage rot was lower than in the previous year (Table 3). In both years the genetic resistance of 75P1 and the application of thiabendazole to D2 controlled storage rot more effectively than did the genetic resistance of 7326 or the application of thiabendazole to 2B, respectively (Tables 2 and 3).

Cultivars 2B and D2 each averaged 2% rotted tissue during the 100-day storage period of 1977–1978 (Table 3). That rot was accompanied by a significant decrease in sucrose content and RWST. There were no significant changes when 2B was wounded only or when D2 was wounded only or not treated. Storage rot was reduced to a trace amount and no significant decreases in quality occurred when both cultivars were treated with thiabendazole.

Resistant breeding line 7326 developed more rot than did 75P1 when both were inoculated. Both breeding lines and cultivar 2B without thiabendazole treatment had similar amounts of RWST at harvest and after storage; however, the loss of RWST from the resistant roots during storage was not statistically significant, whereas that from roots of 2B and D2 not treated with thiabendazole was.

## DISCUSSION

The genetic resistance of the sugar beet breeding lines and the use of thiabendazole on the commercial cultivars satisfactorily reduced quality deterioration and storage rot caused by *P. betae*, *B. cinerea*, and *P. claviforme*.

*Phoma betae* is a seedborne pathogen of sugar beet and it also exists as an inactive pathogen within tissues of mature sugar beets that have not damped-off at the seedling stage of the disease cycle. The fungus becomes active and decays tissue of roots during storage (5,6). Thiabendazole may not control this phase of *Phoma* storage rot if the systemic capability of the fungicide is not extensive enough to contact the fungus embedded in the root tissues.

The differences in root yields between 1976 and 1977 were due to a difference in available moisture; drought conditions existed in 1976 and the first part of 1977. This difference also may account for the differences in susceptibility to storage rot. Roots are more susceptible to *Phoma* storage rot when grown under dry conditions (8). When the susceptible cultivars were grown with adequate moisture in the latter part of 1977, they suffered only 2% rot by weight after inoculation and storage for 100 days. The use of thiabendazole significantly reduced the amount of rot in both cultivars and significantly reduced the sucrose loss in the cultivar 2B. In 1975–1976, 1.77% of rot was found at a Moorhead, Minnesota factory (2). Thus, the severity of rot in the 1977–1978 experimental storage season approximated that in a commercial environment in 1975–1976. If thiabendazole had been used on the 288,000 tons of roots that were stored for more than 100 days at the Moorhead factory and had controlled rot as well as it did in our test, the savings in sucrose would have been about 4.4 kg/ton, or 1.3 million kg of sucrose. Our data suggest that even greater control of storage rot and savings in sucrose may be realized with the use of genetic resistance (Table 3).

The use of a fungicide, such as thiabendazole, to reduce sugar beet losses from storage rot is highly desirable and fills a need in the industry. Storage rot control is especially needed when roots are stored within protective structures (7). Here the humidity is high and the environment is conducive to rot (W. M. Bugbee, *personal*

TABLE 3. Rotted tissue, sucrose content, clear juice purity (CJP), recoverable white sugar per ton (RWST), and change in RWST in two commercial sugar beet cultivars (2B and D2) and in two storage rot-resistant breeding lines (7326 and 75P1) during the 1977-1978 storage season<sup>x</sup>

Cultivar or breeding line	Treatment <sup>y</sup>	Rot (% w/w)	Sucrose content (% w/w)	CJP (%)	RWST (kg)	Change in RWST during storage <sup>z</sup> (kg)
2B	No thiabendazole:					
	Inoculated	2.0 a	12.1 e	86.6 c-f	79 fgh	-17*
	Wounded only	0.1 d	13.0 b-e	86.2 def	83 d-h	-13
	None	0.0 d	12.4 de	87.4 b-f	82 e-h	-14
	At harvest	...	13.5 b	89.6 a-d	96 a-e	...
	Dipped in thiabendazole:					
	Inoculated	0.1 d	13.2 bcd	89.8 a-d	94 b-e	-6
	Wounded only	0.0 d	13.0 b-e	89.9 abc	93 b-e	-7
	None	0.0 d	13.4 bc	88.5 a-f	92 b-f	-8
	At harvest	...	13.6 b	91.1 a	100 abc	...
D2	No thiabendazole:					
	Inoculated	2.0 a	12.9 b-e	89.3 a-e	91 c-g	-18*
	Wounded only	0.0 d	13.6 b	90.2 abc	98 abc	-10
	None	0.0 d	13.7 b	90.3 abc	99 abc	-9
	At harvest	...	14.7 a	91.0 ab	108 a	...
D2	Dipped in thiabendazole:					
	Inoculated	0.02 d	13.4 bc	89.4 a-e	94 b-e	-10
	Wounded only	0.01 d	13.4 bc	89.8 a-d	96 a-d	-8
	None	0.0 d	13.5 b	89.7 a-d	96 a-d	-8
	At harvest	...	14.5 a	90.2 ac	105 ab	...
7326	No thiabendazole:					
	Inoculated	1.2 b	12.2 e	85.2 f	76 h	-8
	Wounded only	0.02 d	12.3 de	86.0 ef	78 gh	-5
	None	0.0 d	12.3 de	87.5 a-f	83 d-h	-4
	At harvest	...	12.5 cde	87.2 c-f	83 d-h	...
75P1	No thiabendazole:					
	Inoculated	0.6 c	13.1 bcd	88.0 a-f	89 c-h	-3
	Wounded only	0.0 d	13.4 bc	85.6 f	84 d-h	-8
	None	0.0 d	13.0 b-e	87.8 a-f	88 c-h	-4
	At harvest	...	13.6 b	87.9 a-f	92 b-f	...

<sup>x</sup> After about 100 days of storage at 5 C, the rotted portions were excised and weighed, and sucrose content was measured by polarimeter. Within each column, means followed by the same letter are not significantly different according to Duncan's multiple range test ( $P = 0.05$ ).

<sup>y</sup> Where indicated, roots were dipped in thiabendazole (1,500  $\mu\text{g/ml}$ ) and allowed to dry for 1 day before inoculation. Where indicated roots were inoculated with *Phoma betae*, *Botrytis cinerea*, and *Penicillium claviforme*.

<sup>z</sup> Symbols and abbreviations: asterisk (\*) indicates statistically significant decrease during storage according to Duncan's multiple range test,  $P = 0.05$ . CJP standing for clear juice purity, is  $100 \times (\text{sucrose content}) / (\text{dry substance of root extract})$  that has been clarified by CaO and filtration. RWST stands for recoverable white sugar per ton of roots calculated with an assumed factory loss of 0.3% and a molasses purity of 62.5%. Breeding lines as described in Table 1.

observation). Storage rot control with the use of resistant cultivars also should be a goal of sugar beet breeders, however, because past experience with other crops has shown such means to be more economical than chemical control. Efforts now are in progress to incorporate storage rot resistance into agronomically acceptable cultivars.

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