Breeding Sugar Beets for Resistance to the Cyst Nematode Heterodera Schachtii

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Received for publication July 27, 1964

Introduction

The cyst nematode *Heterodera schachtii* Schmidt causes a major disease of sugar beets in Europe and in the United States. In the United States, the cyst nematode has been severe chiefly in areas where sugar beets have been grown for many years. In some areas, however, notably in the Imperial Valley of California, sugar beets have been grown a relatively short time, yet the nematode disease is already serious and soil infestation is spreading. Probably initial nematode infestation resulted from cysts carried in particles of soil adhering to farm equipment which had been used in other areas where the soil was infested with sugar beet nematode. The pest was brought to this country from Europe, where it has been known for over 100 years, through the importation of sugar beet seed. In the early 1920's, the author found live nematodes in soil screened from imported sugar beet seed(1)³.

Crop rotation has been effective in keeping the population of nematodes down to a level that will permit one crop of beets to be grown successfully in infested soil once in 4 to 6 years. This is possible only if nonhost plants are grown in rotation with sugar beets. However, intervals of 4 to 6 years between sugar beet crops sometimes are not conducive to favorable economy of small farmers who depend upon sugar beets as a profitable cash crop.

Crop rotation, soil fumigation, and other measures have been recommended for control of the nematode; however, these are not fully satisfactory, and there is still scrious need for varieties that are tolerant of the pathogen. Breeding research for the development of varieties that are resistant to the nematode has been a major activity at the U. S. Agricultural Research Station, Salinas, California. The use of resistant varieties would

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² The author is indebted to Phyllis Emparan. formerly a member of the Crops Research Division, Agricultural Research Service. U. S. Department of Agriculture, for help with statistical analyses, and to C. L. Schneider. Pathologist, Crops Research Division, Agricultural Research Service. Logan, Utah, for isolation of the pathogenic fungi from the field soil used for screening tests.

⁸ Numbers in parentheses refer to literature cited.

reduce the damage caused by the nematode as well as permit growers to shorten the interval between crops of sugar beets in the rotation.

Material and Methods

Hundreds of thousands of plants have been examined in screening tests in an attempt to find resistance to the cyst nematode *Heterodera schachtii*. Material examined has consisted of commercial varieties such as the curly top-resistant varieties US 22, US 56/2, and US 33, and some of the Cercospora leafspot-resistant varieties. Also used were promising monogerm lines, plants from irradiated seeds, and crosses between sugar beets and *Beta maritima* L., as well as crosses between sugar beets and *Beta webbiana* Moq., which is immune to the attack of *H. schachtii*.

Screening tests in the greenhouse are accomplished by means of a technique developed at Salinas, in which seedling sugar beets are exposed to severe nematode attack and the most vigorous plants with fewest cysts are selected for future studies and further selections. The number of plants showing any degree of resistance is so small that it is necessary to examine large populations of sugar beets in order to obtain a few which show promise as breeding material. Many of these are eliminated later when tested under more extensive exposure to nematodes and other soilborne pathogens.

Viable cysts for use in screening breeding material for nematode resistance are collected from fields in which sugar beets have suffered severe damage from the pathogen. The soil is collected immediately after harvest of the infested crop, and by this means, freshly formed and highly viable cysts are obtained. The field soil is screened by means of a machine especially designed for the purpose. Three screens of different size mesh are mounted in a frame which is agitated by an eccentric. The clods and debris are carried to the end of the largest screen and returned to the field; the small soil particles and nematode cysts pass through the smallest screen and collect in a container. The mixture of nematode cysts and small particles of soil are placed in 50-gallon drums for use as inoculum in the evaluation of sugar beets for nematode resistance.

Cyst population in the screened field soil is determined by weighing 100-gram samples of inoculum from each drum. If each of several 100-gram samples contains 200 cysts filled with eggs the inoculum is considered adequate. Cysts in the inoculum, however, frequently exceed 200 per 100 grams of soil. Field soil frequently contains, in addition to nematode cysts, root-rotting

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fungi. *Rhizoctonia solani* Kuehn is the principal pathogenic fungus isolated from diseased seedlings grown in soil infested by the inoculum.

Greenhouse Test

Screening sugar beets for nematode resistance involves planting seeds in sterile sand, and when the seedlings reach the 2-leaf stage they are transplanted in greenhouse flats (Figure 1) containing a special mix of sterilized light sandy soil to which the nematode inoculum is added. The soil and inoculum are thoroughly mixed so that the cysts are uniformly distributed. After 6 or 8 weeks the plants are immersed in water and the adhering soil is carefully removed from the roots without detaching the female nematodes. The sugar beet rootlets are tneh examined



Figure 1.—Testing for resistance to *Heterodera schachtii*. Sugar beet seedlings are transplanted from sterile sand to flats of nematode infested soil. US 41 in 2 center rows; new breeding lines on left and right.

for the presence of female nematodes. The plants are classified into 6 categories (0 to 5 inclusive) according to the number of cysts present on the roots. The plants graded 0, 1, and 2, which have from none to 5 nematodes on the roots, are transferred to aluminum cylinders where they are exposed further to nematode attack before final selections for resistance are made. Plants carded. Frequently, the selected plants which were graded 0, 1, graded 4 or 5 are considered susceptible to nematode and disand 2 and further tested in aluminum cylinders are graded 4 or 5 when removed from the cylinders (Figure 2). Final selections for resistance to nematode are characterized by: a) not more than 3 nematode cysts on the roots, b) high vigor, and c) comparatively large root size.



Figure 2.—Females of *Heterodera schachtii* on roots of susceptible sugar beet grown in infested soil in aluminum cylinder.

After approximately 8 weeks, sugar beets in aluminum cylinders are examined, and the final selections are planted in 8-inch greenhouse pots and placed in a favorable location for growth. After the roots in greenhouse pots have attained approximately 1 inch in diameter, the plants are removed to a cold room, which is equipped with lights, and the temperature maintained at 42° F. The plants remain in the 42° temperature 90 to 120 days, depending upon the thermal induction requirement of the selection. They are then removed to the field or green-

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house for seed increase or hybridization. Selections for nematode resistance are again tested in field and greenhouse and a second cycle of selection and testing is completed. Selections for nematode resistance have also been made from commercial fields in the Salinas, California, area.

Crock Test

Crocks for the test (Figure 3) are partially filled with uniformly mixed fertile soil. Nematode cyst inoculum is added to the soil in half of the crocks. Each line of sugar beet to be evaluated is planted in a set of 10 crocks each of infested and noninfested soil. There are 3 such replications—or 30 crocks with nematode inoculum added and 30 crocks with no inoculum. The sugar beets are thinned to a single plant in each crock. At harvest, tops are removed and roots are weighed. Yields of roots are calculated on the individual root weight basis and each plant is considered a replication.



Figure 3.—Crock test of nematode-resistant lines of sugar beet developed in greenhouse screening program. One plant in each 3-gallon crock of nematode-infested soil or nematode-free soil.

Field Test

In the field test plot (Figure 4) at the U. S. Agricultural Research Station at Salinas, California, the soil is infested with sugar beet nematode and it also contains root-rotting fungi, principally *Rhizoctonia solani*. A high population of nematodes is maintained in the soil by means of frequent applications, with a fertilizer drill, of soil infested with sugar beet nematode. The



Figure 4.—General view of nematode resistance test, U. S. Agricultural Research Station, Salinas, California. The soil in this field is heavily infested with the cyst nematode *Heterodera schachtii* and root-rotting fungi.

contaminated soil is placed approximately 3 inches deep and close to the planted row of sugar beets. Selections to be tested for resistance to nematodes are planted in May, when soil temperatures are favorable for nematode activity and maximum damage occurs. The sugar beets are exposed to nematode attack at a very early stage of growth when they are highly susceptible to damage. The sugar beets are planted on beds and furrow irrigation is used. Plots of each selected line of sugar beet and of the commercial varieties used as checks are single row, 20 inches apart and 25 feet long, with 2 to 6 replications of each line in the test.

Root-rotting fungi play an important part in the damage attributed to nematodes. The punctures made by *Heterodera schachtii* undoubtedly facilitate penetration of pathogenic organisms into root tissue. Therefore, a variety of sugar beet that is resistant to a combination of pathogenic soilborne organisms and nematode is highly desirable. This phase of the work has received much attention in the breeding program.

Results

Greenhouse Test

Screening tests were made in the greenhouse under conditions of exposure to both nematodes and root-rotting fungi. By repeated selections and crosses, some lines have been developed which show remarkable resistance to a combination of root rot and cyst nematode (Figure 5); but much further work is necessary to develop lines of sugar beet that are fully resistant to the combination of nematodes and other soilborne pathogens under field conditions. Studies have indicated strongly that when some

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nematode-resistant lines are exposed to the combination they suffer less damage than the commercial variety US 41 used as check. It is necessary, however, to conduct further tests to determine whether these lines are resistant to the pathogenic fungi or whether nematode tolerance of the sugar beet gives more protection against the root-rotting organisms.



Figure 5.—Sugar beet seeds planted in greenhouse flats in soil heavily infested with the sugar beet nematode and containing root-rotting fungi. Commercial variety US 41 and a monogerm line, SL 9229, are highly susceptible. A selected line, 101-7, shows remarkable resistance.

Field Test

Table 1 shows the results of tests of 10 lines of sugar beet bred for resistance to the cyst nematode *Heterodera schachtii* and two commercial varieties, US 41 and US 75, used as checks. US 41 has been used as a check for several years in nematode tests,

Line or variety	Acre yield	Increase acre yield over check (US 41)	increase acre yield over commercial variety (US 75)	Plant per 100 ft row
	Tons	Tons	Tons	Number
033-1	19.95	10.45	11.98	120
SL060-3	19.59	10.09	11.62	121
856-1	18.71	9.21	10.74	119
150-1	18.10	8.60	10.13	118
C057-15	17.63	8.13	9.66	120
SL054-1	16.52	7.02	8.55	117
896	16.34	6.84	8.37	114
060	15.64	6.14	7.67	120
159-8	15.62	6.12	7.65	118
861-15	15.54	6.04	7.57	117
LSD 5%	5.07			
LSD 1%	6.70			

Table 1.-Root yield of resistant lines of sugar beet grown under severe exposure to cyst nematode and other soil-inhabiting pathogens. Field test, Salinas, California, 1962.

but US 75 was used for the first time in the 1962 tests. Sugar beets were planted in heavily infested soil as already described. Plots were replicated three times for each line and for each of the two commercial checks. Of the 10 nematode-resistant lines included in the test, 7 were significantly superior in yield of roots per acre to US 41 at the 1% level of significance, and 3 were significantly superior at the 5% level. All nematoderesistant lines tested were superior in yield per acre to US 75 at the 1% level of significance.

Crock Test

Table 2 shows results of 12 entries—US 41, used as check, and 11 lines of sugar beet which have been bred for resistance to the cyst nematode. The plants were grown in 3-gallon crocks, as described under "Methods". After the tops were removed, each individual root was weighed and its weight recorded in grams. There were 10 crocks in each of three replications, or a total of 30 beets of each line and 30 beets of US 41. Results are therefore averages of 30 roots for each entry in the test. Three 10-beet samples were taken for sucrose determination. Table 2, column 2, shows weights of roots grown in noninfested soil, and column 4 shows weight of roots grown in infested soil. The percent loss in weight due to nematodes was calculated on the basis of difference between weight of roots grown in noninfested soil and those grown in infested soil. The loss in weight

	Noninfested		Infested		
Variety or line	Average weight per beet	Sucrose	Average weight per beet	Sucrose	Weight loss due to nematode
	Grams	Percent	Grams	Percent	Percent
019	1381.9	13.7	1367.9	13.1	1.0
062-11	1306.3	13.7	1287.0	12.1	1.5
033-1	1198.2	13.0	1127.2	12.1	5.9
1089G	1410.6	13.6	1263.9	12.7	10.4
050-6	1419.8	12.5	1248.7	12.0	12.1
U074	1348.8	14.4	1109.6	13.9	17.7
801-7	1209.9	14.7	991.8	13.4	18.0
1033-1	1492.2	12.5	1186.6	12.1	20.5
057-15	1591.1	14.5	1269.4	13.1	20.2
028	1521.0	14.7	1100.2	13.7	27.8
857-3	1572.2	15.0	1125.6	14.5	28.4
US 41	1273.0	14.0	886.5	13.9	30.4
Average % sucrose		13.86		13.05	
Reduction in sucrose due to nematodes				.81	
LSD 5%	224.9				
LSD 1%	295.6				

Table 2.-Root yield and sucrose percentage of resistant line; of sugar beet grown in crocks containing nematode-infested soil or nematode-free soil. Salinas, California, 1962.

varies from 1% in line 019 to 30.4% in US 41. All lines tested were higher in yield of roots than US 41. There was also a reduction in sucrose percentage due to nematode in all lines tested, with an average of .81 of a percentage point for the 11 entries.

Summary

Seedling sugar beets were planted in nematode infested soil in flats in the greenhouse and later screened for nematode intestation. Plants showing the least number of cysts were retained and planted singly in infested soil in aluminum cylinders for a second exposure to nematodcs. The best of these plants were retained for seed production. Progenies were tested in flats, cylinders, singly in 3-gallon crocks, and in replicated field tests where further selections for resistance were made. Lines which show promise of appreciable nematode resistance have been obtained from the curly top-resistant varieties US 22, US 56/2, and US 33, and from some of the Ccrcospora leaf-spotresistant lines. Some of these selections have shown resistance to a combination of nematode and Rhizoctonia root rot.

Literature Cited

(1) Farmers' Bulletin 1514, p. 6, 1926.