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Host Suitability of Rapeseed for *Heterodera schachtii*[1]

Eric L. Nielsen,[2] David D. Baltensperger,[2] Eric D. Kerr,[3] Charlie L. Rife[4]

Abstract: Because rapeseed, especially canola, has the potential to be grown in rotation with sugarbeet in the north-central region of the United States, this study was initiated to assess its susceptibility to infection by *Heterodera schachtii* and to develop a screening method for *Brassica* germplasm. Existing methodology was adapted for growing *Brassica juncea, B. napus, B. rapa, Brassica* hybrids, and sugarbeet, *Beta vulgaris,* in *H. schachtii*-infested soil to count the females that developed on the roots. Cysts on sugarbeet contained a mean of 130 eggs compared with 240 for *B. napus,* lowest for the *Brassica.* Viability of eggs produced was assessed in soil planted with *Brassica* and sugarbeet and infested with with 0, 100, 1,000, 3,000, and 5,000 eggs to count resulting females and cysts. Number of females (y) was related linearly to infestation rate (x) by the regression equations $y = 2.82 + 0.07(x)$ for the *Brassica* lines ($R^2 = 0.79$; $P < 0.001$) and $y = 0.43 + 0.04(x)$ for sugarbeet ($R^2 = 0.69$; $P < 0.007$). These data indicated the potential for *H. schachtii* population increase if the two crops are used in rotation. All of the 111 germplasm lines tested were susceptible. The methodology developed during this research would benefit attempts to develop rapeseed cultivars resistant to *H. schachtii.*

Key words: Beta vulgaris, Brassica hybrid, *Brassica juncea, Brassica napus, Brassica rapa,* canola, *Heterodera schachtii,* rapeseed, resistance screening, sugarbeet, sugarbeet cyst nematode, susceptibility.

Heterodera schachtii Schmidt, 1871, the sugarbeet cyst nematode, has been a negative factor in sugarbeet production for many years. It is reported in most sugarbeet (*Beta vulgaris* L.)-growing regions of the United States (Nematode Geographical Distribution Committee of the Society of Nematologists, 1984) and, depending on nematode densities, economic losses vary from a trace to 70% (Altman and Thomason, 1971). During his surveys in the 1920s, Thorne (1961) noted rapid distribution of sugarbeet cyst nematodes in the sugarbeet production areas of the western United States due to poor management practices.

Control practices for *H. schachtii* in sugarbeet production involve crop rotation, strict sanitation procedures, early planting, and chemical nematicides. In highly infested fields, yields are often doubled by nematicide treatments, and yield increases have averaged 11 to 15 tons/ha following soil fumigation treatments compared to untreated soil (Altman and Thomason, 1971). Trap crops of radish and white mustard, which stimulate hatching but depress reproduction, have been developed and used in Europe to reduce *H. schachtii* populations. Research to evaluate the effectiveness of this control method has been performed in the United States (Gardner and Caswell-Chenn, 1993; Koch, et al. 1998). Research has been conducted in Europe to develop hybrid species of oil-seed rape with resistance to *H. schachtii* by making crosses with genotypes of related white mustard and fodder radish species that are resistant (Lelivelt et al. 1993a; Lelivelt et al. 1993b). Crop rotation can be an effective control practice when properly managed. *Heterodera schachtii* eggs may have a potential life span of 10 years, with the majority hatching within the first 3 to 5 years (Jones, 1956). Once hatched, the second-stage juveniles (J2) have a relatively short time in which to find a crop or weed host (Roberts and Thomas, 1981).

In recent years some sugarbeet producers in the north-central region of the United States have planted rapeseed (*Brassica* species L.), especially canola, as an alternative rotation crop. This study was initiated to assess the suitability of *Brassica* species with potential for canola-quality oils as hosts for *H. schachtii* and investigate methodologies for rapidly screening new rapeseed lines for resistance.

Materials and Methods

The experiment was conducted in two phases. In the initial phase, soil from a field highly infested with *H. schachtii* was collected and used to fill two sizes of 'Conetainers' (Steuewe and Sons, Inc., Corvallis, OR), 3.8 × 20.9 cm and 3.8 × 13.9 cm. The nematode population in the soil averaged 32 eggs and J2/cm^3 soil, as determined from cysts extracted from the soil by a modified version of the technique developed by Caswell et al., (1985) and crushed in a Tenbroeck Pyrex model 7726 tissue homogenizer (Corning, Inc., Science Products Division, Corning, NY) to free the eggs to be counted at x60 magnification. The soil also contained an undetermined quantity of *Nacobbus abberans* (Thorne, 1935) Thorne and Allen, 1994.

A diverse selection of winter and spring types of *Brassica* germplasm was represented in the experiment (Table 1). Spring types included four hybrids, three *B. juncea* experimental lines, 12 *B. napus* cultivars, and 10 experimental lines. Winter types included five *B. rapa* experimental lines, one cultivar, 14 *B. napus* cultivars, and 62 experimental lines. Sugarbeet, *Beta vulgaris* cy. 'Monohikari', was grown as a standard control.

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[2] Research Assistant and Professor, respectively, Department of Agronomy, University of Nebraska Panhandle Research and Extension Center, 4502 Ave. I, Scottsbluff, NE 69361.

[3] Professor Emeritus, Department of Plant Pathology, University of Nebraska Panhandle Research and Extension Center, Scottsbluff, NE 69361.

[4] Research Assistant Professor, Department of Agronomy, Kansas State University, Manhattan, KS 66506.

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E-mail: dbaltensperger1@unl.edu

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TABLE 1. Germplasm tested and mean scores of *Heterodera schachtii* females from four replications of each line and averaged over species.

Germplasm	Species	Habit	Germplasm mean[a]	Species mean
KSR925	*Brassica rapa*	winter	14	
KSR903	*B. rapa*	winter	13	
KSR912	*B. rapa*	winter	13	
KSR943	*B. rapa*	winter	13	
Debut	*B. rapa*	winter	12	
KSR953	*B. rapa*	winter	12	
				13
KS6120	*Brassica napus*	winter	15	
KSM3-1-120	*B. napus*	winter	15	
Pendleton	*B. napus*	winter	15	
Rapier	*B. napus*	winter	15	
Winfield	*B. napus*	winter	15	
KS8114	*B. napus*	winter	15	
KS8137	*b. napus*	winter	15	
KS8263	*B. napus*	winter	15	
KS8339	*B. napus*	winter	15	
KS8343	*B. napus*	winter	15	
KS8346	*B. napus*	winter	15	
KS8357	*B. napus*	winter	15	
KS8361	*B. napus*	winter	15	
KS8367	*B. napus*	winter	15	
KS7013	*B. napus*	winter	15	
KS7017	*B. napus*	winter	15	
KS7123	*B. napus*	winter	15	
KS7284	*B. napus*	winter	15	
KS7369	*B. napus*	winter	15	
KS7412	*B. napus*	winter	15	
KS7574	*B. napus*	winter	15	
KS7667	*B. napus*	winter	15	
KSB0008	*B. napus*	winter	14	
KSM3-1-124	*B. napus*	winter	14	
Olsen	*B. napus*	winter	14	
Wichita	*B. napus*	winter	14	
WW1089	*B. napus*	winter	14	
KS8071	*B. napus*	winter	14	
KS8202	*B. napus*	winter	14	
KS8313	*B. napus*	winter	14	
KS8369	*B. napus*	winter	14	
KS8381	*B. napus*	winter	14	
KS7007	*B. napus*	winter	14	
KS7513	*B. napus*	winter	14	
KSW001	*B. napus*	winter	14	
ARC91022-59L	*B. napus*	winter	13	
Ceres	*B. napus*	winter	13	
KS3203	*B. napus*	winter	13	
Plainsman	*B. napus*	winter	13	
KS8279	*B. napus*	winter	13	
KS8334	*B. napus*	winter	13	
KS8372	*B. napus*	winter	13	
KS8063	*B. napus*	winter	13	
KS8130	*B. napus*	winter	13	
KS7083	*B. napus*	winter	13	
KS7174	*B. napus*	winter	13	
KS7340	*B. napus*	winter	13	
KS7566	*B. napus*	winter	13	
KS7703	*B. napus*	winter	13	
KS7740	*B. napus*	winter	13	
KSC004	*B. napus*	winter	13	
Bridger	*B. napus*	winter	12	
Casino	*B. napus*	winter	12	
KS1701	*B. napus*	winter	12	
KS8094	*B. napus*	winter	12	
KS8118	*B. napus*	winter	12	
KS8173	*B. napus*	winter	12	
KS8281	*B. napus*	winter	12	
KS8288	*B. napus*	winter	12	
KS8208	*B. napus*	winter	12	
KS7448	*B. napus*	winter	12	
KS7489	*B. napus*	winter	12	
KS7688	*B. napus*	winter	12	
KS7727	*B. napus*	winter	12	
Arctic	*B. napus*	winter	11	
Ericka	*B. napus*	winter	11	
Inka	*B. napus*	winter	11	
Jetton	*B. napus*	winter	11	
KS8189	*B. napus*	winter	11	
KS8284	*B. napus*	winter	11	
KS8309	*B. napus*	winter	11	
KS7756	*B. napus*	winter	11	
KS8326	*B. napus*	winter	10	
KS7534	*B. napus*	winter	10	
KS7210	*B. napus*	winter	9	
KS7473	*B. napus*	winter	8	
Cyclone	*B. napus*	spring	15	
Norseman	*B. napus*	spring	15	
93.SN.195.9.1	*B. napus*	spring	15	
93.SN.21.8.3	*B. napus*	spring	15	
93.SN.545.8.2	*B. napus*	spring	14	
Flint	*B. napus*	spring	13	
Impulse	*B. napus*	spring	13	
Legend	*B. napus*	spring	13	
Sunrise	*B. napus*	spring	13	
G96202	*B. napus*	spring	13	
G97097A	*B. napus*	spring	13	
93.SN.195.10.3	*B. napus*	spring	13	
Brigade	*B. napus*	spring	12	
Westar	*B. napus*	spring	12	
93.SN.545.5.6	*B. napus*	spring	12	
Bingo	*B. napus*	spring	11	
Quantum	*B. napus*	spring	11	
A112	*B. napus*	spring	10	
Jewel	*B. napus*	spring	10	
CL2078	*B. napus*	spring	10	
RPX06.7.5.M4	*B. napus*	spring	10	
Crown	*B. napus*	spring	8	
				13
92X2914512	*Brassica* hybrid-spp. unknown	spring	14	
93BC.21622	*Brassica* hybrid-spp. unknown	spring	12	
				13
92.BJ.13.B.5	*Brassica juncea*	spring	14	
92.BJ.26.B.2	*B. juncea*	spring	13	
ZEM 1	*B. juncea*	spring	13	
				13
CI3	*B. juncea* × *B. napus* hybrid	spring	12	
CII3	*B. juncea* × *B. napus* hybrid	spring	12	
				12
Monohikari, sugarbeet	*Beta vulgaris*		8	
				8
Grand mean			13	12
LSD (0.05)			NS	3.55

[a] Counts were discontinued at 15 adult females and cysts. This level was considered to be high level of infection. Differences in germplasm means were not significant at a level ($P > 0.71$).

The experiment was conducted in a greenhouse during late fall and winter. Four replications of 112 entries each, with two replications grown in the 3.8 × 20.9-cm cones and two grown in 3.8 × 13.9-cm cones, were evaluated for females after 5 weeks. Individual plants were removed from the cones, the roots carefully washed of soil with a gentle spray of water, and both viewed at × 10 to ×30 magnification with dissecting microscopes. For the purpose of this experiment counts of 15 or more females were considered high levels of infection, so females were counted and scored from 0 to 15 for statistical analysis. Roots also were inspected for response to infection by *N. abberans,* and galls were counted and dissected for verification. Data for *H. schachtii* were analyzed by means of the SAS PROC GLM (SAS Institute Inc., Cary, NC) with species as whole plots and germplasm as subplots.

Females were collected from a subset of six *Brassica* experimental lines and six cultivars, as well as *Beta vulgaris,* and examined in more detail. Females from each plant were ground in a tissue homogenizer to free the eggs, which were then counted under ×30 magnification to determine the average number of eggs per female.

The second phase of the experiment was designed to test the viability of the eggs. Roots of remaining plants were washed and all females were collected by the modified version of the technique developed by Caswell et al. (1985). The collected females were ground in a tissue homogenizer, and the freed eggs were placed in 500 eggs/cm^3 water. The eggs were used to infest heat-treated, uncontaminated soil in 3.8 × 20.9-cm 'Cone-tainers' planted with two replications of 10 of the *Brassica* lines and sugarbeet.

Disposable plastic syringes with 14-gauge × 3/75-cm needles were used to infest the soil. The eggs were maintained in a constant state of suspension with a magnetic stirrer. The stirrer was stopped only long enough to fill a syringe to minimize potential variance caused by fluid flow. Injection was as near roots as possible without damage to the plant. Each replication included infestation levels of 5,000 eggs (10-ml suspension), 3,000 eggs (6-ml suspension), 1,000 eggs (2-ml suspension), 100 eggs (1-ml suspension diluted to 100 eggs/cm^3), and a control without eggs. After 34 days the roots were washed and the females were counted. Data were analyzed with SAS contrast statements in PROC GLM and PROC REG to calculate coefficients of linear effects. Infestation levels were inspected for linear, quadratic, and cubic effects.

Results

None of the selections tested displayed resistance to *H. schachtii.* More females developed on roots of plants in the larger 'Cone-tainers' ($P < 0.001$), probably due to the additional soil volume, which allowed increased root development and increased the inoculum level per plant. There was no difference ($P > 0.71$) among the varieties tested and no difference ($P > 0.13$) among species (Table 1). Sugarbeet had fewer females ($P < 0.05$) as well as small root systems compared to the extensively developed rapeseed root systems, which ultimately provided more opportunity for infection.

Nacobbus abberans, which infected several sugarbeet roots, averaged fewer than one per plant. Although present, they had little effect on root systems so no interaction with *H. schachtii* was expected. No *N. abberans* galls were observed on any of the *Brassica* roots, indicating it may not be a suitable host for this particular species.

Females from sugarbeet roots contained a mean of 130 eggs compared with 240 for *B. napus,* the lowest among the *Brassica* species (Table 2). Although eggs per female were lower on sugarbeet than *Brassica* lines ($P < 0.01$), there were no differences ($P > 0.29$) among lines within species of the *Brassica* subset tested.

When eggs from this phase were used to infest soil at various levels, viability of the eggs was demonstrated by the production of large numbers of females. Sugarbeet produced lower numbers of females ($P < 0.04$) at all levels of infestation. Because there was no difference ($P > 0.06$) among the *Brassica* species, they were combined for regression analysis in which only the linear effect was significant for infestation levels (Fig. 1).

Discussion

Sugarbeet had lower numbers of females when grown in naturally infested soil and under all artificial infestation levels compared with any of the *Brassica* lines. It also produced fewer eggs per female than any of the *Brassica* lines. Because this germplasm represents a wide range of the canola types used in current breeding programs, this indicates that sugarbeet cyst nema-

Table 2. Mean number of *Heterodera schachtii* eggs per female averaged over two replications of selected subset.

Line	Species	Germplasm mean	Species mean
Brigade	*Brassica napus*	390	
Wichita	*B. napus*	260	
Plainsman	*B. napus*	210	
Legend	*B. napus*	190	
Ceres	*B. napus*	170	240
92.BJ.13.B.5	*Brassica juncea*	370	
92.BJ.26.B.2	*B. juncea*	290	330
Debut	*Brassica rapa*	270	
KSR953	*B. rapa*	250	
KSR925	*B. rapa*	220	250
CI3	*B. juncea* × *B. napus* hybrid	260	
CII3	*B. juncea* × *B. napus* hybrid	240	250
	Mean	260	270
			NS[a]
Monohikari	*Beta vulgaris*	130	

[a] Differences between lines and *Brassica* species are not significant.

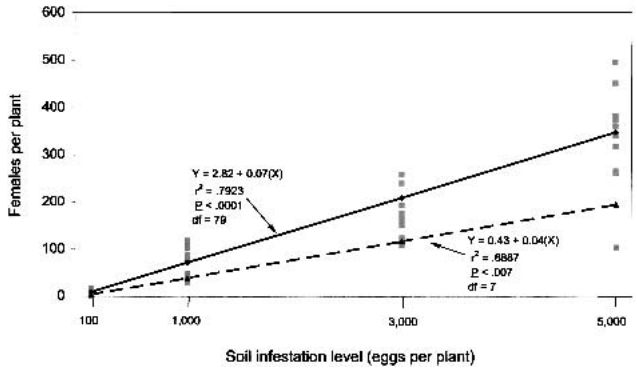

FIG. 1. Relationship between soil infestation level of *Heterodera schactii* (x) and number of females produced (y) on *Brassica* lines (—) and *Beta vulgaris* (- - -).

todes could present major problems in canola production. Furthermore, canola may increase problems with *H. schachtii* on sugarbeet when included with it in the same rotation. The germplasm evaluated represents a small proportion of the total rapeseed and related species germplasm (GRIN, 2001). Although none of the experimental lines or cultivars in the test proved to be resistant to *H. schachtii,* this experiment established a relatively fast and efficient procedure to evaluate rapeseed resistance to *H. schachtii* that may be of value in transferring resistance from poorly adapted types, such as those being developed by Lelivelt et al. (1993a,b), to well-adapted canola quality types.

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