

# Pathological Differences in *Heterodera schachtii* Populations<sup>1</sup>

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**Abstract:** Five populations of *Heterodera schachtii* Schm. from Oregon, Idaho, and Utah did not differ significantly in seedling penetration and rate of emergence and virulence. Another Utah *H. schachtii* population (Utah 2), however, differed from these five populations in all of the above-mentioned characteristics. More *H. schachtii* larvae of the Utah 2 population than the other populations penetrated sugarbeet seedlings at 10, 15, 20, and 25 C. Root and top weights of sugarbeet plants were significantly less when roots were parasitized by the Utah 2 population than when they were parasitized by larvae of the other nematode populations under similar experimental conditions. Also, the period of larval emergence was shorter in the Utah 2 population than in any of the other *H. schachtii* populations. **Key words:** sugarbeet cyst nematode, sugarbeet, *Beta vulgaris*, virulence, soil temperature, penetration, hatching.

The potential value to the sugarbeet industry of sugarbeets resistant to *Heterodera schachtii* Schm. is well documented (5,17, 18,20,21). However, an agronomically suitable variety of sugarbeet with resistance to *H. schachtii* has not yet been developed (15). Without plant resistance, the grower must rely on crop rotation or chemicals for nematode control. Aldicarb (2-methyl-2[methylthio] propiona[dehyde] oxime) and 1-3, dichloropropene have usually given excellent control of the sugarbeet nematode (4,6). But these have sometimes failed, even when used as prescribed. In a northern Utah sugarbeet field, aldicarb failed to give satisfactory results even though recommendations relating to rate, placement, soil temperature, soil moisture, and population density (3,4) were followed. The existence of an unusually virulent physiological strain of *H. schachtii* was suspected.

A study was initiated to determine whether the Utah population differed in virulence from five other geographically separated populations in Idaho, Oregon, and Utah.

## MATERIALS AND METHODS

The Idaho *H. schachtii* populations were from Burley (ID1) and Nampa (ID2); the Oregon populations were from Ontario (OR1 and OR2); the Utah populations were from Lewiston (UT1) and Ogden (UT2). Each population was cultured separately on sugarbeet. Cysts containing viable eggs were surface sterilized with 0.5%

sodium hypochlorite solution, rinsed in distilled water, and second stage larvae were hatched from eggs in a ZnCl<sub>2</sub> solution.

**Root penetration:** Fourteen-day-old sugarbeet seedlings (Tasco AH3) were transplanted into bromomethane fumigated sandy loam soil in 12.5-cm plastic containers and inoculated with 250 *H. schachtii* larvae/plant. Plants, four/container, were replicated (five containers/nematode population) and grown at constant temperatures of 10, 15, 20, and 25 C in water baths. After 14 d of growth, plants were harvested; roots were stained in an acid fuschin-lactophenol solution, and root penetration by *H. schachtii* larvae determined.

Another experiment involved inoculating 14-d-old sugarbeet seedlings (four plants/container) with three cysts from the Idaho 2, Utah 1, and Utah 2 populations. Inoculum was obtained from sugarbeet plants that had been inoculated with larvae and grown for 42 d at 25 C. Replicates (five containers of plants/population) were grown at each temperature (10, 15, 20, and 25 C) for 14 d and root penetration determined.

**Nematode virulence:** Differences in virulence of the six population isolates were studied. Fourteen-day-old sugarbeet was transplanted into 12.5-cm plastic containers of soil (one plant/container) and inoculated with 500 *H. schachtii* larvae. Plants, replicated five times, were grown at each of the four different temperatures (10, 15, 20, and 25 C) for 42 d and root and top weights determined.

Another experiment involved inoculating 14-d-old sugarbeet seedlings with sugarbeet nematode cysts (three cysts/plant) of the Idaho 2 and Utah 1 and 2 populations. These cysts were obtained from sugarbeet

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plants that had been inoculated with larvae and grown for 42 d at 25 C. Plants were replicated five times/population, grown at each temperature (10, 15, 20, and 25 C), and root weights determined.

**Larval emergence from cysts:** An experiment was made to determine possible differences in the rate of larval emergence from cysts of the six nematode populations. Fourteen-day-old sugarbeet seedlings were transplanted into 12.5-cm plastic containers and inoculated with 500 *H. schachtii* larvae/plant. Plants, replicated five times/population, were grown at 25 C for 42 d. Twenty cysts were hand picked at random from each plant, larvae hatched in a ZnCl<sub>2</sub> solution at 25 C, counted daily, and new ZnCl<sub>2</sub> solution added daily for a period of 21 d. A similar experiment involved growing plants for 70 d after inoculation before determining differences in larval emergence from cysts.

A final experiment involved inoculating 14-d-old seedlings with cysts (three/plant) of the Idaho 1 and Utah 1 and 2 populations. These cysts were obtained from plants inoculated with larvae and grown for 42 d at 25 C. After 42 d growth at 25 C, larval emergence was determined in a manner similar to that described above.

**RESULTS AND DISCUSSION**

Differences among the Idaho, Oregon, and Utah 1 populations in seedling penetration, virulence, rate of reproduction, and rate of hatching were not significant. However, the Utah 2 population was physiologically different from the other populations in all above-mentioned characteristics.

**Root penetration:** Significantly more *H. schachtii* larvae of the Utah 2 population penetrated sugarbeet seedlings than did larvae of the other five populations ( $P = 0.01$ ) in 14 d at all temperatures (10, 15, 20, and 25 C) (Fig. 1). Penetration increased with increasing soil temperature in all *H. schachtii* populations, but penetration of sugarbeet seedlings by the Utah 2 population was as great at 20 C and greater at 25 C than that of any other *H. schachtii* populations at 25 C ( $P = 0.01$ ).

When 14-d-old sugarbeet seedlings were inoculated with three cysts/plant, signif-

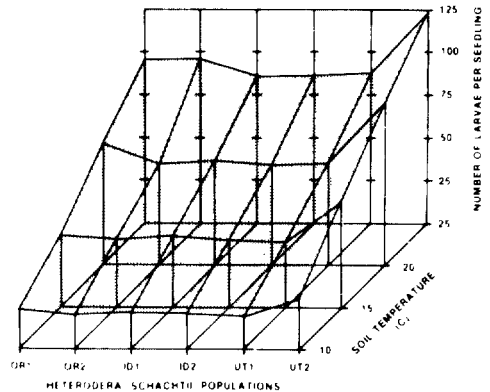


Fig. 1. Differences in penetration of 14-day-old sugarbeet seedling roots by larvae from 6 populations of *Heterodera schachtii* from Oregon (OR1 and 2), Idaho (ID1 and 2), and Utah (UT1 and 2). Each seedling was inoculated with 250 larvae and grown for 14 days at 10, 15, 20, or 25 C ( $P, 0.01 = 22$ ).

icantly more *H. schachtii* larvae from the Utah 2 population than from the Idaho 2 and Utah 1 populations penetrated seedlings ( $P = 0.01$ ) at all temperatures (Table 1).

**Nematode virulence:** The Utah 2 population was more virulent than the other populations; root and top weights of plants parasitized by the Utah 2 populations inoculated as 14-d-old seedlings with 500 larvae and grown at soil temperatures of 10–25 C were significantly lower than that from plants grown at the same soil temperature and parasitized by the other populations ( $P = 0.01$ ) (Figs. 2, 3). Similar results were

Table 1. Penetration of sugarbeet seedling roots by larvae from three *Heterodera schachtii* populations.\*

Soil temp. (C)	Number of larvae/seedling root in indicated nematode populations†			LSD ( $P = 0.01$ )
	ID2	UT1	UT2	
10	12	10	34	7
15	33	36	73	11
20	56	52	102	17
25	72	74	156	13
LSD ( $P = 0.01$ )	13			

\*Fourteen-day-old sugarbeet seedlings were inoculated with three cysts (cultured on sugarbeet at 25 C for 42 d).

†Penetration determined 14 d after inoculation.

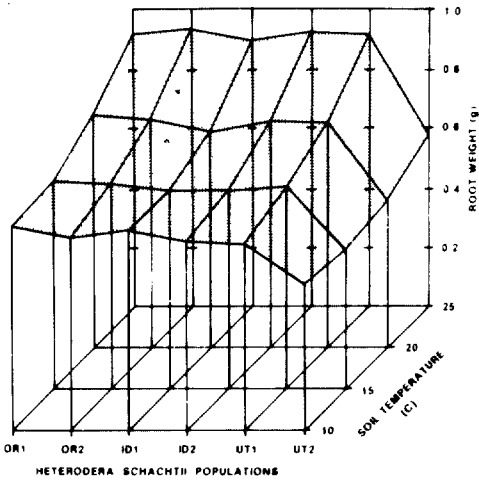


Fig. 2. Effect of 6 populations of *Heterodera schachtii* from Oregon (OR1 and 2), Idaho (ID1 and 2), and Utah (UT1 and 2) on sugarbeet root growth. Fourteen-day-old seedlings were inoculated with 500 larvae/plant and grown for 42 days at 10, 15, 20, or 25 C ( $P, 0.01 = 0.15$ ).

observed when 14-d-old seedlings were inoculated with cysts of Idaho 1 and Utah 1 and 2 populations (Fig. 4). At 10, 15, 20, and 25 C, root growth of the plant inoculated with cysts from the Utah 2 population was 60, 58, 58, and 48%, respectively, of the root growth of plants inoculated with Idaho 2 cysts and 62, 56, 60, and 55%, respectively, of that of plants inoculated with Utah 1 cysts. Likewise, it was 80, 67, 67, and

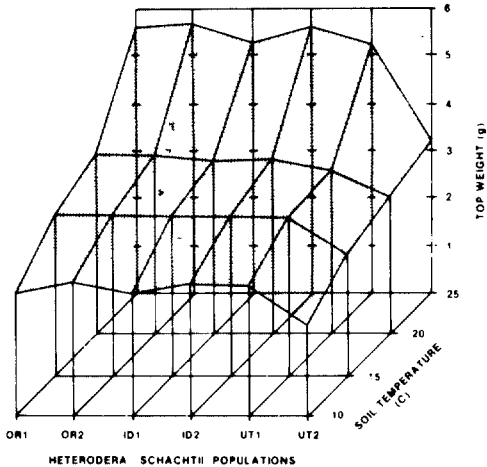


Fig. 3. Effect of 6 populations of *Heterodera schachtii* from Oregon (OR1 and 2), Idaho (ID1 and 2), and Utah (UT1 and 2) on sugarbeet top growth. Fourteen-day-old seedlings were inoculated with 500 larvae/plant and grown for 42 days at 10, 15, 20, or 25 C ( $P, 0.01 = 0.64$ ).

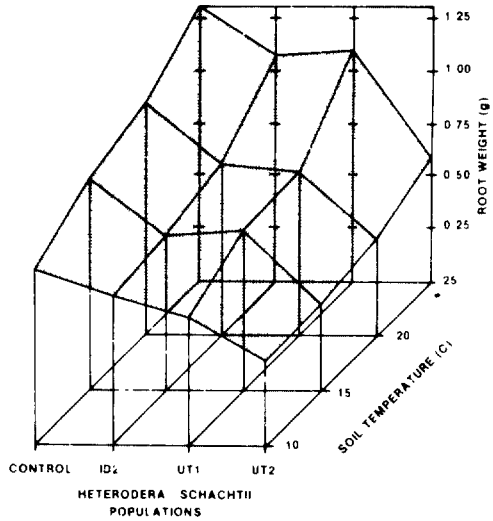


Fig. 4. Effect of 3 populations of *Heterodera schachtii* from Idaho (ID2), and Utah (UT1 and 2) on sugarbeet root growth. Fourteen-day-old seedlings were inoculated with three cysts/plant and grown for 42 days at 10, 15, 20, and 25 C. Inoculum (cysts) were produced on plants inoculated with larvae and grown for 42 days at 25 C. ( $P, 0.01 = 0.22$ ).

64% of the root growth of the plants inoculated with Utah 1 larvae and 76, 69, 68, and 64% of that of plants inoculated with Idaho 2 larvae at 10, 15, 20, and 25 C, respectively.

**Larval emergence from cysts:** The larval emergence was shorter and was significantly higher for the Utah 2 population than for the other populations ( $P = 0.05$ ) (Table 2). After 7 d, 152 (76%) of the viable eggs from this population hatched; next highest was the 36 (22%) for the Idaho 1 populations. The low number of larvae/cyst and the apparent great variation in number of eggs/female were due to the presence of undeveloped as well as viable eggs in the cysts. This was emphasized by the fact that there were no differences in the total number of larvae hatching from any of the populations after 21 d from cysts cultured for 70 d. However, there were significant differences in the rate of emergence of larvae from cysts cultivated on sugarbeet for 70 d (Table 3).

The rate of larvae emergence from cyst reproduction on plants inoculated with cysts (Table 4) were similar to those for nematodes on plants inoculated with larvae (Table 2). After 7, 14, and 21 d of incubation, 57, 19, and 14%, respectively, of the eggs hatched in the Utah 2 population on plants inoculated with cysts; the comparable

Table 2. Emergence of larvae from cysts of six populations of *Heterodera schachtii* cultured on sugarbeet after 42 d.\*

Days of incubation†	Number of larvae emerged/cyst and nematode populations						LSD <i>P</i> = 0.05
	ORI	OR2	ID1	ID2	UT1	UT2	
7	32	24	36	34	26	152	26
14	73	69	52	43	45	30	11
21	62	69	77	97	91	18	12
Total	167	162	165	174	162	200	32
LSD ( <i>P</i> = 0.05)	24						

\*Cysts collected from plants inoculated with 500 larvae and grown at 25 C.

†Cysts (100/population) maintained at 25 C during larval emergence.

Table 3. Emergence of larvae from cysts of six populations of *Heterodera schachtii* cultured on sugarbeet for 70 d.\*

Days of incubation†	Number of larvae emerged/cyst in indicated nematode populations						LSD <i>P</i> = 0.05
	ORI	OR2	ID1	ID2	UT1	UT2	
7	102	99	97	119	103	201	22
14	82	69	94	81	92	46	41
21	75	89	72	77	77	30	18
Total	259	257	263	277	272	276	24
LSD ( <i>P</i> = 0.05)	19						

\*Cysts collected from plants inoculated with 500 larvae and grown at 25 C.

†Cysts (100/population) maintained at 25 C during larval emergence.

figures were 15, 35, and 50% for the Idaho 1 population and 13, 37, and 50% for the Utah 1 population. Likewise, the hatching percentages for eggs on plants inoculated with larvae were 76, 15, and 9% for the Utah 2; 22, 32, and 47 for the Idaho 1;

Table 4. Emergence of larvae from cysts of three populations of *Heterodera schachtii* cultured on sugarbeet for 42 d.\*

Days of incubation†	Numbers of larvae/cyst in indicated nematode populations			LSD ( <i>P</i> = 0.01)
	ID2	UT1	UT2	
7	29	26	186	19
14	69	76	53	18
21	98	101	39	16
Total	196	203	278	13
LSD ( <i>P</i> = 0.01)	23			

\*Cysts collected from plants inoculated with three cysts/plant and grown at 25 C.

†Cysts (100/population) maintained at 25 C during larval emergence.

and 16, 28, and 50 for the Utah 1 population after 7, 14, and 21 days of incubation, respectively.

When penetration of sugarbeet seedlings is compared after inoculation with 250 larvae (Fig. 1) and inoculation with three cysts (Table 1) of the Utah 2 population, significantly more larvae emerging from cysts penetrate sugarbeet seedlings. This difference was apparently a result of more than 250 larvae hatching from the three cysts. With the Idaho 2 and Utah 1 populations, however, the results of the penetration and pathogenicity studies followed the reverse trend: larvae penetration was greater and plant growth was less after larval inoculation than after inoculation with cysts. This difference may indicate a possible shorter reproductive period in the Utah 2 population. This, plus the more rapid larval emergence that would result in an earlier root penetration by a greater number of larvae, resulted in a more severe pathological reaction to the Utah 2 population than to the other populations.

From this study it appears that physiological differences in virulence is apparently due to a faster larval penetration and shorter period of larval emergence that also contributes to an earlier root penetration. A reduced generation time may also be involved, but this is a factor requiring further investigation. Such a phenomenon should be considered when sugarbeet yields after the use of cultural or chemical control practices are less than expected. These factors will influence the response of the host-parasite relationship to temperature, the nematode threshold densities for reduced growth of sugarbeets, and the conditions under which some method of control must be considered (2).

The evidence presented here demonstrates the existence of a physiological variation in *H. schachtii*. Therefore, when economic, nematode-resistant sugarbeet cultivars are introduced, we may encounter the same problems that exist with soybean resistance to *H. glycines* Ichinohe. Physiological strains of *H. glycines* were observed soon after soybean resistance was found (12, 13,14,18,22). Both *H. schachtii* and *H. glycines* are amphimictic diploid species; they have the same number of chromosomes ( $n = 9$ ) (1,10,19), and the two species have been hybridized (11). Miller (8) states that the sugarbeet cyst nematode and the soybean cyst nematode may be subspecies of *H. schachtii*. Since the two species are so closely related (soybean has been shown to be a host of *H. schachtii* and sugarbeet a host of *H. glycines*) (7,9,16), it seems likely that physiological strains with *H. schachtii* is a distinct possibility.

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