The Effects of Hot Water Treatments on Survival of Heterodera schachtii

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Abstract: Cysts of *Heterodera schachtii* were treated in a water bath at constant temperatures ranging from 45 - 62.5 C for 1 sec to 28 hr. Treated and untreated cysts were incubated 8 weeks in sugarbeet root diffusate at 24 C to measure emergence of surviving larvae. Within the temperature range of 49 - 54 C, the minimum lethal temperature was proportional to the log time of treatment. No larvae emerged from cysts exposed 10 sec at 60 C. Although treatment of cysts for 8 hr at 45 C significantly reduced emergence, increasing the treatment period to 28 hr did not completely suppress emergence. *Key Words:* sugarbeet nematode, hatching, emergence, temperature.

The wide distribution of *Heterodera schachtii* Schmidt, 1871 (1, 2), together with a wide host range that includes crop, ornamental and weed plants (4), suggests that this nematode may be transported within infested nursery stock or in soil moved by various means.

Hot water treatment of plants and heat sterilization of small soil lots has been used for many years to eradicate a number of plant parasitic nematode species. However, the effects of high temperatures on survival of the sugarbeet nematode have not been investigated thoroughly. This paper reports a study of the lethal time-temperature relations for *H. schachtii*.

MATERIALS AND METHODS

Cysts of *H. schachtii* were obtained by washing soil from the roots of infected sugarbeet plants grown for 90-120 days in a greenhouse. Newly formed cysts filled with eggs were selected and divided into treatment groups which included four replications each consisting of 40 cysts. Cysts were transferred with pipettes to avoid damage to cyst walls or to cyst contents and stored in water for 10 days at 8 C to delay hatching and larval emergence until initiation of the test. Prior to treatment, the cysts were removed from refrigeration and kept at room temperature for 30 min.

The water bath was a styrofoam container filled with tap water and equipped with a variable thermo-regulator, a heater element, and a variable speed stirrer; it was maintained within ± 0.5 C of a preselected temperature ranging from 45 - 62.5 C (Table 1).

Treatments consisted of submerging weighted paper bags (Lipton's Flo-Thru® tea bags), each containing 40 cysts, in the water bath for 1 sec to 28 hr. When the bags were removed from the water bath, they were plunged immediately into tap water maintained at 24 C.

Treated and untreated cysts were placed in Syracuse watch glasses containing about 15 ml of sugarbeet root diffusate and incubated 8 weeks at 24 C. At weekly intervals the cysts were transferred to watch glasses containing fresh diffusate, and the emerged larvae were preserved in 5% formalin until counted. All larvae that emerged from cysts during the 8-week period were considered alive.

RESULTS

Emergence of larvae from treated cysts (Table 1) showed non-linear time-temperature relationships throughout the tested ranges (Fig. 1). However, within the temperature range of 49 - 54 C, the minimum temperatures which completely inhibited emergence of larvae from cysts was proportional to the log time of treatment (Fig. 2).

No larvae emerged from cysts exposed to 60 C for 10 sec. However, treatment periods of less

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TABLE 1. Effects of hot water treatments on subsequent emergence of larvae from cysts of *Heterodera schachtii* exposed 6 weeks to sugarbeet root diffusate.[1]

Temp. (C)	Minimum Lethal Time (Sec)	Maximum Non-lethal Time (Sec)	Number of Larvae Emerged	Percent Emergence[2]
62.5	-[3]	2.0	98	0.50
61	-	5	6	0.02
60	10	5	329	1.70
59	-	10	12	0.04
58	-	15	91	2.80
57	30	15	2	0.01
56	45	30	4	0.01
55	45	30	119	0.40
54	60	45	145	0.50
53	120	90	23	0.08
52	180	150	142	0.50
51	360	300	71	0.30
50	900	750	3	0.01
49	1800	1500	61	0.30
48	-	3600	115	0.60
47	-	5400	9318	29.00
46	-	10,800	7073	22.00
45	-	100,020	825	3.50

[1] Only data for minimum treatment times which totally stopped emergence and maximum times which did not stop emergence of all larvae are shown.
[2] Emergence from cysts exposed for the maximum non-lethal time expressed as a percent of emergence from untreated cysts.
[3] A lethal time period was not included in the tested range for the given temperature.

than 10 sec did not kill all larvae even at 62.5 C. No larvae emerged from cysts treated 4 hr (14,400 sec) at 47 C, but did emerge from cysts treated 8 hr at 45 C. Although the latter treatment significantly reduced larval emergence, increasing the time to 28 hr did not completely suppress emergence. Figure 3 illustrates that larval emergence from cysts is inversely proportional to the length of time that cysts are submerged in water at 45 C, and that total suppression of emergence would require treatment periods in excess of 28 hr.

The time-temperature combinations required to reduce emergence of larvae from treated cysts by 50% exhibit a linear relationship for treatment periods greater than 5 sec (Fig. 4). These data show that increasing the treatment temperature by 1 degree requires reduction of the treatment period by 2.5 sec to obtain the same effect.

Emergence of larvae from cysts was delayed by treatments which did not totally suppress emergence (Fig. 5, 6). Increased treatment periods at any given lethal temperature delayed emergence of larvae to a greater degree.

DISCUSSION

Endo (3) reported lethal temperatures of 49 - 62.5 C for *H. glycines*. Within this temperature range, the thermal death point was inversely proportional to the log treatment time. The observations that 49 C for 30 min and 60 C for 10 sec were lethal to *H. schachtii* agrees with the findings for *H. glycines* and for *H. rostochiensis* (5). However, in the present study, minimum lethal temperatures above 54 C were not proportional to the log time of treatment (Fig. 1). The dissimilarity of parameters of this study and those found for *H. glycines* is probably related to methods rather than to the physiology of the two species. Since none of the temperatures tested completely prevented hatching if applied for less than 10 sec, paper bags, cyst walls and/or eggs, and larvae adjacent to cyst walls may have insulated some of the more centrally located eggs against brief exposures to high temperatures.

The thermal LD_{50} (Fig. 4) is perhaps a more accurate indicator of time-temperature relationships than are thermal death points based on total suppressions of emergence, or the absence of mobility in a small percent of the population. As Fig. 4 shows, at treatment periods greater than 5 sec, the temperature required to depress emergence by 50% is inversely proportional to the time of treatment.

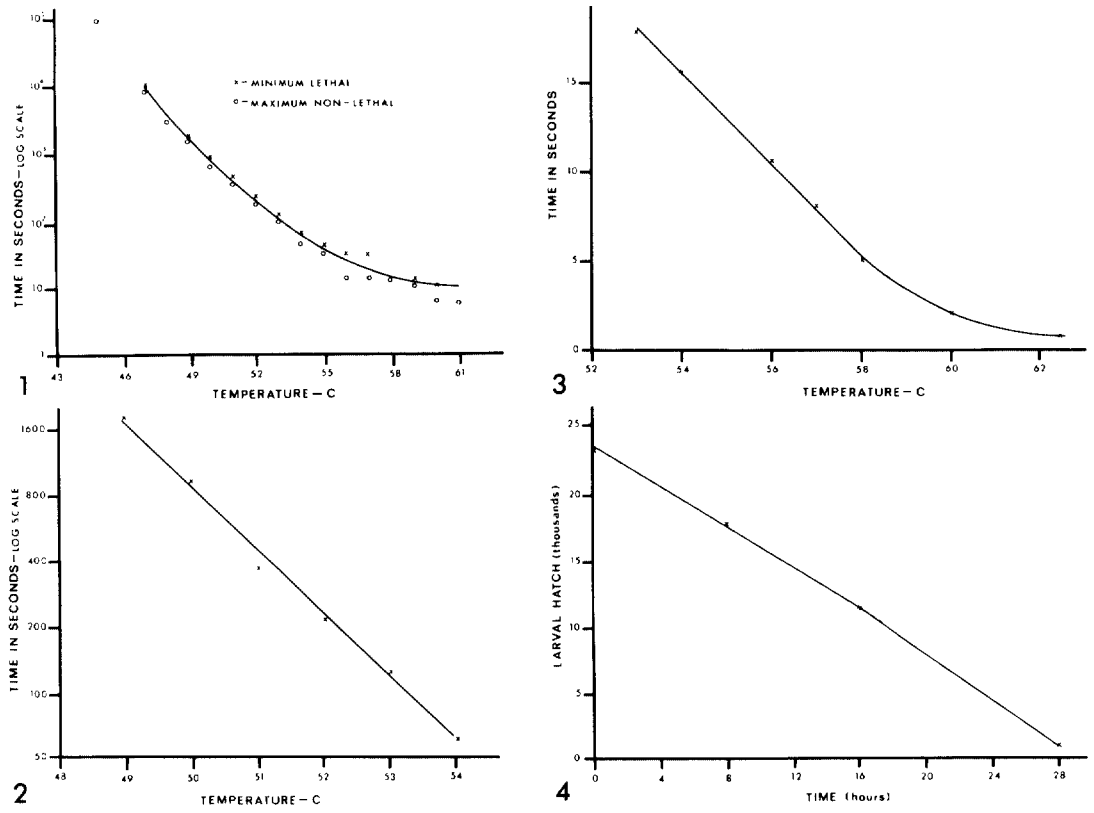

FIG. 1-4. 1. Time-temperature effects on survival of *Heterodera schachtii*. Cysts treated with hot water and then incubated 8 weeks in sugarbeet root diffusate. 2. Expanded segment of Fig. 1 showing proportional time-temperature relationships. 3. Influence of treatment at 45 C for various time periods on subsequent emergence of larvae from cysts in sugarbeet root diffusate. 4. Thermal LD_{50} for *H. schachtii*. Each point represents failure of 50% of the larvae to emerge from cysts incubated in sugarbeet root diffusate after hot water treatment at indicated times and temperatures.

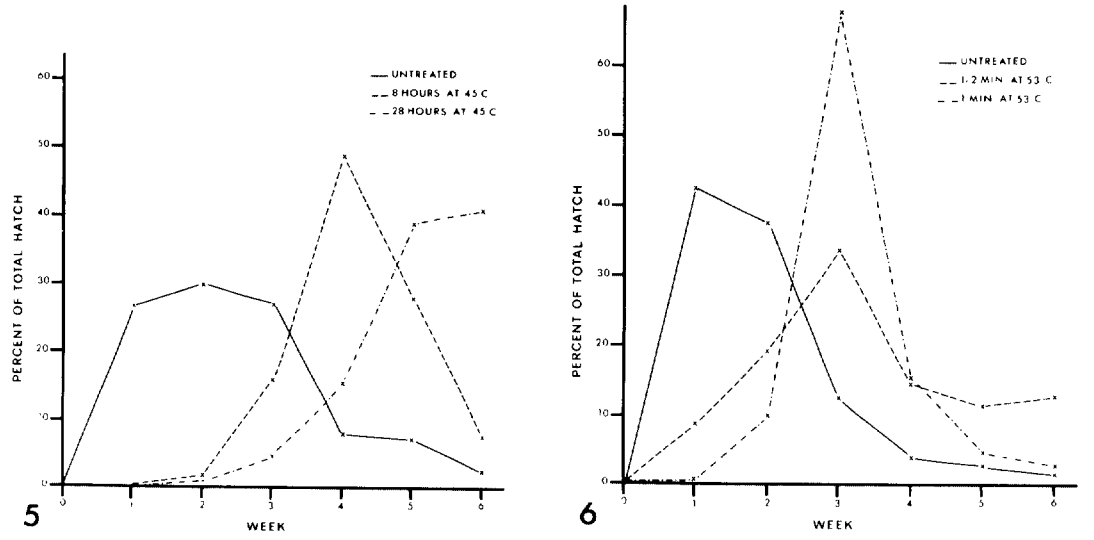

FIG. 5-6. Weekly hatches of larvae from treated and untreated cysts expressed as percents of total emergence during a 6-week period. 5. After 8- and 28-hr treatments at 45 C. 6. After 0.5- and 1.0-min treatments at 53 C.

The non-proportionality of time and temperature at temperatures lower than 49 C (Fig. 1) suggests that cyst contents (i.e., developmental stages) do not respond uniformly to selected temperatures in this range. However, at a given temperature (i.e., 45 C) emergence, and, therefore, survival is inversely proportional to treatment time.

Delayed emergence of larvae from treated cysts (Fig. 5, 6) suggests that larvae within eggs and/or hatched larvae oriented near the periphery of cysts may delay either movement of diffusate into centrally oriented eggs or emergence of newly hatched larvae. On the other hand, thermal stress at sub-lethal temperatures may temporarily suppress emergence of larvae which commences after a period of recovery.

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