Supplement to the Journal of Nematology 31(4S):662–666. 1999.

Damage and Reproduction Potentials of *Heterodera avenae* on Wheat under Outdoor Conditions

A. S. Al-Hazmi, F. A. Al-Yahya, and A. T. Abdul-Razig[1]

Abstract: Two pot experiments, in consecutive years, were conducted under outdoor conditions during the wheat growing season to examine the relationship between increasing initial population densities (Pi: 0–3,000 cysts/pot) of *Heterodera avenae* and corresponding responses of wheat cv. Yecora Rojo. Results of both experiments were very similar. The nematode suppressed plant height, root and biomass dry weights, and grain yield at all Pi's studied. The suppression of these parameters, as well as the final nematode population densities (Pf), increased with increasing Pi levels. The reproduction factor (Pf/Pi) decreased as Pi increased but was always greater than 1.0. When data from both experiments were combined for regression analyses, inverse relationships were found between $\log_{10}$ (Pi + 1) and both plant growth and yield. These negative relationships were highly significant and adequately described by linear models. Final population (Pf) increased linearly with Pi. The wheat cultivar cv. Yecora Rojo was found to be highly vulnerable to damage and a good host for *H. avenae*.

Key words: cereal cyst nematode, growth, *Heterodera avenae*, nematode, pathogenicity, reproduction, *Triticum aestivum*, wheat, yield.

Wheat (*Triticum aestivum* L.) is the most important field crop in Saudi Arabia. The crop is cultivated, under irrigation, throughout most of the country but most intensively in the Central Province. In 1982, the area under wheat cultivation was 735,000 ha and yield was only 187,000 MT. Since then production has increased to a peak in 1992, when the cultivated area was 924,407 ha producing 4.123 million MT (Anonymous, 1996).

The cereal cyst nematode (CCN), *Heterodera avenae* Wollenweber, was first reported on wheat and barley in the Riyadh region in 1992 (Al-Hazmi, 1992). The nematode pathotype infesting the region is very similar to Ha21 (Cook and Al-Hazmi, 1997). *H. avenae* has been spreading and has become a major limiting factor in wheat production in most wheat-producing regions in the country (Al-Hazmi et al., 1994; Al-Yahya et al., 1998; El-Meleigi and Al-Rokaibah, 1996). Most of the wheat cultivars grown in the country are susceptible to CCN (Al-Hazmi et al., 1994). Severe symptoms of stunting and yellowing often occur, and the nematode density in some heavily infested fields may reach 67 second-stage juveniles (J2)/g soil (Al-Hazmi et al., 1994), and yield loss may be as high as 93.5% (Al-Yahya et al., 1996).

Although previous reports or observations have provided evidence of CCN damage to wheat in Saudi fields, experimental studies are still needed to establish the damage function and reproductive potential of *H. avenae* on wheat. The objective of this study was to examine the relationship between increasing population densities (Pi) of *H. avenae* and the corresponding responses of wheat cv. Yecora Rojo in outdoor pot experiments.

Materials and Methods

Clean 12-cm-diam. plastic pots were filled with a steam-sterilized soil mixture of equal parts of sand and loamy sand soil. The soil mixture of each pot (ca. 1,500 cm^3 soil) was mixed thoroughly in a polyethylene bag with the designated inoculum of *H. avenae* and returned to its pot. The inoculum (brown cysts) was obtained from soil collected at the end of the previous season from a heavily infested wheat field in Al-Kharj region near Riyadh and stored at 5 °C. Cysts were extracted from soil with the modified Cobb decanting and sieving method (Barker, 1985). The average number of infective second-stage juveniles (J2) per cyst, determined from a sample of 50 cysts, was 53.5 J2/cyst. Cysts were used as inoculum,

Received for publication 3 March 1999.
[1] Professor, Assistant Professor, and Research Assistant, Department of Plant Protection, King Saud University, Box 2460, Riyadh 11451, Saudi Arabia.
This paper was edited by T. L. Kirkpatrick.

and six initial inoculum levels (Pi) of 0, 187, 375, 750, 1,500, and 3,000 cysts/pot were used. These densities correspond to 0, 10,000, 20,000, 40,000, 80,000, and 160,000 J2/pot. Treatments were replicated five times. The control treatment received the same volume of cyst-free decanting water. Four uniform seedlings of wheat cv. Yecora Rojo, a susceptible cultivar (Al-Hazmi et al., 1994), were transplanted into each pot. Pots were watered after transplanting and arranged in a randomized complete block design on a cardboard outside the greenhouse during the 1996/1997 wheat growing season. As needed, plants were watered and fertilized with water-soluble N-P-K (20-20-20) fertilizer. The experiment was repeated in the following season (1997/1998).

At harvest in both years (120 or 130 days after transplanting), plant heights were measured, and grain weight per spike and per pot were determined. Roots were separated gently from soil and weighed. Cysts on roots were dislodged by first washing roots gently to free them from soil, and then vigorously with a strong jet of water onto a 850-µm-pore sieve over a 150-µm-pore sieve (Hooper, 1986). Soil from each pot was thoroughly mixed, and a 250-cm^3 aliquot was taken to extract cysts and J2 with a modified centrifugation procedure (Barker, 1985) and a dense sucrose solution (Dunn, 1969). Since the numbers of J2 found in the soil at harvest were negligible, final population (Pf) was calculated from cysts collected from roots and soil. The reproduction factor (Rf = Pf/Pi) for each replicate was calculated. The shoot and root systems were oven-dried at 60 °C for a week, and dry weights of shoots, roots, and total biomass were determined.

Data from each experiment were subjected to analysis of variance (ANOVA), and treatment means were separated with Fisher's Protected LSD at $P \leq 0.05$ (SAS Institute, Cary, NC). Data from both experiments were combined and regression analyses were performed on selected plant responses and nematode reproduction versus Pi, $\log_{10}$ (Pi + 1), or $\log_{10}$ Pi.

Results

Results from both experiments were very similar. Plant heights and dry weights of both roots and biomass were suppressed ($P \leq 0.05$) even at the lowest Pi (187 cysts/pot) (Table 1). Suppression increased with Pi and reached up to 50% in plant height, 81% in roots, and 80% in biomass dry weights at the highest Pi level (3,000 cysts/pot) (Table 1). Similarly, yield (grain weight per pot or grain weight per spike) decreased ($P \leq 0.05$) in response to increasing inoculum level (Table 2). The yield reduction (18–35%) was evident at the lowest Pi, but the greatest reduction (80%) occurred at the highest Pi (Table 2).

Final nematode population (Pf) increased ($P \leq 0.05$) with increasing Pi in both years, but the nematode reproduced more in the second than in the first year (Table 3). Reproduction factors (Rf) decreased ($P \leq 0.05$) in both years, except at Pi = 375 in the second year, as Pi increased

TABLE 1. Growth responses of wheat cv. Yecora Rojo to different initial population densities (Pi) of *Heterodera avenae* in two outdoor pot experiments.

Pi (cysts/1,500 cm^3 soil)	Plant height (cm)		Root dry weight (g)		Biomass dry weight (g)	
	1996	1997	1996	1997	1996	1997
0	45.2 a	52.1 a	1.25 a	1.16 a	7.90 a	7.73 a
187	40.7 b	42.4 b	0.55 b	0.58 b	7.02 b	5.80 b
375	33.7 c	38.4 c	0.41 c	0.46 c	4.93 c	4.49 c
750	29.2 d	34.6 d	0.26 d	0.34 d	3.78 d	3.94 d
1,500	27.4 d	28.6 e	0.33 cd	0.27 de	2.85 e	3.44 d
3,000	24.8 e	26.0 f	0.29 cd	0.22 e	2.06 f	2.72 e

Values are means of five replicates. Means in a column followed by the same letter are not significantly different ($P \leq 0.05$) according to Fisher's Protected LSD.

TABLE 2. Effect of different initial population densities (Pi) of *Heterodera avenae* on yield of wheat cv. Yecora Rojo in two outdoor pot experiments.

Pi (cysts/1,500 cm^3 soil)	Grain weight/pot (g)		Grain weight/spike (g)	
	1996	1997	1996	1997
0	3.70 a	3.20 a	0.93 a	0.80 a
187	2.41 b	2.64 b	0.60 b	0.66 b
375	1.85 c	1.95 c	0.46 c	0.49 c
750	1.66 c	1.52 d	0.41 c	0.38 d
1,500	1.14 d	0.98 e	0.29 d	0.24 e
3,000	0.74 e	0.73 e	0.19 e	0.18 e

Values are means of five replicates. Means in a column followed by the same letter are not significantly different ($P \leq 0.05$) according to Fisher's Protected LSD.

(Table 3), but Rf was always greater than one.

When data from both growing seasons were combined in the analysis, inverse relationships were found between inoculum level and wheat growth and yield. These negative relationships were highly significant ($P \leq 0.0001$), and adequately described by linear models (Table 4). Plant height (cm) and dry weight (g) of plant biomass were negatively correlated with $\log_{10}$ (Pi + 1) with $R^2 = 0.78$ and 0.80, respectively. Similarly, yield (grams per pot) was also negatively correlated with $\log_{10}$ (Pi + 1) with $R^2 = 0.83$. Final cyst populations increased linearly with Pi ($R^2 = 0.82$), but the reproduction factor (Rf) was negatively correlated with $\log_{10}$ (Pi) with $R^2 = 0.30$ (Table 4).

DISCUSSION

Heterodera avenae caused significant damage and reproduced well on wheat cv. Yecora Rojo. Suppressions of growth and grain yield were dependent on Pi levels. Even at the lowest Pi of 187 cysts/pot (7 J2/cm^3 soil), *H. avenae* caused significant growth and yield suppression. At the highest Pi of 3,000 cysts/pot (107 J2/cm^3 soil), grain yield was reduced by up to 80%. Consequently, cultivation of wheat would not be profitable at this Pi level, and greater damage and crop losses might be expected under stressful field conditions. For example, in small field plots, naturally infested with *H. avenae*, we have found that the yield loss of wheat cv. Yecora Rojo was 93.5% (Al-Yahya et al., 1996). According to Swarup and Sosa-Moss (1990), yield losses of wheat and barley may be 45–48% in light soils even at a density of 6 eggs/g soil. In the Central Province of Saudi Arabia many wheat fields are heavily infested with *H. avenae*, and damage in some fields is so great that some growers plow the soil and grow alfalfa or vegetable crops instead (Al-Hazmi et al., 1994). Although yield depends on many interacting factors, *H. avenae* disturbs several physiological aspects of the infected wheat plants, such as photosynthesis, mineral uptake, transpiration, temperature of the plant canopy, and water content of leaves and roots (Al-Yahya et al., 1998). Our results on suppression of growth and yield of wheat caused by *H. avenae* support different reports from other countries (Brown, 1982; Dhawan and Nagesh, 1987; Greco et al., 1993; Rivoal and Sarr, 1987; Sabova et al., 1986; Williams and Beane, 1982).

In our study, the damage threshold level was 7 J2/g soil (187 cysts/pot). This level is similar to the threshold levels on wheat and barley reported by Swarup and Sosa-Moss (1990) from India (6 eggs/g soil), by Shahina and Maqbool (1990) from Pakistan (100–150 cysts/plant), and by Sabova et al. (1981) from Slovakia (180 cysts/pot). In the temperate semi-arid regions of Australia, which have a somewhat similar climate to that of Saudi Arabia, *H. avenae* reduced yield of wheat and barley by 20% at a Pi of 2 eggs and J2/g soil, and by 40% at a Pi of 16 eggs and J2/g soil (Meagher and Brown, 1974).

TABLE 3. Reproduction of *Heterodera avenae* on wheat cv. Yecora Rojo at different initial population densities (Pi) in two outdoor pot experiments.

Pi (cysts/1,500 cm^3 soil)	Pf (cysts/pot)		Reproduction factor (Rf)	
	1996	1997	1996	1997
187	622 d	744 e	3.32 a	3.97 b
375	733 d	1,884 d	1.95 b	5.02 a
750	1,434 c	2,696 c	1.91 b	3.60 c
1,500	2,464 b	4,273 b	1.64 bc	2.85 d
3,000	4,786 a	8,268 a	1.60 c	2.76 d

Values are means of five replicates. Means in a column followed by the same letter are not significantly different ($P \leq 0.05$) according to Fisher's Protected LSD.

Reproduction factor (Rf) = Pf/Pi.

TABLE 4. Summary of relationships between initial inoculum density (Pi) of *Heterodera avenae* (X) and damage and nematode reproduction (y) on wheat cv. Yecora Rojo, 1996–1998.[a]

		Linear regression			
X	y	a	b	R^2	P
$\log_{10}$ (Pi + 1)	Plant height (cm)	50.96	−6.54	0.78	0.0001
$\log_{10}$ (Pi + 1)	Dry biomass (g/pot)	8.35	−1.5	0.80	0.0001
$\log_{10}$ (Pi + 1)	Grain yield (g/pot)	3.68	−0.75	0.83	0.0001
Pi	Pf	438.96	2.02	0.82	0.001
$\log_{10}$ Pi	Rf	6.85	−1.39	0.30	0.001

[a] Data were combined from two outdoor pot experiments (n = 10). Pi and Pf are based on number of cysts per pot (1,500 cm^3 soil). Rf = Pf/Pi.

Although several investigators reported varying damage threshold levels of *H. avenae* on wheat and barley, generally 1 to 40 eggs and J2/g soil have been reported from different countries (Dhawan and Nagesh, 1987; Gill and Swarup, 1971; Greco and Brandonisio, 1987; Meagher and Brown, 1974; Stone, 1968; Swarup and Sosa-Moss, 1990). Many biotic and abiotic factors in different regions may affect the estimates of the damage thresholds of *H. avenae* on wheat or other cereals. These factors include: soil type, rainfall, cereal plant or cultivar, nematode pathotype, nutrients, type or location of test, presence of other nematodes or pathogens, and nematicides used in trials (Rivoal and Cook, 1993; Simon, 1980; Williams and Beane, 1982).

Heterodera avenae reproduced readily on wheat cv. Yecora Rojo. Final numbers of nematode (Pf) increased with Pi (r = 0.90, P = 0.0001), and the reproductive factor (Rf) was always greater than 1.0 (1.6–5.0). The cultivar was a good host for *H. avenae* but relatively less suitable than some barley cultivars and lines (Al-Hazmi et al., 1994). As previously reported (Dhawan and Nagesh, 1987; O'Brien and Fisher, 1978; Rivoal and Sarr, 1987), Rf was inversely related to $\log_{10}$ Pi (r = −0.55, P = 0.001). The decrease of Rf is most likely due to the greater damage of roots with increasing inoculum levels.

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