Effects of Fall Infection by *Gaeumannomyces graminis* var. *tritici* and Triadimenol Seed Treatment on Severity of Take-All in Winter Wheat

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ABSTRACT

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Treatment of winter wheat seed with the systemic fungicide triadimenol at 14.8 and 29.6 ml a.i./45.4 kg of seed reduced take-all under artificially and naturally infested field conditions. Yield losses due to take-all were reduced 60–75% depending on rate of triadimenol applied. In greenhouse experiments, the treatments protected seedling roots from infection by *Gaeumannomyces graminis* var. *tritici* (Ggt) for 4 and 8 wk, respectively. In the field, the lower rate of fungicide delayed infection by Ggt approximately 6 wk during the fall. Conversely, increases in the incidence of infection by Ggt during the winter and spring were virtually the same for plants derived from treated or nontreated seed. In six fields with known take-all levels, the high correlations for incidence of Ggt and percentage of whiteheads obtained when incidence was measured 4 or 8 wk after planting indicated that fall infections by Ggt strongly influenced the amount of take-all yield loss in conducive soils under Kansas conditions. Seed treatment with triadimenol protects wheat plants from Ggt during this critical period resulting in control.

Take-all is a severe disease of wheat (*Triticum aestivum* L.) caused by the soilborne fungus *Gaeumannomyces graminis* (Sacc.) von Arx and Olivier var. *tritici* Walker (Ggt). When roots and crowns are damaged to the extent that symptoms such as plant death or whiteheads are obvious, yields are usually reduced by more than 50% (24). Controls for take-all include crop rotation (14,20), proper fertility utilizing chloride (5,17) or ammoniacal forms of nitrogen (12,25), tolerant cultivars (16), and late planting (8). However, in Kansas, many producers either do not follow these recommendations or sustain take-all losses in spite of the procedures.

Crop rotation is not popular in regions of Kansas where continuous wheat production is practiced due to the lack of suitable alternate winter crops or the loss of one cropping season in three when winter wheat is rotated with summer crops. Thus, over 50% of the wheat is grown on continuous-cropped land (1). Furthermore, over 70% of the total nitrogen fertilizer used in Kansas is in the ammoniacal form (2), yet take-all continues to rank as the state's fourth most important wheat disease (23). Similarly, delayed planting is not a widespread practice because it increases soil erosion and directly reduces yields. During the period from 1977 through 1981, take-all caused an estimated average annual yield loss in Kansas of 150 million kg (5.6 million bushels) (23). Clearly, additional control procedures are necessary for wheat production under Kansas conditions.

Chemical control of take-all of wheat using foliar or soil drench fungicides has not been effective in the past (3,13,18) and most seed treatment fungicides tested have not effectively controlled the disease (13,18). Chemical control with soil fumigants or specific fungicides also may eliminate microorganisms antagonistic to the take-all fungus (6,9,15,19). Although benomyl at 35 kg/ha incorporated below wheat seed reduced take-all in field trials, this high rate would preclude its commercial use in the USA (4). Soil incorporation of triadimefon had no effect upon take-all development in pot experiments (3). Conversely, triadimefon used as a seed treatment was shown to reduce take-all in an artificially inoculated field trial (8) and indicated some promise for reduction of disease severity with chemicals.

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Preliminary field experiments with the systemic seed treatment fungicide triadimenol (β-[4-chlorophenoxyl]-α-[1,1-dimethylethyl]-1*H*-1,2,4-triazole-1-ethanol) indicated that it reduced take-all severity on winter wheat. This study was initiated to determine the potential of triadimenol to control take-all losses under naturally and artificially inoculated conditions, to study the effect of triadimenol on the disease incidence curve, and to relate the percentage of plants infected by Ggt in the fall to final take-all severity. The latter would provide information needed to determine the importance of reduced fall infections on yield losses due to take-all.

MATERIALS AND METHODS

Artificially inoculated field experiment. Winter wheat seed (cultivar Newton was used throughout) was treated with triadimenol (Baytan 150 FS, Mobay Chemical Corporation, Kansas City, MO 64120) at rates of 0, 14.8 ml (0.5 oz a.i./cwt) or 29.6 ml a.i./45.4 kg seed (1.0 oz a.i./cwt). To quantify the effect of seed treatment on take-all, an uninoculated-inoculated split plot field experiment with four replications was used. Subplots were 4.6-m paired rows spaced 31 cm apart that received either autoclaved oat kernels or oat kernels colonized by Ggt at the rate of 1 g kernels per meter of drill row. The kernels were introduced along with the seed at planting (6). At growth stage 85 (soft dough) (26), 50 randomly selected tillers in each plot were rated for premature ripening (whiteheads). Upon maturity, each plot was individually harvested to determine grain yields with weights adjusted to 10% moisture. Percentage yield loss due to take-all was determined by subtracting the yield of an inoculated plot from the yield of the adjacent uninoculated plot.

Naturally infested field experiment. To determine the effect of triadimenol on take-all under natural disease conditions, a randomized block experiment with four replications was established in a field, in which moderate take-all losses (50% whiteheads) had been sustained the previous season (1979–1980). Treatments included winter wheat seed left either nontreated or treated with triadimenol at the rate of 14.8 ml a.i./45.4 kg seed. Seed was planted with a grain drill into 1.2 × 7.6-m plots. Periodically throughout the growing season, notes were taken on the occurrence of various diseases including leaf rust, speckled leaf blotch, Cephalosporium stripe, and tan spot. At the soft-dough growth stage, percentage whiteheads was determined by rating 100 random tillers per plot for premature ripening. Upon maturity, each plot was harvested with a combine and grain weights adjusted to 10% moisture.

Greenhouse experiment. Winter wheat seed was left nontreated or treated with triadimenol at rates of 14.8 or 29.6 ml a.i./45.4 kg seed. Three seedlings were grown in vermiculite in 2.5- × 12.5-cm plastic tubes (Ray Leach Nursery, Canby, OR 97013) with six autoclaved oat kernels colonized by Ggt placed 1 cm below the seed in each tube (7,11). To determine the duration of protection from Ggt provided by triadimenol, tubes planted to treated or nontreated seed were inoculated at planting, watered and fertilized as needed, and maintained in the greenhouse (19 ± 4 C). Either 4, 8, or 12 wk after tubes received inoculum, roots were washed free of vermiculite and percentage root destruction on seedlings was rated in shallow white pans under a dissecting microscope (25). Similarly, there were treatments in which inoculation occurred 4 or 8 wk after planting time and percentage of root rot was rated 4 wk after inoculation. Inoculation at these times involved removing the plants from the tubes, placing the kernels 1 cm below the seed in contact with the roots, followed by replanting. Appropriate noninoculated control treatments received autoclaved oat kernels not colonized by Ggt, and there were 20 replicate tubes for each treatment. At the time seedling roots were rated for take-all, plant parts above the seed level were removed and oven-dry weights were determined.

Fall incidence of Ggt on wheat roots. Six grower's wheat fields were selected, which were in the first 4 yr of wheat production and which had various take-all levels ranging from none (no whiteheads) to severe (75% whiteheads). At 2-, 4-, and 8-wk intervals after planting, samples were collected from a 1,000-m^2 area in each field with a minimum of 100 random plants dug from each area on each sample date. The roots were washed free of soil and all plant parts above the crown were detached and discarded. The roots and crown of each individual plant were placed in a plastic tube two-thirds filled with sterile vermiculite. This "bait" was covered with 1 cm of additional vermiculite, three wheat seeds were added, and the tube was filled to the top with vermiculite. Tubes were fertilized and watered as needed for 4 wk, then the roots of the seedling trap plants were washed free of vermiculite and rated for the presence of take-all lesions by observation with a dissecting microscope. The percentage of tubes with trap plants infected with take-all was assumed to reflect the percentage of field-collected "bait" plants with Ggt associated with their root systems or crowns at the time of sampling. At soft dough stage, areas in the fields where sample collection occurred were rated for percentage whiteheads.

Effect of triadimenol on the incidence of Ggt. Winter wheat seed treated with triadimenol (14.8 ml a.i./45.4 kg seed) or left nontreated was planted into 1.2- × 7.6-m plots and artificially inoculated with Ggt-colonized oat kernels. A randomized block design with two treatments and five replications was used. Periodically during the growing season, 25 random plants per plot were dug, their roots and crowns were washed free of soil, and used as "baits" as described above. After the 4-wk growth period in plastic tubes, the percentage tubes with infected trap plants was assumed to reflect the percentage field-collected plants with Ggt associated with their root systems and crowns at the time of sampling.

RESULTS

In the artificially inoculated plots of the field experiment, triadimenol seed treatment at 14.8 ml a.i./45.4 kg of seed reduced whiteheads 63% and take-all yield loss 61% (Table 1). The higher rate of seed treatment (29.6 ml a.i./45.4 kg of seed) reduced whiteheads 74% and take-all yield loss 78%. Similarly, in the field naturally infested with Ggt, the low rate of triadimenol seed treatment reduced whiteheads 57% and increased yield 38% compared to the nontreated control (Table 1). At the test site, take-all was the only disease that significantly affected yield.

In the greenhouse experiment, both rates of triadimenol seed treatment significantly reduced the incidence of take-all root rot 4 and 8 wk after seeding when inoculum of Ggt was added at planting (Table 2). The higher rate gave significantly greater control than the lower rate when severity was rated 8 and 12 wk after seeding. At all sample periods, dry weights of plant parts above seed level were reduced by an amount that reflected the amount of root rot (Table 2). At the 4- and 8-wk sample periods, plant dry weights of inoculated triadimenol treatments were not significantly different from the uninoculated controls. Delays in germination, reduced early seedling growth, and darker green leaves were noted on plants from triadimenol-treated seed; however, these differences disappeared after 4 wk of growth as evidenced by the dry weights of uninoculated treatments (Table 2).

When inoculum of Ggt was added 4 wk after planting, only the treatment with the higher rate of triadimenol resulted in significant reduction in take-all root rot (Table 2). In this experiment, root rot did not progress to the point that plant dry weights of either treated or nontreated plants were reduced. Neither rate of triadimenol reduced root rot relative to the nontreated check when inoculum was added 8 wk after planting (Table 2). Although plant dry weights were reduced in these treatments, they were not significantly different from the uninoculated controls, indicating that during the 4 wk after inoculation, root rot had not progressed sufficiently to significantly reduce top growth.

The percentage of 2-wk-old wheat plants collected from naturally infested fields with Ggt associated with their root systems was low and poorly correlated ($r = 0.71$) with the percentage of whiteheads the following spring (Fig. 1). Estimates of Ggt association taken 4 or 8 wk after planting increased progressively and were highly correlated ($r = 0.88$ and 0.95, respectively) with percentage whiteheads.

Triadimenol seed treatment significantly reduced fall incidence of Ggt on wheat roots and crowns (Fig. 2). When wheat was entering the winter dormant period (5 December sampling date), plants derived from triadimenol-treated seed exhibited a Ggt incidence equivalent to the Ggt incidence on nontreated plants

TABLE 1. Effect of seed treatment with triadimenol on take-all disease severity, yield loss, and yield of winter wheat

Inoculum	Seed treatment (ml a.i./45.4 kg of seed)	Percentage whiteheads[v]	Take-all yield loss (%)[w]	Yield in kg/ha (bu/ac)
Artificial[x]	None	48[y] a	57 c	1344 (20) f
	Triadimenol (14.8)	18 b	22 d	2420 (36) g
	Triadimenol (29.6)	13 b	13 e	2689 (40) h
Natural[z]	None	35 i	...	2151 (32) k
	Triadimenol (14.8)	15 j	...	2958 (44) l

[v]At least 50 randomly selected tillers per plot were rated for premature ripening at soft-dough growth stage.
[w]Yield loss determined by subtracting the yield of an inoculated plot from the yield of the appropriate uninoculated plot.
[x]Oat kernels colonized by *Gaeumannomyces graminis* var. *tritici* (Ggt) introduced into the drill row with the seed at planting (1 g/m).
[y]Means of four replications and are significantly different ($P = 0.05$) according to Duncan's multiple range test or LSD if followed by different letters.
[z]Grower's field naturally infested with Ggt.

collected approximately 6 wk earlier (20 October sampling date). The increase in incidence of Ggt during the winter and spring months was approximately the same for both treatments (Fig. 2).

DISCUSSION

Treatments of winter wheat seed with the systemic fungicide triadimenol significantly reduced yield loss due to take-all under artificially and naturally infested field conditions. Although the lower treatment rate (14.8 ml a.i./45.4 kg seed) provided moderate to good control (approximately 60% reduction in yield loss) the higher rate (29.6 ml a.i./45.4 kg seed) was significantly better (75% reduction). Since neither rate gave complete control, some whiteheads and yield losses are expected to occur under high Ggt pressure even though seed is treated with triadimenol. It is not known if continuous triadimenol seed treatment will prevent the buildup of take-all during the first few years of wheat monoculture or if it will affect the amount of carryover of Ggt inoculum between susceptible crops. Further research will be necessary to determine the full prophylactic and/or eradicative potential of this control procedure.

Effective take-all control appears to be due to the protection of winter wheat plants from infection by Ggt during the fall growth period. In greenhouse studies, the lower fungicide rate protected

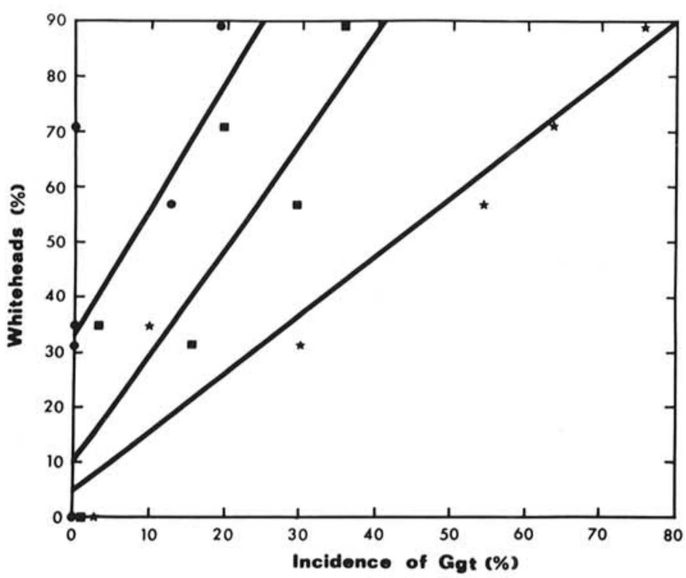

Fig. 1. Correlation of percentage winter wheat plants having *Gaeumannomyces graminis* var. *tritici* (Ggt) associated with their root systems with percentage prematurely ripe tillers (whiteheads) at soft-dough growth stage. Plants sampled from six grower's fields 2 wk (circles: $r = 0.71$), 4 wk (squares: $r = 0.88$), or 8 wk (stars: $r = 0.95$) after seeding.

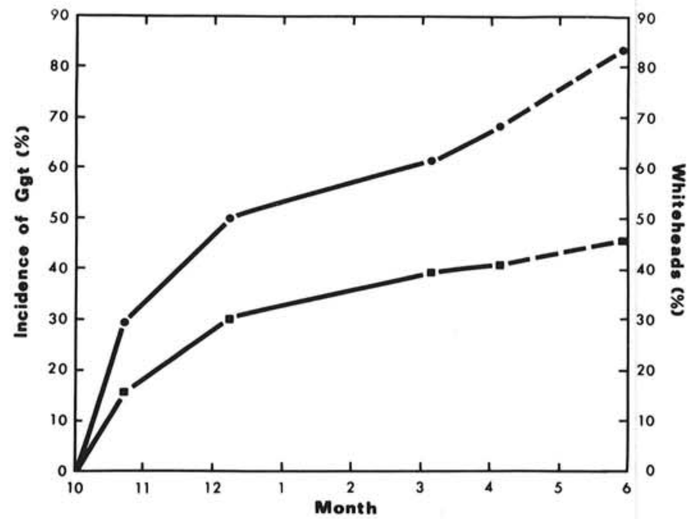

Fig. 2. Relationship between percentage of winter wheat plants having *Gaeumannomyces graminis* var. *tritici* (Ggt) associated with their root systems and the month the sample was collected. Artificially inoculated plots planted 1 October using nontreated seed (circles) or seed treated with triadimenol at the rate of 14.8 ml a.i./45.4 kg seed (squares). Values represent the means of five replications ($LSD_{0.05}$ = 7.4, 14.5, 9.7, and 9.1 for sample dates 20 October, 5 December, 3 March, and 2 April, respectively). Percentage prematurely ripe tillers (whiteheads) determined at the soft-dough growth stage (29 May).

TABLE 2. Effect of postplant time of inoculation on control of take-all root rot with triadimenol seed treatments

Seed treatment (ml a.i./45.4 kg seed)	4 wk after planting		8 wk after planting		12 wk after planting	
	Take-all root rot (%)[w]	Dry weight per plant (g)[x]	Take-all root rot (%)	Dry weight per plant (g)	Take-all root rot (%)	Dry weight per plant (g)
Uninoculated						
Nontreated	0[z] a	0.031 f	0 g	0.098 lm	0 n	0.158 st
Triadimenol (14.8)	0 a	0.033 f	0 g	0.101 lm	0 n	0.163 st
Triadimenol (29.6)	0 a	0.027 f	0 g	0.091 lm	0 n	0.168 st
Inoculated at planting[y]						
Nontreated	66 c	0.016 e	83 j	0.028 k	90 p	0.048 q
Triadimenol (14.8)	17 b	0.034 f	54 i	0.079 l	78 p	0.118 r
Triadimenol (29.6)	4 ab	0.029 f	7 h	0.104 m	36 o	0.174 t
Inoculated 4 wk after planting[y]						
Nontreated	...	...	37 i	0.092 lm	...	...
Triadimenol (14.8)	...	...	37 i	0.098 lm	...	...
Triadimenol (29.6)	...	...	7 h	0.095 lm	...	...
Inoculated 8 wk after planting[y]						
Nontreated	...	...	...	...	31 o	0.138 rs
Triadimenol (14.8)	...	...	...	...	37 o	0.143 s
Triadimenol (29.6)	...	...	...	...	29 o	0.147 s

[w] Percentage root system with characterisitic lesions and/or runner hyphae of *Gaeumannomyces graminis* var. *tritici* (Ggt).
[x] Plant parts above the seed removed, oven dried, and weighed.
[y] Six oat kernels colonized by Ggt placed 1 cm below the seed in each tube.
[z] Means of 20 replicate tubes and are significantly different ($P = 0.05$) within the same column according to Duncan's multiple range test if followed by different letters.

seedlings for about 4 wk after inoculum was added at seeding. When inoculum was added 4 wk after planting and incubated an additional 4 wk, the lower rate did not reduce the percentage of root rot. After 4 wk of plant growth, the fungicide had presumably been reduced to ineffective levels due to translocation, dilution, leaching, and/or inactivation. The higher fungicide rate significantly reduced root rot an additional 4 wk even when inoculum was added 4 wk after seeding. However, when the inoculum was added 8 wk after planting, it was apparent that the higher rate had also been reduced to ineffective levels. Thus, this rate apparently reduced infection by Ggt and root rot activity for about 8 wk. This is supported by the results obtained with plants inoculated at planting, in which root rot had not developed significantly at 8 wk (7%), but had developed to 36% at the 12-wk sampling period.

The field data confirmed the greenhouse data. When seed was treated with triadimenol at the lower rate, there was approximately a 6-wk delay in the incidence of Ggt at the beginning of winter, indicating that chemical seed treatment protected roots from colonization by Ggt during the fall. The relative increases in incidence of Ggt during the winter and spring were virtually identical for wheat from treated or nontreated seed because the fungicide apparently had been reduced to ineffective levels at those times.

Results of this study indicate that under Kansas conditions, early fall infections of winter wheat by Ggt strongly influence the amount of yield loss due to take-all. Although triadimenol protection of roots is virtually eliminated 4–8 wk after planting, it results in effective take-all control. Since infections by Ggt can occur at any time during the season (27), the early fall infections would have a longer time to become established and subsequently destroy the root system than would late fall or spring infections. Thus, protection of wheat plants in the fall with triadimenol seed treatment reduces the time for root rot to develop.

Explanations for the mechanism of take-all control provided by late planting (15) may also apply to the control obtained with triadimenol. Since Ggt is a poor competitive saprophyte (10), delayed planting results in a longer time between harvest of one crop and sowing of the next, and thus more time for inoculum degradation by soil microorganisms to reduce Ggt inoculum potential. Triadimenol seed treatment would have the same effect by inhibiting Ggt root-colonizing activity during the fall an additional 4–8 wk. Confining Ggt to its saprophytic state for this 4- to 8-wk period would provide time for the inoculum to undergo an equivalent period of additional decay. However, unlike delayed seeding, triadimenol seed treatment would not increase soil erosion or reduce winter survival and yields because early planting could be practiced.

The baiting procedure used in this study was found to be a more sensitive method for detecting Ggt associated with wheat roots than direct observation of field-collected roots. This was true especially during fall sampling periods when initial stages of Ggt infection are difficult to detect on field-collected roots and other soil microorganisms or injuries cause root discoloration mimicking early take-all. However, after the 4-wk incubation in vermiculite, characteristic runner hyphae of Ggt (7,11,27) on trap plant roots enable positive diagnosis of take-all. In no instance was there a 1:1 correlation between the incidence of Ggt determined by the baiting procedure and the development of whiteheads, perhaps due to poor root sampling efficiency or insensitivity of the assay in detecting early infections by Ggt.

The baiting procedure also indicated the importance of fall infections in Kansas since high correlations between incidence of Ggt and whitehead development were obtained when fields were sampled in the fall, 4 and 8 wk after seeding. However, since only a small number of fields were used, and these had previously determined levels of Ggt, it is not known if the baiting procedure will have any value in forecasting take-all severity. This is particularly true since none of the grower's fields used in this study had been in long-term wheat production (21,24) nor were they displaying suppressiveness to take-all (22). Nevertheless, for the take-all conducive fields used in this study, fall incidence of Ggt was an important determinant in the amount of loss due to take-all.

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