

Influence of Flutolanil and Tolclofos-Methyl on Root and Culm Diseases of Winter Wheat

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ABSTRACT

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Two fungicides with high toxicity to species of *Rhizoctonia* were adsorbed onto a granular fertilizer and banded in, below, or between the seed furrows at the time of planting winter wheat. Experiments were performed under conventional tillage (chisel plow and disk) and no-tillage systems. Flutolanil and tolclofos-methyl were each highly toxic to growth of *R. solani* AG-8 and *R. oryzae* in vitro, were inconsistent in suppressing the incidence and severity of *Rhizoctonia* root rot in the field, and did not significantly reduce the incidence of take-all or eyespot. Mixing metalaxyl with each of the primary fungicides did not improve disease control. Application of a starter fertilizer at planting increased the severity of *Rhizoctonia* root rot in a no-till system but had no effect on the disease when wheat was grown with a conventional-tillage system. Fungicides did not improve the yield of grain in these experiments.

In 1984, *Rhizoctonia* root rot was identified as an important disease of wheat (*Triticum aestivum* L.) and barley (*Hordeum vulgare* L.) in the Pacific Northwest region of the United States (13,15). The disease commonly reduces wheat yields by at least 4%, and the loss in one field was determined to be at least 17% (Smiley, unpublished). In a field infested by *Rhizoctonia*, the fumigation of soil with methyl bromide during August of 1986 led to an increase in production of spring wheat by 32% during 1989 and also allowed crop maturation to occur 21 days earlier than in the non-fumigated portion of the field (Smiley, unpublished). The dominant and most virulent species and intraspecific groups (12) of *Rhizoctonia* in the affected fields are *R. solani* Kühn AG-8 (11) and *R. oryzae* Ryker & Gooch.

Smiley et al (14) evaluated fungicide seed treatments for their efficiency in controlling *Rhizoctonia* root rot. Several of the fungicides were highly inhibitory to growth of *R. solani* AG-8 and *R. oryzae* in vitro, but did not reduce the incidence of *Rhizoctonia* root rot or improve grain yields in field studies. Most of the fungicides reduced tillering of winter wheat, thereby precluding the examination of higher application rates as seed treatments. Tolclofos-methyl was especially toxic to both of the dominant pathogenic species of *Rhizoctonia*. This fungicide consistently reduced root rot,

although not at a significance level of $P < 0.05$. Unfortunately, tolclofos-methyl was also among the fungicides that were phytotoxic to wheat.

Recent studies have illustrated that much higher efficiency can be achieved from certain fungicides when they are banded below or with the seed, rather than coated onto the seed (3,4). The fungicides were applied by adsorbing them onto sand or fertilizer granules, and then dispensing them at the time of planting. This placement technique has the potential to reduce phytotoxicity and to position the fungicide so that it can protect roots below the caryopsis, through acropetal translocation subsequent to absorption. These are essential attributes for fungicides that may be capable of controlling root diseases of wheat. This technique becomes especially important in that acropetally translocated fungicides highly toxic to species of *Rhizoctonia* are now available (2,6,7).

Flutolanil is a fungicide that appears to possess properties desired for controlling *Rhizoctonia* root rot (2,6,10). Although flutolanil has been proven toxic ($EC_{80} = 0.2-2.1 \mu\text{g/ml}$) to anastomosis groups AG-1, 1A, 2-2, and IIIB of *R. solani*, it has apparently not been tested for inhibition of the most important group (AG-8) causing *Rhizoctonia* root rot of cereals in the Pacific Northwest, and it is reported as relatively ineffective against *R. oryzae* ($EC_{80} = 29 \mu\text{g/ml}$) (1). It was important to determine the toxicity of flutolanil to Pacific Northwest isolates of *R. solani* AG-8 and *R. oryzae*. Likewise, it was important to determine whether placement of the fungicide in the root zone could effectively control *Rhizoctonia* root rot.

Inoue et al (7) determined (*personal communication*) that tolclofos-methyl

and flutolanil were the most fungistatic of the compounds evaluated for suppressing growth of most anastomosis groups of *R. solani*. Both compounds were much more fungistatic than quintozene (pentachloronitrobenzene). Tolclofos-methyl also suppressed growth of many ascomycetous fungi and, to some extent, *Pythium debaryanum* Auct. non R. Hesse. Inoue et al (7) also reported that tolclofos-methyl was compatible with all fertilizers evaluated and that the pH of soil did not influence the efficiency of seed treatments applied to beans.

In addition to examining the effects of placement on the fungicidal and phytotoxic activities of fungicides targeted toward species of *Rhizoctonia*, it is also important to determine the impact of such treatments on other diseases that occur in the region. Take-all (caused by *Gaeumannomyces graminis* (Sacc.) Arx & Olivier var. *tritici* J. Walker), eyespot (caused by *Pseudocercospora herpotrichoides* (Fron) Deighton), and *Pythium* root rot (caused by species of *Pythium*) are also frequently observed on winter wheat plants in the Pacific Northwest. Complexes of two or more of these agents with species of *Rhizoctonia* are commonly observed. Much of the commercial wheat seed in this region is treated with a mixture of carboxin plus thiram. In areas where *Pythium* root rot is common, metalaxyl is often added to the basic seed treatment.

The objectives of this study were: 1) to determine the toxicity of flutolanil to the important agents of *Rhizoctonia* root rot in the Pacific Northwest and 2) to determine whether banding flutolanil and tolclofos-methyl below or between the seed rows at the time of planting could reduce the incidence and severity of *Rhizoctonia* root rot and increase the yield of winter wheat.

MATERIALS AND METHODS

Laboratory study. A laboratory study was performed to examine the sensitivity of the dominant species of *Rhizoctonia* to flutolanil and tolclofos-methyl. Isolates of *R. solani* AG-8 and *R. oryzae* were grown on half-strength potato-dextrose agar medium for 7 days. Agar plugs (3 mm in diameter) were removed and placed on potato-dextrose agar media amended with flutolanil or tolclofos-methyl at rates of 0, 0.5, 1, 5, 10, and 50 $\mu\text{g a.i./ml}$. Measurements of radial growth rates were made daily.

Field studies. Three experiments were performed on two fields at the Columbia Basin Agricultural Research Center at Pendleton during 1988-89. The climate and soils at the Center are typical of those for Pacific Northwest Agronomic Zone 2 (5). The Center is in a 430-mm precipitation zone 13 km NE of Pendleton, with a Walla Walla silt loam (coarsely mesic Typic Haploxeroll) that is deep (>100 cm to hardpan), is well drained, and has a surface horizon pH (in 0.01 M CaCl₂) of 5.3-5.6.

One experiment was performed on a field that had a long history of winter wheat/fallow rotation, using a conventional tillage system (twisted-shank chisel followed by a chisel with sweeps) that buries most plant residue. Weed control was accomplished by three passes of a rod weeder during the fallow season, with the last weeding occurring 1 wk before planting, on the same day that inoculum was applied (described below). Fertilizer delivering 90 kg N/ha and 5 kg S/ha was broadcast-applied during September, 1988. Weed control during the experiment consisted of a single application of bromoxynil + MCPA (each at 0.561 kg/ha, as "Bronate") during the spring. The field is naturally infested with *R. solani* (AG-8 and other intraspecific groups), *R. oryzae*, *Pythium* spp., *G. g. var. tritici*, *P. herpotrichoides*, *Pratylenchus thornei* Sher & Allen, and several foliar pathogens. Wheat grown in this field typically has a low to very low incidence of root rot and a low incidence of eyespot.

Two experiments were performed on a field that had 3 yr of annual no-till spring barley. Experiments differed in planting equipment and placement of the fungicide + fertilizer mixture. Weeds were managed by periodic applications of glyphosate ("Roundup"), chlor-sulfuron ("Glean"), dicamba ("Banvel"), or bromoxynil + MCPA. Chlorsulfuron (0.017 kg/ha) and a fertilizer top-dressing delivering 110 kg N/ha and 6 kg S/ha were applied after the 1988 barley crop was harvested. Inoculum of the pathogen (described below) was placed into the soil 2-7 days before winter wheat was planted, during October of 1988. Bromoxynil + MCPA (each at 0.561 kg/ha) was applied for weed control during the spring. This field is infested with the same pathogens listed for the conventionally tilled field, but the incidence and severity of root diseases are typically higher. Root damage occurs from a combination of *R. solani*, *R. oryzae*, *Pythium* spp., and *G. g. var. tritici*, with the dominant damage being caused by *R. solani* AG-8. These root rots are typically expressed as patches of stunted plants that are more severely affected than the overall plant population in a field.

Inoculum of *R. solani* AG-8 was placed in the soil to increase the unifor-

mity of this pathogen's spacial distribution over each experimental area. Autoclaved millet seeds colonized by the isolate examined in the laboratory study were inserted into the soil by multiple passes of a double disk seed drill. The inoculum was placed 7-8 cm deep in rows 8 cm apart. It was applied 2-7 days before planting, and at the rates of 30 and 6 kg/ha for the tilled and no-till fields, respectively. This delivery rate provided ca. 60 and 12 pathogen foci per meter of row.

The soft white winter wheat cultivar Stephens was planted at the rate of 90 kg/ha in all experiments. This rate delivers ca. 60 seeds per meter of row. The seed was commercially treated with carboxin + thiram ("Vitavax 200") and lindane for suppressing seedling diseases and wire-worms (Coleoptera:Elateridae), respectively. These compounds may delay the colonization of seminal roots by the above-mentioned pathogens, but they do not have the persistence to control *Rhizoctonia* root rot (14).

Fungicide treatments consisted of banded applications of flutolanil (SN 84364 50W), tolclofos-methyl ("Rizolex 50W") or metalaxyl ("Apron 25W"), each applied at the rates of 75 (1X) and 150 (2X) g a.i./ha. These fungicides were delivered into soil by adsorbing their wettable powder formulations onto fertilizer granules. The fertilizer had an N-P-K-S-Fe ratio of 7-7-7-11-11 (Best Chem. Co.'s "Super Iron Plant Food" for lawns and ornamentals; manufactured by J. R. Simplot Co., Lathrop, CA). Uniformly sized granules were collected by sieving. The fraction used in this study passed through a 9-mesh screen and was retained by 8-mesh. The fertilizer plus fungicide mixture was applied at the rate of 11 kg N/ha (ca. 125 granules per meter of row) in all experiments. A fertilizer only (without fungicide) and a nonfertilized treatment were used as double controls. The deeply convoluted microtopography of the fertilizer granule provided a high level of adsorption and/or entrapment for the fungicide. In preliminary mixing tests, the dry mixture process was as efficient as wet mixtures for accomplishing a complete adherence of the fungicide to the fertilizer and for preventing separation of the two ingredients during subsequent handling.

Disease assessments were restricted to a single sampling during the spring, when plants were at Haun growth stage 7-8 (8). Twenty seedlings with intact root systems were dug from three randomly selected positions in each plot. The plants were rinsed to remove adhering soil from the root systems, and were evaluated for incidence and severity of all visible diseases. Eyespot was quantified as the percentage of tillers containing lesions that penetrated into the culm beneath the leaf sheath. Take-all was rated as the per-

centage of seminal root main axes affected by characteristic blackening of the root cortex or vascular system. *Rhizoctonia* root rot causes a light to dark brown cortical rot, which then intensifies to the point of severing the root, usually with a characteristic "spear point" created by the extension of the vascular system beyond the margins of cortical rot. These symptoms were evaluated separately for the seminal and coronal root systems. The *Rhizoctonia* root rot ratings for seminal roots were as follows: 0 = no lesions, 1 = lesions on <25% of first-order and <50% of second-order lateral branches, 2 = lesions on 25-50% first-order and >50% second-order lateral branches, 3 = lesions on >50% of first-order lateral branches, 4 = lesions on 1-2 main axes, and 5 = lesions on 3 or more main axes. For coronal roots the ratings were as follows, based on percentages of main root axes with lesions: 0 = none, 1 = <25%, 2 = 26-50%, 3 = 51-75%, and 4 = >76%.

Periodic isolations were made from symptomatic roots. Root segments were washed under running water for 3 hr and then, without surface-disinfection, plated onto 2% water agar amended with 50 µg rifampicin per milliliter, and then emerging fungal isolates were transferred onto 0.5 strength potato-dextrose agar. Grain yields were measured after plants matured in late July, by threshing plants in all 5 rows of each plot.

Conventional tillage experiment. The experiment was designed as a randomized complete block design with 6 replications for each of 10 treatments. Treatments included flutolanil, metalaxyl, and tolclofos-methyl at 1X and 2X rates; mixtures of metalaxyl (2X) with either flutolanil (2X) or tolclofos-methyl (2X); and the fertilizer and no-fertilizer controls. Plots measured 1.5 × 14 m and contained 5 rows of plants spaced at 25-cm intervals. Wheat seed was planted 2.5 cm deep into dry soil on 7 October 1988, using a 3-m-wide Great Plains drill equipped with 11 double-disk openers. The center opener was blocked, and different treatments were delivered from each half of the drill during each pass through the experimental area. The fungicide and fertilizer treatments were placed in bands 1.5 cm below the seed. Delayed autumn rainfall caused seedling emergence to be delayed until November 22. Seedlings were collected for assessments of diseases on April 25, 1989.

No-till experiment. With few exceptions, this study was identical to that described for the conventional tillage experiment. Ten replicates were used for each of the 10 treatments. The plot length was 30 m and the planting date was 14 October. Seedlings emerged on 26 November 1988, and root rot ratings were performed on 7 June 1989.

Fungicide placement experiment. A 3

× 3 factorial design with 8 replicates was used to investigate 3 fungicide placements for 3 fungicide treatments. Each plot measured 3 × 30 m. The fungicide treatments included the carrier fertilizer

without fungicide or with flutolanil or tolclofos-methyl, each at the 2× rate. Two of the three placement systems were performed with a 3-m-wide experimental model of the Yielder no-till drill. The drill

is designed with paired rows spaced 12 cm apart, with 38 cm between pairs. The fungicide and/or fertilizer combinations were placed either 1.5 cm below the seed (2.5 cm deep) in each row or were deep banded between the paired rows (6 cm below and 6 cm beside the seed placement). The Great Plains drill was used as the third placement system, to deliver the treatments 1.5 cm below the seed, as described for the conventional tillage experiment. Wheat seed was planted on 18 October 1988, and seedling emergence occurred on 30 November. Root rot ratings were performed on 9 June 1989.

Table 1. Influence of fungicides and their fertilizer carrier on diseases and yield of winter wheat planted with a Great Plains no-till drill

Treatment and rate (g/ha)	Rhizoctonia root rot ^a				Grain yield (kg/ha)
	Seminal roots	Coronal roots	Take-all (% roots)	Eyespot (% tillers)	
Conventional tillage					
Nonfertilized control	3.6	3.4	9.0	14.4	5,911*
Fertilizer control ^b	3.6	3.2	10.3	12.2	6,665
Flutolanil (75)	3.0	2.8	6.6	8.9	6,502
Flutolanil (150)	1.8*	2.6*	6.5	8.9	6,706
Metalaxyl (75)	3.3	3.1	10.6	2.1	6,583
Metalaxyl (150)	3.3	3.2	3.9*	7.7	6,543
Tolclofos-methyl (75)	2.0*	2.7*	9.1	5.6	6,563
Tolclofos-methyl (150)	2.0*	2.7*	7.2	5.6	6,564
Metalaxyl + flutolanil (150 + 150)	2.0*	2.9	8.0	16.7	6,685
Metalaxyl + tolclofos-methyl (150 + 150)	2.3*	3.0	6.7	7.8	6,767
Significance of F	0.01	0.08	0.04	NS ^c	0.01
LSD (0.05)	0.8	0.5	3.9	...	412
No-till					
Nonfertilized control	1.5*	1.0*	0.4	6.7	4,952*
Fertilizer control ^b	2.6	1.8	0.5	15.6	5,128
Flutolanil (75)	1.3*	1.4	0.6	1.1	5,235
Flutolanil (150)	1.4*	1.1*	0.3	11.1	5,190
Metalaxyl (75)	1.8*	2.0	0.7	7.7	5,074
Metalaxyl (150)	1.8*	1.7	0.7	12.2	5,188
Tolclofos-methyl (75)	1.6*	1.4	0.7	5.5	5,242
Tolclofos-methyl (150)	1.8*	1.7	0.9	8.9	5,229
Metalaxyl + flutolanil (150 + 150)	1.7*	1.6	0.4	7.7	5,190
Metalaxyl + tolclofos-methyl (150 + 150)	1.5*	1.2*	0.6	13.3	5,161
Significance of F	0.01	0.01	NS ^c	NS	0.01
LSD (0.05)	0.6	0.5	154

^a Refer to text for root rot indices for seminal and coronal roots.

^b The fertilizer control (11 kg N/ha, as 7-7-7) serves as the basis for comparison of all treatments since all fungicides were applied on this carrier.

^c NS = not significant ($P > 0.10$). Asterisk denotes values that differ significantly from the nontreated control.

Table 2. Influence of fungicides and their placement with two no-till drills on diseases and yield of winter wheat

Treatment and rate (g/ha)	Rhizoctonia root rot ^a				Grain yield (kg/ha)
	Seminal roots	Coronal roots	Take-all (% roots)	Eyespot (% tillers)	
Yielder drill					
Placement below seed					
Fertilizer control	3.1	2.4	6.8	30.2	6,281
Flutolanil (150)	2.8	2.1	2.7	33.3	6,271
Tolclofos-methyl (150)	2.7	2.3	4.8	25.0	6,369
Significance of F	NS ^b	NS	NS	NS	NS
Placement Between Rows					
Fertilizer control	2.9	2.1	2.8	36.7	6,391
Flutolanil (150)	2.8	2.2	2.0	38.3	6,389
Tolclofos-methyl (150)	2.8	2.3	3.2	26.7	6,379
Significance of F	NS	NS	NS	NS	NS
Great Plains drill					
Placement below seed					
Fertilizer control	2.8	2.1	1.9	33.3	6,423
Flutolanil (150)	2.0	1.6	1.4	22.0	6,336
Tolclofos-methyl (150)	2.2	1.5	1.6	23.2	6,376
Significance of F	NS	NS	NS	NS	NS

^a Refer to text for root rot indices for seminal and coronal roots.

^b NS = not significant ($P > 0.10$).

RESULTS AND DISCUSSION

Both *Rhizoctonia solani* AG-8 and *R. oryzae* were highly sensitive to flutolanil and tolclofos-methyl. Radial growth of these fungi on fungicide-amended agar medium was reduced by more than 50% at fungicide concentrations of 0.5 µg/ml. Growth of *R. solani* AG-8 was completely inhibited (EC₁₀₀) at concentrations of 0.5 and 1.0 µg/ml for flutolanil and tolclofos-methyl, respectively. This appears to be the first report of the sensitivity of *R. solani* AG-8 to flutolanil. The EC₁₀₀ against *R. oryzae* occurred between 1 and 5 µg/ml for both fungicides but the precise concentrations were not determined. These results indicate that the sensitivity of Pacific Northwest isolates of *R. oryzae* to flutolanil is much greater than the sensitivity (EC₈₀ = 29 µg/ml) reported for Japanese isolates (1). It remains to be determined whether this is a characteristic of a broader spectrum of isolates of *R. oryzae* from cereals, rice, and turfgrasses produced elsewhere in North America.

Pathogenic fungi isolated from affected roots included *Fusarium culmorum* (Smith) Sacc., *F. graminearum* Schwabe, *R. solani* AG-8, *G. g.* var. *tritici*, and species of *Pythium* (data not presented). *F. culmorum* and *F. graminearum* were most frequently isolated but are not known to cause the types of cortical and stelar root rots observed; they were isolated from roots that exhibited symptoms of root rot as well as from roots with no visible infections. *R. solani* was isolated regularly from roots at all three locations, although at a lower frequency than for the fusaria. *G. g.* var. *tritici* and species of *Pythium* were isolated infrequently (0–7% of roots). Root rots observed in this study were characteristic of those caused by species of both *Rhizoctonia* and *Pythium*. Roots with blackened vascular tissue or darkly pigmented ectotrophic “runner” hyphae typically yielded *G. g.* var. *tritici*.

Fungicides placed below the seed at planting reduced *Rhizoctonia* root rot in both the conventional and no-till management systems (Table 1). Under conventional tillage both flutolanil and tolclofos-methyl reduced the root rot

index on seminal roots. Only the high rate of flutolanil reduced root rot on both the seminal and coronal roots. Flutolanil was also noted for its ability to suppress *Rhizoctonia* root rot in the no-till experiment. This is particularly important because these measurable reductions in root rot severity occurred on both seminal and coronal roots and were made nearly 8 mo after treatments were applied. The level of *Rhizoctonia* root rot control with this placement technique exceeded that in previous studies performed with seed treatments (14), thus supporting an earlier report of this finding by Ballinger and Kollmorgen (3). Unfortunately, our results indicate also that variable control may occur in response to the seed drill used. Although there were no statistical differences in *Rhizoctonia* root rot in the experiment comparing fungicide placement systems (Table 2), the severity of *Rhizoctonia* root rot was decreased when fungicides were delivered with the Great Plains drill but not with the Yelder drill. Additionally, our results indicate that the level of disease control in each of these experiments was insufficient to have an impact on grain yield.

The severity of *Rhizoctonia* root rot was affected by the presence or absence of starter fertilizer in the no-till but not in the conventionally tilled soil (Table 1). In the no-till system the starter fertilizer increased ($P = 0.05$) the incidence of *Rhizoctonia* root rot. This observation conflicts with previous reports. Experiments conducted in nitrogen-depleted soils in Australia have demonstrated that increasing amounts of nitrogen fertilizer can be used to reduce the severity of *Rhizoctonia* root rot (9). Fertilizers applied before planting have had no influence on the severity of *Rhizoctonia* root rot in irrigated winter wheat produced on nitrogen-depleted sandy soils in Oregon (13). The soils used for our

study are not depleted in nitrogen and were treated with moderately high rates of fertilizer 1 mo before placing the starter fertilizer below or beside the seed at planting. The ability of starter fertilizer to increase the severity of *Rhizoctonia* root rot in our no-till study is important and deserves further investigation. This is particularly pertinent in that fertilizers are, by definition, applied at planting during the no-till process.

Application of a starter fertilizer at planting also caused a significantly positive impact on grain yield in both of the experiments where this could be evaluated (Table 1). The yield advantage from starter fertilizer was 13% and 4% in the conventional and no-till systems, respectively.

Flutolanil and tolclofos-methyl did not significantly reduce the incidence of take-all or eyespot (Tables 1 and 2), and the results of these tests were highly variable. It was clear, however, that in the seven instances where tolclofos-methyl was applied individually on a fertilizer carrier, the incidence of eyespot was considerably less than for the fertilized control treatment. Likewise, in the experiments where take-all affected more than 2% of the roots, flutolanil consistently reduced the incidence of this disease. However, as stated earlier, when compared to the fertilized carrier alone, the addition of fungicides failed to improve grain yields in each of these experiments. We conclude that flutolanil and tolclofos-methyl are not effective for increasing yields of winter wheat through management of root and culm diseases.

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