Evaluation of Benomyl and Propiconazole for Controlling Sheath Blight of Rice Caused by *Rhizoctonia solani*

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ABSTRACT

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Twenty-one isolates of *Rhizoctonia solani* were sensitive to benomyl (mean EC_{50} 0.5 μg a.i./ml) and propiconazole (mean EC_{50} 5.2×10^{-2} μg a.i./ml) in vitro. Radial growth rates for all isolates on unamended PDA averaged 0.96 mm/hr. In field trials, propiconazole applied twice or propiconazole followed by benomyl significantly reduced disease severity and increased yields in three of six studies during 1984–1985 on the semidwarf rice cultivar Lemont. Yield response was related to incidence of sheath blight at the panicle differentiation growth stage. Benomyl applied twice did not significantly reduce disease or increase yields. An economic return from propiconazole/propiconazole or propiconazole/benomyl could be anticipated when more than 5% diseased tillers were observed at the panicle differentiation growth stage.

Additional key word: fungicides

Sheath blight, caused by *Rhizoctonia solani* Kühn, anastomosis group 1, sasakii form (1), has become the major rice disease in Texas and other south central long-grain rice production areas of the United States since the late 1960s (11,23). Producers continue to adopt production practices that favor the development of sheath blight, including 1) use of rotation crops susceptible to *R. solani*, particularly soybean (19) and sorghum (18); 2) shortened rotation intervals resulting from economic constraints on land ownership; 3) shallow disk cultivation instead of the once widely used practice of moldboard plowing as a consequence of escalating fuel and equipment costs; and 4) use of very susceptible semidwarf cultivars planted in dense stands under high nitrogen fertilizer regimes (14,24). Chemical control represents the most significant practice implemented in the last decade to limit yield loss caused by *R. solani* in rice production areas of the southern United States (10,11,20,24).

Benomyl (Benlate) has been used in Texas to control a broad range of rice diseases since its registration in 1975. Initially, control programs were designed to limit yield loss caught by the rice blast fungus, *Pyricularia oryzae* Cav. (25); however, the identification and deployment of broadly based resistance has resulted in the virtual elimination of blast as a significant production problem on the long-grain rice presently grown in Texas (13,16). Federal registration of benomyl for control of stem rot (*Sclerotium oryzae* Catt.) remains included in the labeling despite field research that shows it to be ineffective (8).

In 1977, the Environmental Protection Agency (EPA) approved Section 24c Special Local Need (SLN) registration of benomyl to allow for further uses: reduced use rates (0.28 vs. 0.56 kg a.i./ha); earlier timing of first applications (panicle differentiation vs. boot stage); and additional target diseases adding sheath rot (*Acrocylindrium oryzae* Sawada), narrow brown leaf spot (*Cercospora oryzae* Miyake), panicle blight (*Cercospora* spp.), leaf smut (*Entyloma oryzae* H.&P. Sydow), and sheath blight (*R. solani*). The SLN was modified in 1981 to delete panicle blight when Marchetti (12) demonstrated this to be a physiological disorder.

A major change in the use pattern of foliar fungicides in rice occurred in 1981, when triphenyltin hydroxide (TPTH) was registered under Section 18 of the Federal Insecticide, Fungicide, and Rodenticide Act (FIFRA) for use in control of *R. solani* and *R. oryzae* Ryker & Gooch. *R. oryzae* causes the sheath spot disease of rice (21). Emergency exemptions were granted for three consecutive years (1981–1983), and producers rapidly adopted a cost-effective fungicide program that included TPTH (0.6 kg a.i./ha) applied at panicle differentiation for stem and sheath diseases followed by benomyl (0.28 kg a.i./ha) applied at heading. TPTH has been shown to be highly effective in controlling rice diseases caused by *Rhizoctonia* spp. (10,11,20,24) and also stem rot (8,20).

In 1984, a request for a fourth-year exemption for TPTH was denied by EPA, citing missing toxilogical and oncogenological data. An exemption request for the use of propiconazole to control sheath blight was also denied in 1984 by EPA, citing the "marginal nature of sheath blight as an emergency situation" and the existing registration of benomyl (EPA SLN No. TX-770004) for control of sheath blight.

In 1984, we began studies to evaluate the relative efficacy of benomyl and propiconazole in controlling *R. solani* in rice. We considered the lack of producer satisfaction with benomyl control programs as potentially resulting from the development of benomyl-insensitive strains, ineffective application timings, or the heightened susceptibility of newly adopted semidwarf cultivars. Since its introduction in 1982, the semidwarf cultivar Lemont has accounted for 1,200 (0.5%), 7,000 (5.3%), 22,000 (13.5%), and 69,000 (55.0%) ha planted in successive years. In 1986, it is estimated that 70% of the rice planted in Texas is of this cultivar (R. K. Jones, *unpublished*).

An additional objective of this study was to assess disease incidence immediately before fungicide application and evaluate the potential use of this information to improve the economic returns from foliar fungicide programs in rice. Integrating treatment thresholds with cultural practices and field selection based on soil assays of preplant inoculum (2,3) could increase the range of producer options for minimizing loss caused by sheath blight in long-grain rice production systems.

MATERIALS AND METHODS

Fungicidal activity in vitro. Twenty-one isolates representing collections from 15 commercial rice fields in six Texas counties were tested for sensitivity to benomyl and propiconazole in vitro. Isolates were obtained from sclerotia of *R. solani* collected from field soil (2,3). Identification consisted of sclerotial morphology and color; hyphal branching habit and colony growth characteristics on water agar (WA) and potato-dextrose agar (PDA); and the ability of each isolate to produce typical disease symptoms on both rice and soybean in

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greenhouse pathogenicity tests. In addition, selected isolates were tested for their ability to anastomose with AG-1 tester isolate R-43 (provided by P. S. Gunnell and E. E. Butler, University of California, Davis).

Dilutions of each fungicide were prepared by mixing appropriate amounts of each fungicide (based on active ingredient) in 5 ml of 95% ethanol. Equal volumes of ethanol containing diluted fungicides were added to sterile, cool PDA to achieve concentrations of 0.01, 0.1, 1, and 2 μg a.i./ml of PDA for benomyl and 0.01, 0.1, 1.0, 10, and 100 μg a.i./ml of PDA for propiconazole. A zero concentration treatment was prepared for each isolate × fungicide combination and contained 1% (v/v) of 95% ethanol to ensure equivalent ethanol concentrations in all treatments. Fungicide-amended PDA was dispensed aseptically into 9-cm-diameter plastic petri dishes (about 20 ml per dish).

Mycelial plugs (7 mm diameter) were cut from the margins of actively growing 2-day-old PDA cultures of each isolate and inverted in the centers of fungicide-amended and unamended PDA plates, with three replicate plates for each isolate-fungicide combination. Plates were incubated (inverted) for 36 hr at 28 C in the dark. Colony diameters were measured twice (random and right angles) and adjusted for the diameter of the inoculating plug. The effective fungicide concentration was determined by interpolating plots of growth inhibition probits for concentrations that give 50% (EC_{50}) or 90% (EC_{90}) growth inhibition for each fungicide (4).

Field efficacy. Field trials were conducted to evaluate the efficacy of benomyl and propiconazole for control of sheath blight at six locations in 1984 (four sites) and 1985 (two sites). Studies were conducted in commercial fields on the semidwarf rice cultivar Lemont. Plots were 2.2 × 7.3 m with 0.6-m alleys and replicated four times in a randomized complete block design. Seeding rates, planting dates, total nitrogen applied, and insecticidal control programs varied from location to location. Disease incidence was determined by examining 500 tillers (25 tillers at each of 20 randomly chosen sites within the test area) for symptoms of sheath blight at the panicle differentiation growth stage immediately before initial fungicide applications.

Fungicides were applied in 290 L/ha water at the panicle differentiation (PD), boot (B), or heading (H) stage of plant growth (22) with a Solo Mist Blower (Model 410, Forestry Suppliers, Inc., Jackson, MS). Treatments included: 1) benomyl (Benlate 50WP) at 0.28 kg a.i./ha applied at PD and H (registered rate and timing), 2) propiconazole (Tilt 3.6 EC) at 0.14 kg a.i./ha applied at PD followed by benomyl (0.28 kg a.i./ha) at H, and 3) propiconazole (0.11 kg a.i./ha) applied at PD and B. Untreated plots were included as controls.

Sheath blight was assessed 2 wk before harvest using the International Rice Research Institute visual index of canopy damage based on a linear scale of 0–9 (7). Plots were harvested by hand-clipping panicles from two 0.85-m^2 areas, one from each end of the plot, avoiding edges. Harvested panicles from each subplot were separately placed in paper bags and air-dried for 2 wk in a greenhouse before threshing for grain yield data.

Levels of economic return were determined using the 1984–1985 target price for long-grain rice of $0.26/kg. The cost for fungicides was calculated using $59.52/kg a.i. for benomyl and $215.56/kg a.i. for propiconazole. Custom aerial application was included in the program cost estimate at $10.38/ha. This estimate included application in 65.4 L of water per hectare with flagmen or automatic field markers.

RESULTS

Fungicidal activity in vitro. Mycelial growth inhibition increased with increasing concentrations of either fungicide. The mean EC_{50} for 21 isolates of *R. solani* tested against benomyl was 0.50 μg a.i./ml. The mean EC_{50} to propiconazole was 0.05 μg a.i./ml. The effective concentrations of benomyl and propiconazole that inhibited 90% growth were 1.6 and 7.9 μg a.i./ml, respectively. The overall response of the isolate means are illustrated in Figure 1.

There were significant ($P = 0.01$) differences in mean mycelial growth measurements among isolates at all concentrations tested including the zero concentration. The mean growth rate for all isolates on unamended PDA was 0.96 mm/hr. Standard errors are given in Table 1. Significant ($P = 0.01$) main effects and interactions of isolate × concentration were observed for both fungicides tested.

Spearman's coefficient of rank correlation was used to compare the relative growth of isolates on amended vs. unamended agar. Significant rank correlations ($P|t| < 0.01$) were observed between isolates grown on agar amended with 0.01, 0.1, and 1 μg a.i./ml benomyl compared with growth on unamended PDA. This suggests that differences in observed growth on benomyl-amended agar may be a function of differences in the growth rates of the isolates in the absence of benomyl. Rank correlation coefficients among isolates for growth on propiconazole-amended vs. unamended PDA were all nonsignificant ($P\ |t| > 0.05$), whereas rank coefficients for isolate growth among propiconazole-amended PDA (0.01 vs. 0.1, 0.1 vs. 1.0, and 0.01 vs. 1.0 μg a.i./ml) were all significant ($P|t| < 0.05$). The differences in observed growth on propiconazole-amended agar may not be a function of the growth rate of the isolates in the absence of propiconazole.

Propiconazole was nearly 10-fold more effective than benomyl in inhibiting radial growth of *R. solani* in vitro at biologically relevant concentrations (EC_{50}). Differential sensitivity to benomyl was observed among the isolates examined in this study but was relatively small and appeared to be correlated to the growth rate of the isolates in the absence of the fungicide. All isolates examined in this study had individual EC_{50} values of <1 μg a.i./ml for benomyl and would be considered sensitive (17).

Field efficacy. Propiconazole or propiconazole/benomyl sequential sprays significantly reduced sheath blight and increased yields ($P = 0.05$) at three of six locations tested (Table 2). Response was related to the amount of disease before

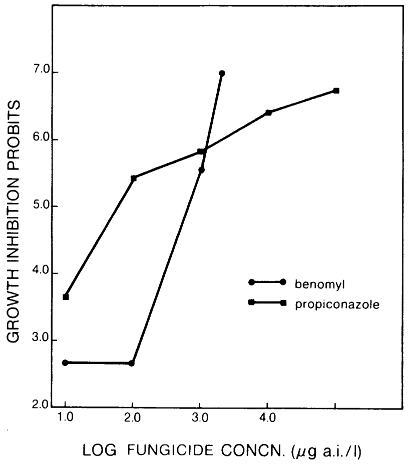

Fig. 1. Mean radial growth inhibition (probits) for 21 isolates of *Rhizoctonia solani* grown on potato-dextrose agar amended with various concentrations of benomyl and propiconazole after 36 hr at 28 C.

Table 1. Influence of benomyl and propiconazole concentration on radial growth of *Rhizoctonia solani* in vitro

Fungicide concentration (μg a.i./ml)	Growth[a] (mm)	Growth inhibition (%)
Benomyl		
None	70.6 (±0.6)	···
0.01	69.5 (±0.8)	1
0.1	69.9 (±0.7)	1
1.0	20.3 (±1.1)	71
2.0	1.2 (±0.2)	98
Propiconazole		
None	67.3 (±0.7)	···
0.01	62.0 (±2.6)	9
0.1	22.9 (±1.0)	66
1.0	14.3 (±0.6)	79
10.0	5.3 (±0.4)	92
100.0	2.7 (±0.3)	96

[a] Growth of 21 isolates on PDA after 36 hr of dark incubation at 28 C ± standard error of mean. Values represent average of two measurements per plate, with three replicates.

application. A maximum yield increase (1,229 kg/ha) was obtained when propiconazole was applied sequentially (0.11 kg a.i./ha at PD + 0.11 kg a.i./ha at B) at an incidence of 16.3% diseased tillers ($Y_3 = -194 + 174X - 5.3X^2$, $R^2 = 0.99$). This increase is significantly greater than the level of economic return (390 kg/ha) at $P = 0.01$ ($t = 13.28$, 2 df). An economic return can be anticipated with this treatment when preapplication disease incidence exceeds 3.8% tillers at the PD growth stage (Fig. 2).

The maximum yield increase (850 kg/ha) obtained when propiconazole and benomyl were applied sequentially (0.14 kg a.i./ha at PD + 0.28 kg a.i./ha at H) occurred at 13.8% diseased tillers ($Y_2 = -254 + 160X - 5.8X^2$, $R^2 = 0.98$). This increase was significantly greater than the level of economic return (350 kg/ha) at $P = 0.01$ ($t = 6.62$, 3 df). An economic return can be anticipated with this treatment when preapplication disease incidence exceeds 4.5% tillers at the PD growth stage.

The maximum yield increase (496 kg/ha) obtained when benomyl was applied sequentially (0.28 kg a.i./ha at PD + 0.28 kg a.i./ha at H) occurred at 13.5% diseased tillers ($Y_1 = -210 + 105X - 3.9X^2$, $R^2 = 0.87$). This increase was not significantly greater than the level of economic return (206 kg/ha) at $P = 0.05$ ($t = 2.31$, 3 df).

The intercepts for propiconazole/propiconazole (−194 kg/ha) and propiconazole/benomyl (−254 kg/ha) were significantly less than zero ($P = 0.05$). Yield reduction associated with propiconazole treatments in the absence of disease suggests possible phytotoxicity. The intercept for benomyl/benomyl was not significantly different from zero ($P = 0.05$).

From these results, we conclude that benomyl/benomyl should not be applied at this rate and timing for control of sheath blight because it is unlikely to provide an economic return. Propiconazole/benomyl is likely to provide an economic return when 5–10% of the tillers are diseased at PD. Propiconazole/propiconazole, applied when 10–20% of the tillers are diseased at the PD stage, will probably show an economic return. It is not possible at present to advise in situations where disease incidence at PD exceeds 25%. The downward trend in yield response (Fig. 2) and the trend toward reduced control as evidenced by the disease index for treatments at site 6 vs. site 5 (Table 2) suggest that higher rates or increased application frequency may be required to achieve economic control where disease incidence at PD exceeds 25%. Extrapolating treatment thresholds from quadratic response curves for use in locations other than those tested is statistically invalid. However, the relationship between yield response (Y) and preapplication disease incidence (X) in the range of 0–15% diseased tillers could be fit with the linear terms of the quadratic equations for each of the three fungicide treatments. Solving the linear terms for X at the level of economic return for each of the treatments resulted in thresholds of 6.9, 6.4, and 5.4%

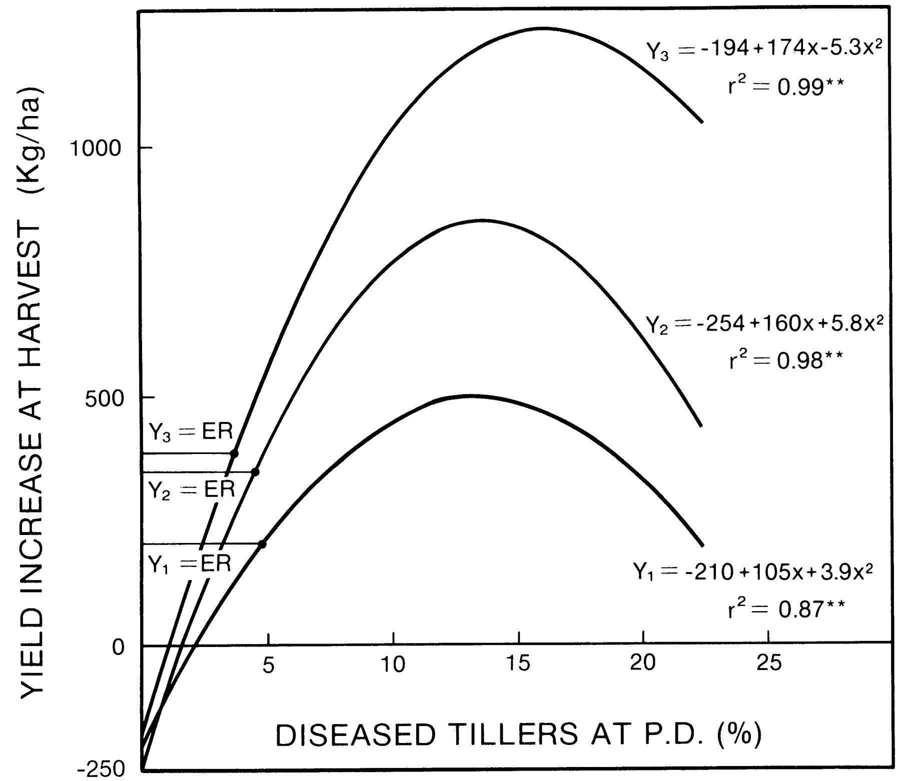

Fig. 2. Yield increase in kg/ha (Y) over untreated control at various levels (X) of percent sheath blight-diseased tillers at the panicle differentiation (PD) growth stage (before fungicide application). Break-even cost of fungicide program and application expressed as level of economic return (ER).

Table 2. Effectiveness of fungicide applications on sheath blight disease index and yield of rice at six locations in the Texas Upper Gulf Coast during 1984 and 1985[a]

	Site 1		Site 2		Site 3		Site 4		Site 5		Site 6	
Treatment	Disease index[b]	Yield[c] (kg/ha)	Disease index	Yield (kg/ha)	Disease index	Yield (kg/ha)	Disease index	Yield (kg/ha)	Disease index	Yield (kg/ha)	Disease index	Yield (kg/ha)
Benomyl/benomyl	0.0	6,825	0.3	7,983	3.0	7,089	3.3	5,975	4.8	8,710	7.0	5,911
Propiconazole/benomyl	0.0	6,820	0.0	7,921	2.3	7,222	2.5*	6,196*	3.3*	9,056*	6.5	6,158
Propiconazole/propiconazole	0.0	6,857	0.0	7,989	2.0*	7,372	–[d]	–[d]	2.8**	9,398**	5.3*	6,767*
No fungicide	0.0	7,070	0.3	7,915	3.5	7,175	3.8	5,718	5.3	8,177	6.8	5,732
Disease incidence[e] (% diseased tillers)	0.0		1.2		2.8		5.8		14.4		22.3	

[a] Tests were conducted at six locations, sites 1, 3, 4, and 6 in 1984 and sites 2 and 5 in 1985. All tests were conducted in commercial fields using a randomized complete block design with four replicates on the rice cultivar Lemont. Results in the same column are statistically different from the untreated controls (* = $P = 0.05$ and ** = $P = 0.01$).

[b] Disease index on a scale of 0–9, where 0 = no disease and each unit increase represents an additional 10% damage to the canopy (flag leaf, f-1, and f-2) and 9 = 90–100% canopy damage. Disease assessed 2 wk before harvest.

[c] Yields expressed as kg/ha rough rice.

[d] Missing data.

[e] Disease incidence before fungicide application was determined by examining 500 rice tillers (25 tillers at each of 20 randomly chosen sites) within the test area for symptoms of sheath blight at the panicle differentiation growth stage.

diseased tillers for Y_1, Y_2, and Y_3, respectively.

DISCUSSION

The current status of foliar fungicide registration on rice in the United States allows for chemical use patterns that are ineffective in providing economic control of *R. solani* on newly released semidwarf cultivars. Failure to achieve control in commercial fields treated with registered rates of benomyl are not the result of insensitivity among the natural populations of *R. solani*. These failures may be a function of inadequate rates at registered use patterns, the increased susceptibility of semidwarf cultivars, or an interaction of both factors. Marchetti (14) reported twofold greater reductions in yield and fivefold greater reductions in milling quality between semidwarf and standard rice varieties with equivalent levels of sheath blight.

Higher rates of benomyl (0.56 kg a.i./ha) are federally registered for application on rice applied at B and H for control of blast and stem rot. However, yield differences between this use pattern and untreated plots evaluated in 1985 at sites 2 and 5 (R. K. Jones, *unpublished*) were 140 and 276 kg/ha, respectively. These differences were not significant ($P = 0.05$).

Sheath blight is only one of many diseases that attack rice in Texas, and others were observed during this study. Propiconazole significantly reduced brown spot at all locations where it occurred (R. K. Jones, *unpublished*), although Marchetti (15) recently demonstrated that significant yield responses were not observed when moderately resistant varieties were inoculated with *B. oryzae* and treated with propiconazole. The Lemont cultivar shows moderate resistance to *B. oryzae* (24).

Yield loss associated with stem rot (*Sclerotium oryzae*) on the new semidwarf cultivars is relatively uncharacterized in Texas and other long-grain rice production areas of the south central United States. Current recommendations for controlling stem rot include rotation and inversion tillage (24). Benomyl, though labeled for control of stem rot in the United States (excluding California), has been shown to be ineffective in reducing disease incidence or increasing yields in the absence of other rice diseases (8). Critical evaluation of propiconazole for control of stem rot is needed.

Federal registration of propiconazole is currently awaiting EPA approval. Proposed use patterns on rice for control of sheath blight include applications of 0.11 kg a.i./ha at PD and B or 0.14 kg a.i./ha at PD followed by an application of benomyl (0.28 kg a.i./ha) at heading. These use patterns may provide an economic advantage over sequential applications of benomyl (0.28 kg a.i./ha) alone. The potential for yield reductions in the absence of disease needs to be evaluated further.

The assessment of disease incidence at key growth stages shows promise as a decision-making tool to improve the economic return from foliar fungicide programs in rice. In Japan, the forecasting of sheath blight has received considerable attention (5,9). Hori (6) reported that 15–20% sheath blight incidence at the booting stage was the critical point for achieving a yield response from organoarsenical fungicides on medium-maturing rice varieties in Japan. Earlier timing of applications and lower thresholds are required on semidwarf cultivars under Texas conditions.

The vertical and horizontal development (secondary disease spread) of sheath blight on rice plants is greatly influenced by temperature and relative humidity. Lesions can expand vertically as much as 0.66 mm/hr at 100% relative humidity and 28 C on susceptible varieties but drops rapidly to 0.07 mm/hr at 84% RH (5). The shortened plant height and dense compact canopy associated with production of cultivar Lemont increases the potential for rapid disease development and yield loss (14). High humidity, warm temperatures (26–30 C), and overcast weather conditions from the boot stage through grain-fill could alter the thresholds and/or expected returns from foliar programs established under the conditions of this study.

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