

SEED TREATMENT WITH BACTERIAL BIOCONTROL AGENTS TO CONTROL HEAD BLIGHT

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ABSTRACT

Selected *Bacillus* and *Paenibacillus* strains isolated from South Dakota wheat foliage and residue have been shown to antagonize isolates of *Fusarium graminearum* causing FHB, as well as antagonizing *Pyrenophora tritici-repentis* which causes tanspot. Application of whole live cells of these bacterial biocontrol agents (BCAs) as well as application of concentrated ethyl acetate extracts of broth culture supernatants of the bacteria, have previously been shown to reduce symptoms of FHB and tanspot in greenhouse and field-plot trials. Another method of using these BCAs would be applying them to wheat seed before planting. By manipulating the bacteria to remove their walls and ensure their osmotic protection, cell wall deficient (CWD) forms of the bacteria such as protoplasts and L-forms can be produced. These CWD bacterial forms have been shown by other researchers to form putative intracellular associations with the cells of a variety of plants. It would be desirable to develop seed-treatment methods to reliably establish such associations between wheat plants and CWD-forms of bacterial BCAs, since it could afford protection against infection by *F. graminearum* without need for spray application of bacteria onto mature wheat plants. Alternatively, seed treatment with wall-bearing cells of bacterial BCAs could result in protection against FHB. In one greenhouse trial, soaking wheat seeds with wall-bearing cells of bacterial strain 1BE (*Paenibacillus lentimorbus*) resulted in 26% less FHB symptoms than soaking seeds with live protoplasts of the same bacterium. In a greenhouse trial involving tanspot, however, soaking wheat seeds with live protoplasts of *P. lentimorbus* 1BE resulted in 22% less FHB symptoms than soaking seeds with live wall-bearing cells of the same bacterium. More work is needed to explore the potential of seed treatment with bacterial BCAs to control FHB.

CONTROL OF FUSARIUM HEAD BLIGHT WITH BIOLOGICAL ANTAGONISTS

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INTRODUCTION AND OBJECTIVES

Fusarium head blight (scab) has been a serious concern for wheat producers in South Dakota for the past several years. Scab and low market prices are the two reasons most often cited by growers as they decrease the number of acres they plant to wheat. Fungicide alternatives for disease control are available on special year-to-year labeling options.

The objectives of this study were to evaluate the efficacy of various bacterial strains employed as biological control agents for their fungicidal properties for the suppression of Fusarium head blight (scab).

MATERIALS AND METHODS

The bacterial isolates from SDSU screened in this study were selected in laboratory assays where both live cells and ethyl acetate extracts were shown to antagonize growth of *Fusarium graminearum* isolate Fg 4. Isolates from SDSU are classified as strains of *Bacillus* or *Paenibacillus*. Two of these strains were applied to two spring wheat cultivars, Oxen and Ingot, planted at Brookings, South Dakota. The previous crop was soybean. No *Fusarium* inoculum was added to the sites nor was the environment modified with mist irrigation. Six microbial isolates were tested. Isolate SDSU #1-1BA is a *Paenibacillus lentimorbus*. All other strains are putative *Bacillus* sp. Bacterial treatments were applied and compared to an untreated control and a chemical control of Folicur (4 fl. oz./A), plus Induce non-ionic surfactant (NIS) (0.125%). Treatments were replicated four times and applied at Feekes 10.3-10.54 as a 10⁴ CFU/ml suspension of whole bacterial cells in nutrient broth. Ingot typically reaches anthesis was about two days later than Oxen, but all plots were flowering on early tillers when treatments were applied. Plots were evaluated for scab and leaf disease 21-28 days after treatment, about soft dough. Yield, test weight, DON, protein, and FDK measurements were taken following harvest.

RESULTS AND DISCUSSION

The Brookings environment was particularly dry in 2000. Little scab developed as a result and significant differences were not observed among most of the variables measured (Table 1). Folicur did reduce scab numerically and leaf disease significantly from the untreated control. However, at such low levels of scab, it is difficult to interpret these data. Failure to increase yield was not a reliable measurement in 2000 because of the low levels of disease. While significant differences were reported among treatments for leaf disease, the

biological control agents were not significantly different from the untreated. The biological control agents did not approach the activity of the Folicur standard.

No product significantly reduced FDK below the level observed in the control or increased protein, yield or test weight significantly higher than the control.

Nonetheless, trends observed for scab incidence and head severity may indicate that the SDSU isolates have activity in the range of Folicur. Additional testing is needed to substantiate this possible difference. These isolates are scheduled for field screening in 2001 under mist irrigation. A mist system intended for use in 2000 was not completed in time due to problems with work completion and the environment in 2000 pushing development of the crop earlier than in most years.

The levels of scab observed in South Dakota in 2000 were lower than have been observed for several years.

Table 1. Measurements of disease and yield in HRSW Scab Biocontrol Trial – SDSU Agronomy Farm, Brookings, SD, 2000.

Treatment	FHB Inc. ^b	FHB Sev. ^c	FHB Ind. ^d	Whole Plot Disease Rating ^e	FDK	Yield	Test Weight	Protein
	%	%	%		%	bu/A	lb/bu	%
Untreated	2	14.54	0.59	4.13	1	51.76	58.39	13.76
Folicur + NIS	1.25	4.29	0.19	3.31	0.5	52.15	58.54	14.14
SDSU #1- 1BA	0.75	2.63	0.07	4.06	0.75	50.47	58.57	13.79
SDSU #2- 1 BC	1.5	5.25	0.12	4	0.75	51.51	58.63	13.81
Experimental A	2.25	12.96	0.35	4.19	0.75	51.11	58.49	13.88
Experimental B	2.25	12.38	0.41	4.19	0.88	50.54	58.67	13.64
Experimental C	1	9.75	0.2	4	0.75	49.33	58.61	13.89
Experimental D	0	0	0	4.13	0.88	50.13	57.29	13.71
LSD (0.05)	NS	NS	NS	0.4	NS	NS	NS	NS

^a Data represents an average of disease impact on Oxen and Ingot.
^b % of infected heads, based on a 50 head sample.
^c % infection of blighted heads.
^d % blighted heads x % infection on blighted heads.
^e Subjective rating (0=green – 5=necrotic) of tissue necrosis in the plot at evaluation.

BIOCONTROL OF FUSARIUM HEAD BLIGHT IN BRAZIL

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INTRODUCTION

Fusarium graminearum Schw. (Teleomorph=*Gibberella zeae* Schw. Petch.) is the *Fusarium* species most frequently responsible for scab of wheat and barley in Brazil. This disease, also known as Fusarium head blight (FHB), is responsible for major losses which vary from 10% (Luz, 1984); to 54% (Picinini & Fernandes, 1994). At present, available and affordable control measures, such as resistant varieties, cultural practices and foliar fungicides, are only partially effective.

Only modest levels of resistance have been deployed in cultivars in commercial fields; the most widely grown cultivars are often most susceptible. Furthermore, the benefit of crop rotation as a control measure is reduced by the wide host range of the pathogen, especially on grasses (Costa Neto, 1976; Luz, 1982). Treatment with foliar fungicides remains the most important (Picinini and Fernandes, 1994) and recommended (Reunião da Comissão Sul-Brasileira de Pesquisa de Trigo, 2000) tool for reducing scab in Brazil, despite its shortcomings as a control measure. The use of certain effective fungicides has been restricted in some countries because application at late developmental stages, that is, during heading and flowering, can result in chemical residues in the harvested grain.

Biological control is an additional strategy that may eventually play an important role in an integrative approach to scab management of cereals.

RESEARCH IN BIOCONTROL OF FUSARIUM HEAD BLIGHT

The chronology of publications on the biocontrol of FHB from 1988 to 2000 is listed in Table 1.

Screening of microorganisms to control wheat scab was initiated in Brazil in the 80's (Luz, 1988). At the beginning, over 300 bacteria and yeasts isolated from wheat were screened in vitro against *F. graminearum*. This work was followed by that of Perondi et al. (1990a, 1990b, 1996) in which microbial strains were tested for their antagonistic action against the pathogen. Potential antagonists were selected by the funnel method (Luz, 1990) which compared the effect of individual test organisms on the radial growth of *F. graminearum*. Promising isolates were further tested in the greenhouse and in the field for their ability to control wheat scab. Individual bioprotectants significantly diminished the severity of the disease under field conditions, raising the yield of wheat between 7 and 31% when compared to nontreated plants.

The most recent published work, (Luz, 2000) showed that *Bacillus megaterium* (Embr.9790) *Bacillus subtilis* (Embr.9786) and *Paenibacillus macerans* (Embr.9770) the best isolates

significantly diminished the disease incidence and severity up to 50% and 67% respectively. The yield increase varied from 701 Kg to 818 Kg/ha.

Isolates that significantly reduced the percentage of scabby spikelets when tested under greenhouse conditions, frequently reduced disease in field trials as well, even though they may have not inhibited growth in the laboratory assays. This suggests that greenhouse evaluation for biocontrol of scab is more reliable as an early selection method than in vitro assays in large-scale screening of bioprotectants. However, since greenhouse screening requires a substantial investment in equipment and space, in vitro assays are likely to continue to be used as an initial step in selecting potential bioprotectants.

From 1988 up to now, thousands of microorganisms have been tested for scab control in Brazil. The biodiversity of microorganisms that show potential for managing the disease comprises the species described in Table 2.

Some other workers outside of Brazil have been investigating antagonists to control FHB (Khan et al., 1998;1999, Boeham et al., 1999; Luo & Bleakley, 1999; Schisler et al., 1999; Stockwell et al., 1997,1999,2000), under greenhouse or field conditions. Some strains have reduced the FHB severity and significantly reduced vomitoxin contamination in grains (Stockwell et al., 1997,2000).

The constraints to the application of bioprotectants to the ears of wheat and barley at flowering such as the timing of application, inoculation technology, physiological state of the organisms, spike colonization, survival of the organisms under the harsh environmental conditions, variability of biocontrol from year to year, fermentation, formulation, and storage will be discussed. The partial control of any tactics to protect against FHB up to this moment indicates that the integration of protection measures would provide the best disease management.

(continued on next page)

Table 1. Chronology of works done on biocontrol of Fusarium Head Blight of wheat

Literature	Bioprotectants
Luz, 1988	Bacteria, Yeast
Perondi;N.L., Luz,W.C.da & Thomas,R, 1990 a,1990 b, 1996	<i>Bacillus subtilis</i> <i>Bacillus</i> spp. <i>Pseudomonas fluorescens</i> <i>Sporobolomyces roseus</i>
Stockwell, C.A; Luz,W.C. da, and Bergstrom, G.C., 1997	<i>Paenibacillus macerans</i> <i>Pseudomonas putida</i> <i>Sporobolomyces roseus</i>
Khan, N.I.,Schisler,D.A.,Boehm,M.J, Lipps,P.E., Slininger,P.J. and Bothast, R.J., 1998	<i>Bacillus</i> spp.
Boehm,M.J., Khan, N.J., and Schisler, D.A,1999	Yeast, <i>Bacillus</i> sp.
Khan, N.J., and Schisler, D.A.,and Boehm, M.J.,1999	Yeast, <i>Bacillus</i> sp.
Luo, Y. & Bleakley, B. 1999	<i>Bacillus</i> spp.
Schisler, D.A., Khan, N.J. and Boehm, M.J. 1999	<i>Bacillus</i> spp.
Stockwell, C.A., Bergstrom, G.C. and Luz, W.C. da. 1999	<i>Paenibacillus macerans</i> <i>Pseudomonas putida</i> <i>Sporobolomyces roseus</i>
Luz, 2000	<i>Bacillus megaterium</i> <i>Bacillus subtilis</i> <i>Pantoea agglomerans</i> <i>Kluyvera cryocrescens</i> <i>Paenibacillus macerans</i> <i>Bacillus licheniformans</i> <i>Pseudomonas putida</i> <i>Sporobolomyces roseus</i>
Stockwell, C.A., Bergstrom, G.C. and Luz, W.C. da.,2000	<i>Paenibacillus macerans</i> <i>Bacillus</i> spp.

Table 2. Biodiversity of microorganisms for biocontrol of Fusarium Head Blight of Wheat in Brazil.

Bacillus licheniformans,
Bacillus megaterium,
Bacillus subtilis, *Kluyvera cryocrescens*,
Paenibacillus macerans,
Pantoea agglomerans,
Pseudomonas putida,
Pseudomonas fluorescens,
Sporobolomyces roseus,
Rhodotorula sp.

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INTERACTION OF 28% NITROGEN WITH FOLICUR FUNGICIDE WHEN APPLIED AT HEADING AS A TANK MIX

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INTRODUCTION AND OBJECTIVES

It is a common practice of wheat producers in the Dakotas and Minnesota to treat heading wheat with 28-0-0 nitrogen fertilizer to increase the protein in the grain and attempt to earn a premium at the elevator. This premium may only be offered in years where weather conditions favor exceptionally low protein in hard wheats. 28-0-0 is a solution of about 30# of actual nitrogen/gallon. It is a very viscous substance that has wetting properties. As such it may be used in place of other surfactants when a fungicide is applied. With the interest in applying fungicides at heading to suppress scab, growers have shown asked many questions about the compatibility of 28-0-0 and Folicur (tebuconazole), the most widely used fungicide for suppression of scab. Unfortunately, that information was not available. The Folicur label recommends the product be applied with a non-ionic surfactant (NIS) at a concentration of 0.06-0.125%.

The objective of this study was to determine the compatibility of Folicur with 28-0-0 with or without an added NIS.

MATERIALS AND METHODS

Two spring wheat cultivars, Oxen and Ingot, were planted at three South Dakota locations (Brookings, Groton, and South Shore). At the Brookings and Groton location the previous crop was soybeans, while the South Shore location was planted into corn residue. No inoculum was added to the sites nor was the environment modified with mist irrigation. Four treatments, Folicur (4 fl. oz./A) with NIS (0.125%); Folicur (4 fl. oz./A) tank mixed with 28-0-0 (28.5# actual nitrogen/A); Folicur (4 fl. oz./A) plus NIS (0.125%), tank mixed with 28-0-0 (28.5# actual nitrogen/A); compared to an untreated check. The treatments were replicated six times at all locations. Treatments were applied at Feekes 10.3-10.54. Ingot is about three days later in reaching anthesis than Oxen. At least some tillers were entering anthesis at the time of treatment. Plots were evaluated for scab and leaf disease 21 (Groton) or 28 days (South Shore and Brookings) after treatment, about soft dough. Following harvest, yield, test weight, DON, protein, and FDK measurements were taken.

RESULTS AND DISCUSSION

Substantial damage was observed on the flag leaves and awns of both cultivars after treatment. By the rating date, these differences were not as apparent. Greater differences were observed between the Oxen and Ingot cultivars than between 28-0-0 treatments when compared to the untreated of the same cultivar. Mixed results were observed, with no definitive

advantage to using 28-0-0 on the crop in 2000. Nitrogen fertilizer as the carrier did not significantly alter the performance of the Folicur for scab suppression at any location. Differences were greater between the two varieties tested than among treatments.

Drought conditions caused the crop to mature very rapidly, making leaf disease evaluations difficult. However, by the end of the season the damage sustained in association with 28-0-0 was not as obvious and did not appear to result in serious losses of yield. When Folicur was applied, leaf disease was generally reduced, when only 28-0-0 was applied leaf disease ratings were similar to the untreated. Similar results were observed at Groton with leaf rust suppression across the treatments.

Yields were generally unchanged from the untreated of each cultivar, as a result of including 28-0-0 as a component of the treatment (Table 1 & 3); however, at one location yields were significantly higher when Folicur was included (Table 2). The addition of 28-0-0 did not significantly decrease the yield response realized from the typical Folicur treatment. A Folicur component always reduced leaf disease, usually significantly. Only at the South Shore location were the reductions in leaf disease insignificant. The addition of 28-0-0 did appear to increase protein content in the harvested grain, but not reliably across locations or cultivars.

At the time of this writing, not all data were available for inclusion.

Table 1. Measurements of disease and yield in HRSW Scab 28-0-0/Folicur Trial – SDSU Agronomy Farm, Brookings, SD, 2000.

Treatment Name (Cultivar – Application)	FHB Inc. ^a %	FHB Sev. ^b %	FHB Ind. ^c %	Whole Plot Disease Rating ^e (0-5)	Yield bu/A	Test Weight #/bu	Protein %
Oxen - Untreated	4	35.37	1.4	4.5	52.21	56.94	14.27
Oxen - Folicur + NIS	2.67	24	0.83	3.83	52.62	57.63	14.6
Oxen - Folicur + NIS + 28-0-0	1.33	27.75	0.72	3.92	50.6	57.13	14.88
Oxen - Folicur + 28-0-0	3.33	28.04	1.2	3.83	55.16	57.7	14.87
Oxen – 28-0-0	1.67	12.22	0.35	4.5	53.46	57.98	14.38
Ingot - Untreated	1	7.61	0.46	5	50.33	60.2	13.97
Ingot - Folicur + NIS	1.33	17.92	0.54	4.17	50.63	60.49	14.88
Ingot - Folicur + NIS + 28-0-0	0.67	20.17	0.4	4.67	49.54	60.29	15.18
Ingot - Folicur + 28-0-0	1.67	22	0.61	4.25	53.48	60.32	15.33
Ingot - 28-0-0	1	21.33	0.43	4.92	48.97	60.51	14.52
LSD (0.05)	NS	NS	NS	0.3	3.47	0.83	0.43

^a % of infected heads, based on a 50 head sample
^b % infection of blighted heads (head severity)
^c % blighted heads x % infection on blighted heads
^d Subjective rating (0=green – 5=necrotic) of tissue necrosis in the plot at evaluation.

Table 2. Measurements of disease and yield in HRSW Scab 28-0-0/Folicur Trial – Abelin Farm, Groton, SD, 2000.

Treatment Name (Cultivar – Application)	FHB Inc. ^a %	FHB Sev. ^b %	FHB Ind. ^c %	Leaf Disease ^d % LAR	Leaf Rust ^d % LAR	Yield bu/A	Test Weight lbs/bu	Protein %
Oxen – Untreated	7	8.4	0.88	23.88	4.77	48.33	56.41	15.92
Oxen - Folicur + NIS	8.67	18.67	1.57	5.7	0.28	56.46	57.32	15.78
Oxen - Folicur + NIS + 28-0-0	6.33	12.44	0.74	9.03	0.03	49.53	55.01	15.75
Oxen - Folicur + 28-0-0	6	14.88	0.91	7.3	0.9	52.33	56.51	15.78
Oxen - 28-0-0	5	21.53	1.2	13.3	4.52	51.13	56.46	15.65
Ingot - Untreated	3	6.13	0.28	41.98	18.47	44.36	60.21	15.8
Ingot - Folicur + NIS	2.67	8.94	0.57	3.08	0.3	53.01	60.42	16.07
Ingot - Folicur + NIS + 28-0-0	2.33	7.42	0.27	21.38	0.62	47.71	59.66	16.07
Ingot - Folicur + 28-0-0	2.33	5.44	0.21	11.47	0.38	51.96	60.43	15.92
Ingot - 28-0-0	2.33	6.81	0.26	37.25	16.78	46.93	59.57	15.65
LSD (0.05)	NS	NS	NS	11.23	4.67	4.25	1.19	NS

^a % of infected heads, based on a 50 head sample
^b % infection of blighted heads (head severity)
^c % blighted heads x % infection on blighted heads (field severity)
^d % leaf area damaged by general leaf diseases (largely tan spot) or leaf rust.

Table 3. Measurements of disease and yield in HRSW Scab 28-0-0/Folicur Trial - NE Farm, South Shore, SD, 2000.

Treatment Name (Cultivar - Application)	FHB Inc. ^a %	FHB Sev. ^b %	FHB Ind. ^c %	Whole Plot Disease Rating ^d (0-5)	Leaf Rust % LAR ^e	Yield bu/A	Test Weight lbs/bu	Protein %
Oxen - Untreated	12	56.76	6.3	3.88	0.4	45.22	58.38	15.73
Oxen - Folicur + NIS	6.67	50.82	3.37	3.83	0.2	44.51	59.98	15.9
Oxen - Folicur + NIS + 28-0-0	9.33	57.39	5.12	3.67	0	43.51	59.3	16.42
Oxen - Folicur + 28-0-0	8	52.86	4.51	3.67	2.4	44.98	59.12	15.5
Oxen - 28-0-0	7.33	40.83	3.39	3.83	1.8	43.76	58.95	14.68
Ingot - Untreated	3	32.88	1.41	4.38	3.2	45.53	58.32	15.68
Ingot - Folicur + NIS	5.33	49.04	2.61	4.17	0.6	44.58	59.07	16.35
Ingot - Folicur + NIS + 28-0-0	4.33	33.33	1.8	3.4	0	42.89	58.19	16.42
Ingot - Folicur + 28-0-0	6.67	44.3	3.4	4.08	1.6	45.13	58.77	16.12
Ingot - 28-0-0	4	67.42	2.63	4.08	5.6	42.43	58.27	15.37
LSD (0.05)	5.24	NS	3.12	NS	N/A	NS	NS	1.1

^a % of infected heads, based on a 50 head sample
^b % infection of blighted heads (head severity)
^c % blighted heads x % infection on blighted heads (field severity)
^d Subjective rating (0=green – 5=necrotic) of tissue necrosis in the plot at
^e % leaf area covered by leaf rust pustules

PERFORMANCE OF VARIOUS FUNGICIDES FOR SUPPRESSION OF FUSARIUM HEAD BLIGHT (SCAB) IN SOUTH DAKOTA – 2000

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INTRODUCTION AND OBJECTIVES

Along with 14 other states, South Dakota entered three locations in the Uniform Fungicide Trial for the suppression of Fusarium head blight or scab on hard red spring wheat and one location for scab suppression studies on hard red winter wheat.

The objectives of this study were to evaluate the efficacy of various fungicides or fungicide combinations for the suppression of Fusarium head blight (scab).

MATERIALS AND METHODS

Two spring wheat cultivars, Oxen and Ingot, were planted at three South Dakota locations (Brookings, Groton, and South Shore). At the Brookings location the previous crop was soybeans, while the Groton and South Shore locations were planted into corn residue. No inoculum was added to the sites nor was the environment modified with mist irrigation. Eleven treatments were applied and compared to an untreated control. The treatments included Folicur (4 fl. oz./A), plus Induce non-ionic surfactant (NIS) (0.125%); Tilt (4 fl. oz./A) plus Induce NIS (0.125%); Stratego (14 fl. oz./A) plus Induce NIS (0.125%); BAS 500 (12.3 fl. oz./A) plus Agridex crop oil concentrate (COC) (1%); BAS 500 (6.2 fl. oz./A), plus Folicur (2 fl. oz./A) and Induce NIS (0.125%); Quadris (0.15# a.i./A) plus Benlate (0.25# a.i./A); BAS 500 (6.2 fl. oz./A) plus Agridex COC (1%); Quadris (0.125# a.i./A); Caramba (13.5 fl. oz./A); Folicur (6 fl. oz./A) plus Induce NIS (0.125%); and, Tilt (6 fl. oz./A) plus Induce NIS (0.125%). Treatments were replicated six times at all locations. Treatments were applied at Feekes 10.3-10.54. Ingot is about two days later in reaching anthesis than Oxen, but all plots were flowering on early tillers when treatments were applied. Plots were evaluated for scab and leaf disease 21-28 days after treatment, about soft dough. Following harvest, yield, test weight, DON, protein, and FDK measurements were taken. The winter wheat trial was lost due to heavy pressure from cheat grass and loss of stand due to root rotting fungi.

RESULTS AND DISCUSSION

At the Brookings location (Table 1), little scab developed and, as such, significant differences were not observed among most of the variable measured. Only Quadris, Caramba, and BAS 500 at the lower rate resulted in significantly higher yield, while most products reduced leaf disease as reported as a whole plot rating of general necrosis.

Even less scab was observed at Groton, SD (Table 2); however, leaf disease was more easily quantified. All products significantly reduced leaf diseases in general and leaf rust in

particular. These rating were based on a percent leaf are that was necrotic due to disease. No product significantly reduced FDK below the level observed in the control or increased protein significantly higher than the control. Test weight was significantly increased by Quadris and the high rate of Folicur (6 fl oz/A). All products significantly increased yield.

The highest levels of scab were recorded at the NE Research Farm (Table 3). Nonetheless, significant differences were not discernable. At this location, no significant differences from the untreated control were recorded for any of the measured variables.

The levels of scab observed in South Dakota in 2000 were lower than have been observed for several years.

TABLE 1. Measurements of disease and yield in HRSW Scab Fungicide Trial – Brookings, SD, 2000.

Treatment	FHB Inc. ^b	FHB Sev. ^c	FHB Ind. ^d	Whole Plot Disease Rating ^e	FDK Score	DON (ppm)	Yield (bu/A)	Test Weight (#/bu)	Protein (%)
Product (rate)	(%)	(%)	(%)	(0-5)	(%)	(ppm)	(bu/A)	(#/bu)	(%)
Untreated	2.5	21.49	0.93	4.75	0.75	0.09	51.27	58.57	14.12
Folicur (4) + NIS	2	20.96	0.68	4	1	0	51.62	59.06	14.74
Tilt (4) + NIS	1.5	30.92	0.83	4.38	0.67	0	51.16	58.69	14.54
Stratego + NIS	1.67	30.13	0.8	4.13	0.58	0	51.6	58.84	14.24
BAS 500 (12.3) + COC	2.17	23.35	0.9	4.33	0.67	0	51.74	58.96	14.4
BAS 500 + Folicur + COC	1.67	26.97	0.84	4.54	0.83	0	50.54	58.8	14.08
Quadris + Benlate	2.33	14.94	0.67	4.13	0.92	0.04	52.66	58.89	14.57
BAS 500 (6.2) + COC	2.17	30.38	0.89	4.67	0.58	0	49.99	58.66	14.06
Quadris	0.83	20.08	0.4	3.96	0.67	0.07	54.83	58.98	14.72
Caramba	2.17	15.65	0.76	4.08	0.67	0	54	58.77	14.56
Folicur (6) + NIS	1.83	24.53	0.78	4.63	0.75	0	50.04	58.53	14.29
Tilt (6) + NIS	1.33	12.32	0.59	4.42	0.83	0	52.75	58.91	14.22
LSD (0.05)	NS	NS	NS	0.24	NS	NS	2.46	NS	0.3

^a Data represents an average of disease impact on Oxen and Ingot.
^b % of infected heads, based on a 50 head sample
^c % infection of blighted heads
^d % blighted heads x % infection on blighted heads
^e Subjective rating (0=green – 5=necrotic) of tissue necrosis in the plot at evaluation.

Table 2. Measurements of disease and yield in HRSW Scab Fungicide Trial - Groton, SD, 2000.

Treatment	FHB Inc. ^b	FHB Sev. ^c	FHB Ind. ^d	Leaf Disease ^e	Leaf Rust ^e	FDK	Yield	Test Weight	Protein
Product (rate)	(%)	(%)	(%)	(%)	(%)	(%)	(bu/A)	(#/bu)	(%)
Untreated	5	7.26	0.58	32.93	11.62	1	46.35	58.31	15.86
Folicur (4) + NIS	5.67	13.81	1.07	4.39	0.29	1	54.73	58.87	15.93
Tilt (4) + NIS	6	14.46	1.05	5.67	2.5	1	53.23	58.77	15.72
Stratego + NIS	4	10.99	0.61	11.88	4.45	1.08	52.45	58.76	15.72
BAS 500 (12.3) + COC	5.67	14.95	1.38	4.69	0.21	1.08	55.66	58.62	16.02
BAS 500 + Folicur + COC	5.5	10.28	0.76	8.09	1.73	1.25	50.27	58.46	15.78
Quadris + Benlate	6.83	13.48	1.23	5.91	2.03	1.33	55.05	58.52	15.81
BAS 500 (6.2) + COC	6.17	10.9	0.93	5.97	0.75	1.08	56.92	58.72	15.96
Quadris	6.17	11.81	0.96	9.72	2.27	1.33	50.41	59.26	15.8
Caramba	2.67	9.02	0.37	3.69	0.19	0.83	52.94	58.9	15.94
Folicur (6) + NIS	4.5	19.5	1.02	4.43	0.59	0.92	52.92	59.21	15.88
Tilt (6) + NIS	3.67	15.35	0.76	3.57	1.42	1	56.69	58.72	15.54
LSD (0.05)	NS	NS	NS	7.04	2.88	0.27	4.66	0.93	0.27

^a Data represents an average of disease impact on Oxen and Ingot.

^b % of infected heads, based on a 50 head sample

^c % infection of blighted heads

^d % blighted heads x % infection on blighted heads

^e % leaf area damaged by general leaf diseases (largely tan spot) or leaf rust.

Table 3. Measurements of disease and yield in HRSW Scab Fungicide Trial - NE Research Farm, South Shore, SD, 2000.

Treatment	FHB Inc. ^b	FHB Sev. ^c	FHB Ind. ^d	Whole Plot Disease ^e	Leaf Rust ^e	FDK ^f	Yield	Test Weight	Protein
Product (rate)	(%)	(%)	(%)	(0-5)	(%)	(%)	(bu/A)	(#/bu)	(%)
Untreated	7.5	44.82	3.86	4.13	1.8	1.25	45.37	58.35	15.71
Folicur (4) + NIS	6	49.93	2.99	4	0.4	0.92	44.54	59.53	16.13
Tilt (4) + NIS	7.33	56.59	4.07	4.02	5	1.25	43.57	59.08	15.63
Stratego + NIS	9.83	56.39	5.13	4.08	1.6	1.17	44.83	59.29	15.89
BAS 500 (12.3) +COC	6.17	31.55	3.01	4.08	6.1	1.33	45.85	58.72	16.06
BAS 500 + Folicur + COC	6.67	40.24	3.02	4.21	0.6	1.25	44.74	59.48	15.69
Quadris + Benlate	6.5	54.47	3.75	3.9	2.3	1.33	41.79	59.25	15.53
BAS 500 (6.2) + COC	7	52.72	3.76	4.29	0.2	1.42	44.9	59.29	15.74
Quadris	7.5	48.06	4.23	4.15	1.7	1.42	45.7	53.69	14.66
Caramba	4	45.39	2.2	4.08	0.1	1.08	42.51	58.75	15.64
Folicur (6) + NIS									
Tilt (6) + NIS									
LSD (0.05)	NS	NS	NS	0.34	N/A	NS	NS	NS	NS

^a Data represents an average of disease impact on Oxen and Ingot.
^b % of infected heads, based on a 50 head sample
^c % infection of blighted heads
^d % blighted heads x % infection on blighted heads
^e Subjective rating (0=green – 5=necrotic) of tissue necrosis in the plot at evaluation.

EFFICACY OF THE FUNGICIDE FOLICUR IN CONTROLLING BARLEY FUSARIUM HEAD BLIGHT IN GENOTYPES WITH PARTIAL RESISTANCE

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OBJECTIVES

The objective of this study is to determine if the integrated use of fungicides and resistant or moderately resistant barley genotypes reduces *Fusarium* head blight (FHB) severity and accumulation of deoxynivalenol (DON) to levels acceptable to the malting and brewing industry.

INTRODUCTION

Fusarium head blight (FHB), incited primarily by *Fusarium graminearum*, adversely affected the quality of barley grown in eastern North Dakota and northwestern Minnesota the last eight years. Quality of harvested grain was reduced because of blighted kernels and the presence of DON, a mycotoxin produced by the pathogen. Zero or low levels of DON are needed for malting barley because DON has been found to carry through malting and brewing into finished beer (Schwarz et al., 1995). Anheuser-Busch, Inc., the largest brewer in the U.S., will not purchase malt produced from barley with DON levels greater than 0.5 ppm.

Research conducted to test the efficacy of fungicides in controlling FHB and DON levels in barley has been conducted using cultivars susceptible to FHB. In a study by Pederson and McMullen (1999), they found that the fungicides Folicur, Tilt, Benlate, Mancozeb, and Quadris significantly reduced FHB severity and DON content. However, the fungicides were not successful in reducing the DON content to a level that would be acceptable to maltsters and brewers. The DON content of the barley for the most successful fungicide treatment was 17.2 ppm. Control of FHB in barley probably will require an integrated approach that includes use of cultivars with genetic resistance, proper cultural practices, and fungicides.

Minimal information is available on control of FHB using fungicides on moderately resistant barley genotypes. In a preliminary study conducted by Horsley et al. during the 1999 growing season (unpublished data), the fungicide folicur was sprayed on 13 genotypes of barley at the recommend rate and growth stage. Genotypes resistant to FHB ('Chevron', 'Svanhals', and 'Kaoto Nijo 2'), partially resistant ('MNBrite', F101-78, F103-61, F103-52, and F102-61), and susceptible ('Foster', 'Stander', 'Conlon', 'Logan', and 6B93-2978) were evaluated. The fungicide by genotype interaction was non-significant ($P>0.05$). It was determined that the interaction was not significant because the fungicide reduced the level

of FHB similarly for all genotypes. Averaged across all genotypes, fungicide application reduced FHB severity nearly 40%; yet, the fungicide main effect was not significant ($P>0.05$).

MATERIALS AND METHODS

Fourteen barley genotypes with different levels of FHB resistance were grown at three locations (Osnabrock, Langdon, and Carrington) in North Dakota during the 2000 growing season. Treatments were assigned to experimental units using a randomized complete block design with a split plot arrangement. Each treatment was replicated three times at each location. Whole plots were fungicide levels (0 and 4 oz. acre⁻¹ of folicur) and subplots were genotypes. Evaluated genotypes were either resistant to FHB (Chevron, Svanhals, and Kaoto Nijo 2), moderately resistant to FHB (MNBrite, F101-78, F103-61, F103-52, and F102-61), or susceptible to FHB (Foster, Stander, Conlon, Logan, ND15477, and 6B93-2978). Plots were not inoculated with *F. graminearum*.

The fungicides were applied using a CO₂-pressurized handheld boom sprayer operating at 40 psi, and delivering four ounces of chemical in 18 gallons of water acre⁻¹. Fungicides were applied when 50% of the spikes in a plot were completely emerged from the plants. Fusarium head blight severity was assessed at the soft dough stage by determining the ratio of infected kernels to total kernels on 10 spikes per row. Disease severity was expressed as percent FHB severity. At maturity, grain was harvested from each plot with a plot combine, dried, and cleaned. Grain from each experimental unit were submitted to Dr. Paul Schwarz's laboratory in the Department of Cereal Science, North Dakota State University for DON analysis. To date, DON data are not available.

Data from the experiments were analyzed as an RCBD with a split plot arrangement. Data from individual locations were analyzed separately using analysis of variance (ANOVA) and error mean squares from each location were tested for homogeneity of variance. Combined ANOVA's were done using data from locations in which error mean squares were homogeneous. Mean separation was done using an F-protected LSD ($P=0.05$). In the combined analyses, fungicide and genotypes will be considered fixed effects and environment a random effect.

RESULTS AND DISCUSSION

Environmental conditions at Langdon and Osnabrock, ND were more conducive for development of FHB than conditions at Fargo. Mean FHB severity was 4.8% at Langdon, 4.2% at Osnabrock, and 0.8% at Fargo. The fungicide x genotype interaction was not significant for FHB severity ($P>0.05$). This suggests that Folicur similarly affected FHB severity of all genotypes; however, this was not observed (Table 1). Folicur did not significantly reduce FHB severity in the resistant or moderately resistant genotypes. Thus, it appears that the integrated use of folicur and a resistant or moderately genotype may not be sufficient in reducing FHB severity to levels acceptable to the malting and brewing industry. However, we need to wait until the DON data are available before more definitive conclusions can be made. In the susceptible genotypes Drummond, Foster, Logan, and Stander, folicur gener-

ally reduced FHB severity. Conversely, slight increases in FHB severity were observed in the susceptible genotypes 6B93-2978 and Conlon when sprayed with folicur.

The fungicide x genotype interaction was significant for yield and kernel plumpness ($P < 0.05$). Genotypes sprayed with folicur generally had greater yield, test weight, and kernel plumpness than unsprayed genotypes (Table 2). Much of the improvements in these traits may be due to reductions of foliar disease in genotypes sprayed with folicur (Table 1). Significant yield increases were observed only for the cultivars developed and released from upper Midwest barley breeding programs (i.e. 6B93-2978, Conlon, Drummond, Foster, Logan, MNBrite, and Stander.) This suggests that factors other than foliar diseases were limiting yield in the other genotypes. Foliar disease severity data were collected at Langdon and Osnabrock. The predominant foliar disease at each location was septoria leaf blotch, incited by *Septoria spp.* Foliar diseases were not prevalent at Fargo.

Test weight of genotypes receiving and not receiving folicur was generally acceptable (> 46.0 lb/bu). Kernel plumpness of all adapted genotypes was below the minimum required by the malting and brewing industry, 65%. Thus, folicur application did not result in improvements of kernel plumpness to acceptable levels.

In the barley-growing region in the upper Midwest U.S., it costs growers about \$12/acre for folicur and its application. For this cost to be recovered, a yield increase of at least 9.2 bushels/acre is needed based on a farmgate-selling price of \$1.30/bushel. Based on the yield increases observed in this study, the cost of folicur and its application was recovered only when applied to the adapted cultivars 6B93-2978, Conlon, Foster, Logan, MNBrite, and Stander.

CONCLUSIONS

- Folicur application did not significantly reduce the level of FHB in resistant, moderately resistant, or susceptible genotypes.
- Genotypes sprayed with folicur generally had greater yield, test weight, and kernel plumpness than unsprayed genotypes.
- Folicur application reduced levels of foliar disease.
- Yield gains due to control of foliar diseases tended to be sufficient to cover the cost of folicur and its application in cultivars developed and released by upper Midwest barley breeding programs.

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Table 1. Effect of folicur rate and genotype on percent Fusarium head blight and foliar disease severity of barley.

Genotype	FHB Severity [†]		Foliar disease	
	No folicur	Folicur	No folicur	Folicur
	-----%-----		-----score [‡] -----	
Chevron	1	1	4.2	2.8
Svanhals	1.2	1.3	5.7	3.3
F101-78	3.7	2.5	6.2	4.5
F102-61	1.2	1.7	6.5	3.7
F103-52	1.7	1.6	4.8	4
F103-61	5.1	3.4	5.8	3.7
Kaoto Nijo 2	3.1	3.6	5.7	3.8
MNBrite	3.7	3.5	3.8	2.2
6B93-2978	3.5	4.1	4.2	3
Conlon	3.4	4.9	5	2.7
Drummond	4.4	3.4	5.2	3.5
Foster	5.6	3.5	6.7	4.3
Logan	4.6	3.1	5.8	3
Stander	6.4	4.8	5.7	4.5
LSD (0.05)	-----ns-----		-----ns-----	
[†] Percent of infected kernels per spike. [‡] 1 = no foliar disease, 5 = severe disease.				

Table 2. Effect of folicur and genotype on yield, test weight, and kernel plumpness of barley.

Genotype	Yield		Test weight		Kernel plumpness [†]	
	No folicur	Folicur	No folicur	Folicur	No folicur	Folicur
	bushels/acre		pounds/bushel		%	
Chevron	50.8	54	48.6	49.4	12.5	17.2
Svanhals	41.7	48.6	48.5	49.6	64.1	71.6
F101-78	61.3	62.8	51.5	52	79.9	83.9
F102-61	59.3	61.1	49.2	49.1	70.3	75.1
F103-52	46.3	51.1	49.9	50.5	74.6	72.9
F103-61	63.8	68.9	50.9	51.3	71.3	73.1
Kaoto Nijo 2	61.5	69.1	49.3	50.6	53.5	66.5
MNBrite	72.9	83.6	48.6	49.5	45.5	54.7
6B93-2978	80.8	91.5	48.4	49	43.8	42.7
Conlon	62.8	75.1	47.9	50	51.1	60.6
Drummond	75.1	84.2	48.7	50.2	49.1	58.9
Foster	75	95.7	46.6	48.2	43.6	58.5
Logan	71.1	89.7	49.5	51.1	44.3	60.3
Stander	79.8	90.8	48.7	49.9	52.5	59.1
‡LSD (0.05)	-----9.0-----		-----ns-----		-----7.6-----	
†Kernels remaining on top of a 6/64 inch slotted sieve were considered plump.						
‡Calculated LSD is for comparisons of same subplot treatments in different whole plots						

EFFECTS OF APPLICATION PARAMETERS ON CONTROL OF FUSARIUM HEAD BLIGHT WITH FUNGICIDES

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OBJECTIVE

To find fungicide application techniques that will improve control of Fusarium head blight in spring wheat, durum wheat, and spring barley.

INTRODUCTION

Success with fungicides in reducing Fusarium head blight (FHB) is dependent on a number of factors, including efficacy of the fungicide, appropriate timing of application, and deposition and retention of the fungicide on the grain head (Halley et al., 1999). A number of fungicides have been evaluated since the late 1970s (Wilcoxson, 1996). Folicur (tebuconazole) has shown consistency in reducing levels of FHB over a number of locations in the US in recent years (McMullen et al., 1999).

In addition to fungicide efficacy, timing of application needs to be optimal. In early studies, fungicides were applied at early or partial head emergence or at early grain fill (Milus and Parsons, 1994; Wilcoxson, 1996). Studies in Europe indicate that the flowering period is the optimum growth stage for fungicide application on wheat (Mauler-Machnik and Zahn, 1994). For barley, which flowers when the heads are still in the boot, early results in North Dakota indicate that optimum fungicide application on barley is shortly after head emergence.

Adequate deposition and retention of fungicide on grain heads also is needed for optimum FHB control. Adjuvants have been shown to have positive effects in improving fungicide deposition and retention on grain heads, resulting in enhanced FHB control (Lukach et al., 1999). Recent studies in greenhouse and field environments also have indicated that spray systems that direct the fungicide spray toward the grain head at an angle rather than vertical to the grain head provide better fungicide deposition and FHB control (Lukach et al., 1999). Our objective was to further evaluate the impact of application timing and the impact of adjuvant types and rates on fungicide effectiveness in controlling FHB.

MATERIALS AND METHODS

Application timing - Folicur fungicide was applied at various growth stages of spring wheat and barley in the greenhouse and field in 2000. 'Grandin' spring wheat, 'Munich' durum, and 'Robust' barley were sown separately into artificial potting mix in 6" by 12" plastic trays at a seeding rate equal to one million seeds/acre. Five trays of each crop were treated with Folicur (4 fl oz + 0.06% v/v Induce in 9 gpa) at three separate growth stages (Feekes 10.3 = one-half head emerged; Feekes 10.5 = full head emergence in barley; Feekes 10.51 =

flowering in wheat; Feekes 10.54 = kernel developing and watery ripe). Fungicides were applied with a track sprayer using two XR8001 flat fan nozzles oriented forward and backwards towards the grain heads at 30° from horizontal. Grain heads were inoculated at Feekes 10.51 using an atomizer with 5000 spores/ml of *Fusarium graminearum*. Plants were placed under intermittent mist for two days following inoculation.

'Russ' hard red spring wheat was planted into barley stubble that had been chiseled the prior fall, and 'Stander' barley was planted into corn stubble that had been chiseled the prior fall. Fungal inoculum came from natural sources in the barley field, while natural sources were augmented with inoculated grain spawn spread among rows in the wheat field. Four replicated plots (9' x 20') per treatment were sprayed with Folicur fungicide (4 fl oz/acre + 0.06% Induce in 18 gpa) using the same nozzle type and orientation as in the greenhouse. FHB was evaluated at soft dough stage of kernel development. Data was statistically analyzed using ANOVA.

Adjuvants - Various adjuvants were tested in conjunction with Folicur or Tilt fungicide to determine their effects on efficacy of these products. Experiments were conducted in field trials at Fargo with Stander barley and Grandin hard red spring wheat, both planted into corn stubble. Adjuvants tested included two rates of a non-ionic surfactant (Induce), two rates of an organosilicone adjuvant (Silwet), and two humectants, L64 and L88. Humectants, such as glycerol and sorbitol, are substances that enhance retention of moisture. Fungicides + adjuvants were applied with a hand held boom equipped with forward/backward flat fan nozzles, as described above. FHB was evaluated at soft dough stage of kernel development. Data was statistically analyzed using ANOVA.

RESULTS AND DISCUSSION

Application timing - Optimum growth stage for fungicide application for FHB control was at Feekes growth stage 10.51 for spring wheat and durum (Table 1). In barley, FHB field severity did not differ significantly among application timings, but was numerically lower when fungicide was applied either at Feekes 10.3 or Feekes 10.5 than at Feekes 10.54 (Table 1). Optimum timing of application resulted in 73-92% reduction in FHB field severity in spring wheat, 89% in durum wheat, and 59-63% in barley.

Adjuvants - With Folicur fungicide on barley and wheat, Induce adjuvant at 0.06% v/v consistently resulted in lower FHB field severity ratings than at 0.03% v/v, although differences were not statistically significant (Table 2). Over all of the treatments with Folicur or Tilt, the lowest FHB field severity ratings were with Folicur + 0.06% v/v Induce. A comparison of Silwet rates with Tilt indicated that the 0.03% v/v rate of Silwet gave a significantly lower FHB field severity rating than use of the 0.06% v/v rate. Results indicate that benefits with adjuvants may be rate dependent. Addition of humectants resulted in similar FHB field severity ratings as with the Induce or Silwet, but slightly lower wheat leaf disease levels than with other adjuvants.

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Table 1. Field severity of Fusarium head blight (FHB) following application of Folicur fungicide (4 fl oz/acre + 0.06% v/v Induce) at various growth stages, Fargo, 2000.

Feekes growth stage* at fungicide application	% FHB Field Severity (Field Severity = Incidence x Head Severity)				
	'Grandin'	'Russ'	'Munich'	'Robust'	'Stander'
	HRSW <i>greenhouse</i>	HRSW <i>field</i>	Durum <i>greenhouse</i>	Barley <i>greenhouse</i>	Barley <i>field</i>
Untreated	7	23.4	32.1	3.2	8.7
10.3	1.6	11.2	16.4	1.5	3.2
10.5-10.51	0.5	6.3	3.5	1.3	4.1
10.54	7	8.8	22.5	1.9	4.5
LSD P= 0.05	3.9	4.6	17.2	2.8	2.8

* Feekes 10.3 = head half emerged; Feekes 10.5 = full head emergence; Feekes 10.51 = early flowering in wheat; Feekes 10.54 = kernel watery ripe

Table 2. Effects of adjuvants on efficacy of Folicur and Tilt fungicides in reducing FHB in two field experiments with barley, one field experiment with spring wheat, Fargo, ND, 2000.

Cultivar	Fungicide	Adjuvant and rate	FHB Field Severity (%)	Barley Yield (bu/ac)
Stander barley	Untreated		6.1	83.6
	Folicur 4 fl oz/A	Induce 0.03% v/v	4.4	87.9
	Folicur 4 fl oz/A	Induce 0.06% v/v	3.7	90.1
LSD P = 0.05			1.5	ns
Stander barley	Untreated		9.4	84
	Folicur 4 fl oz/A	L64 1% v/v	5	87.1
	Folicur 4 fl oz/A	L88 1% v/v	4.5	86.5
	Folicur 4 fl oz/A	Induce 0.03% v/v	4.7	83.9
	Folicur 4 fl oz/A	Induce 0.06% v/v	3.8	89.5
	Folicur 4 fl oz/A	Silwet 0.03% v/v	4	86.5
	Tilt 4 fl oz/A	Silwet 0.03% v/v	4.1	82.4
	Tilt 4 fl oz/A	Silwet 0.06% v/v	6.2	86.9
	Tilt 4 fl oz/A	Induce 0.03% v/v	4.8	86.3
	Tilt 4 fl oz/A	Induce 0.06% v/v	5.7	84.1
	LSD P = 0.05			2.1
				% Leaf necrosis
Grandin HRS	Untreated		39.5	60.3
	Folicur 4 fl oz/A	L64 1% v/v	15.1	5.2
	Folicur 4 fl oz/A	Induce 0.03% v/v	17	8.5
	Folicur 4 fl oz/A	Induce 0.06% v/v	11.8	8.7
	Folicur 4 fl oz/A	Silwet 0.03% v/v	11.9	7.5
	Tilt 4 fl oz/A	Induce 0.06% v/v	13.1	11.8
LSD P = 0.05			7.1	6.1

UNIFORM FUNGICIDE TRIAL FOR CONTROLLING FHB IN SPRING WHEAT, ND, 2000

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ABSTRACT

A core set of seven fungicide treatments was evaluated on hard red spring wheat for efficacy against Fusarium head blight (FHB = scab) and leaf diseases at three locations (Fargo, Carrington, and Langdon) in North Dakota in 2000. A single study on durum wheat also was done with the fungicide treatments at the Carrington Research and Extension Center. The evaluation of the fungicide treatments was part of a national effort to evaluate a uniform set of treatments across multiple states and environments. The core set of treatments included Folicur (4 fl oz/acre), Tilt (4 fl oz/acre), Stratego (14 fl oz/acre), BAS 500 (12.3 fl oz/acre), BAS 500 + Folicur (6.2 fl oz + 2 fl oz/acre), and Quadris + Benlate (9.2 fl oz + 0.25 lb/acre). Folicur (tebuconazole) had a Section 18 for use in ND in 2000. Tilt (propiconazole) has a state label for heading application on wheat in ND. Quadris (azoxystrobin) is registered for wheat. Stratego (combination product of Tilt + trifloxystrobin [Flint]) was recently sold by Novartis to Bayer, and its availability status for 2001 is uncertain. BAS 500 is a strobilurin fungicide and is not yet labeled for wheat in the US. An additional fungicide, Caramba 90SL (metconazole at 13.5 fl oz/acre), was included in the evaluations in ND. Caramba is not labeled in the US and this was the first year of testing of this product. The fungicide treatments were applied at early flowering to 'Russ' hard red spring wheat at Fargo and Carrington, to 'Grandin' hard red spring wheat at Fargo and Langdon, and to 'Munich' durum at Carrington. Fungicide treatments were applied at 35-40 psi in 17-18 gpa, using flat fan nozzles oriented forward and backward at an angle 30° from the horizontal. Fusarium inoculum was added in the form of inoculated grain kernels to the plots in Fargo and Langdon. Wheat straw was distributed among plots at Carrington. Plots were mist irrigated at Fargo and Langdon, while sprinkler irrigation was used at Carrington. Fusarium head blight and leaf disease ratings were recorded at soft dough stage of kernel development. Disease levels were high at all three locations. Mean data across sites was analyzed using ANOVA. Results on spring wheat and durum showed that all of the fungicide treatments significantly reduced leaf disease and FHB field severity when compared to the untreated check. In the hard red spring wheat trials, all fungicide treatments resulted in significantly lower DON levels, as well. All fungicide treatments significantly increased yield (7 to 15 bu in hard red spring and 12 to 18.4 bu in durum) and test weight (up to 2.3 lb in hard red spring and up to 3.4 lb in durum). The Caramba fungicide resulted in significantly lower DON (vomitoxin) levels across all sites than other fungicide treatments and significantly higher yields than several of the core treatments. The BAS 500 treatment resulted in the lowest leaf disease rating and the highest test weight.

UNIFORM FUNGICIDE TRIAL FOR CONTROLLING FHB IN BARLEY, ND, 2000

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ABSTRACT

A core set of seven fungicide treatments was evaluated on 'Stander' six-row barley for efficacy against Fusarium head blight (FHB = scab) and leaf diseases at two locations in North Dakota in 2000. The evaluation of the fungicide treatments was part of a national effort to compare a uniform set of treatments across multiple states and environments. The core set of treatments included Folicur (4 fl oz/acre), Tilt (4 fl oz/acre), Stratego (14 fl oz/acre), BAS 500 (12.3 fl oz/acre), BAS 500 + Folicur (6.2 fl oz + 2 fl oz/acre), and Quadris + Benlate (9.2 fl oz + 0.25 lb/acre). Folicur (tebuconazole) had a Section 18 for use on wheat and barley in ND in 2000. Tilt (propiconazole) has a federal label for application to wheat and barley through flag leaf emergence stage, and a state label for heading application on wheat in ND. Quadris (azoxystrobin) is registered for wheat. Stratego (combination product of Tilt + trifloxystrobin [Flint]) was recently sold by Novartis to Bayer, and its availability status for 2001 is uncertain. BAS 500 is a strobilurin fungicide and is not yet labeled for wheat or barley in the US. An additional fungicide, Caramba 90SL (metconazole at 13.5 fl oz/acre), was included in the evaluations. Metconazole is not labeled in the US and this was the first year of testing in ND. The fungicide treatments were applied to Stander barley at early full head emergence at both locations. Fungicide treatments were applied at 35-40 psi in 17-18 gpa, using flat fan nozzles oriented forward/backward at a 30° angle from the horizontal. Fusarium inoculum was added in the form of inoculated grain kernels to the plots in Fargo and Langdon, and mist irrigation was applied to the plots at both locations as needed. Fusarium head blight and leaf disease ratings were recorded at soft dough stage of kernel development. FHB, DON, and leaf disease levels were higher at Langdon than Fargo. Data across sites was analyzed using ANOVA. Compared to the untreated check, all fungicide treatments significantly reduced leaf disease, which was primarily Septoria. Among fungicide treatments, the only significant difference observed was the BAS 500 + Folicur treatment having significantly lower leaf disease than the Tilt treatment. Significant differences were not observed among treatments for FHB field severity, but fungicide treatments reduced severity levels by 45 to 66.7%. DON levels were very high in these studies and were not significantly affected by fungicide treatments, although fungicides reduced DON levels up to 45.8%. Yield differences also were not statistically significantly different because of wide yield variations between the two sites, but yield improvement averaged from 10 to 20.4 bushels, with Caramba and Stratego treatments resulting in the highest yields. At Langdon, test weights were significantly improved by all but the Tilt fungicide treatment, with Folicur giving the highest test weight.

ANALYSIS OF THE 2000 UNIFORM WHEAT FUNGICIDE TRIALS ACROSS LOCATIONS

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INTRODUCTION

Identifying fungicides that reduce incidence and severity of FHB and levels of damage and mycotoxins in the grain could have immediate and wide spread benefits to growers and users of all market classes of wheat in the event of a FHB epidemic. The overall objective of the uniform wheat fungicide trials is to hasten the integration of fungicides that are effective against FHB into cost-effective and environmentally-safe wheat disease management strategies. Because most fungicides do not have a high level of activity against FHB, current emphasis has been on identifying the most efficacious fungicides. Consequently, this analysis will include only variables that are directly related to FHB.

METHODS

Plant pathologists in 15 states (Table 1) participated in the 2000 wheat uniform fungicide trials. These states represented hard red spring wheat, hard red winter wheat, soft red winter wheat, soft white winter wheat, and durum wheat production areas. The seven uniform treatments for 2000 (Table 2) included well-known fungicides that either are registered or are in the process of being registered for use on wheat in the United States. In addition to the seven uniform treatments, some cooperators included Caramba (Metconazole) which is being used for control of FHB in Europe.

All treatments were applied at flowering stage using a CO₂-powered sprayer. Details such as plot size, number of replications, spray volume, and nozzle configuration varied among the locations but were not considered to significantly affect the results. Inoculation and/or some form of overhead misting were used at some locations to promote head blight development, and these practices likely increased the incidence and severity of head blight. Disease variables included incidence (% of heads showing symptoms) and head severity (% of head area affected) measured at soft dough stage, field severity (= FHB index = incidence x head severity), and deoxynivalenol (DON) content in the grain and percentage of Fusarium-damaged kernels (FDK) measured after harvest. Cooperators analyzed results of their individual locations and provided treatment means to the authors for analysis across locations.

RESULTS

Sixteen locations across seven of the participating states reported some level of FHB (Table 3). The trial from Maryland appeared to be a good test, but a nontreated check was not included because of unusual circumstances. In order to utilize data from this location, data

for two extra, apparently ineffective treatments were averaged, and these averages were substituted for the nontreated check. Field severity data from ten locations, FDK data from four locations, and DON data from five locations were used to analyze treatment effects across locations. All fungicides significantly reduced field severity compared to the nontreated check, but none of the treatments significantly reduced the percentage of Fusarium-damaged kernels or the level of DON (Table 4).

Seven locations included the Caramba treatment and reported some level of FHB (Table 5). Data for the nontreated check at the Maryland location were calculated as described above. Field severity data from five locations, FDK data from three locations, and DON data from four locations were used to analyze treatment effects across locations. All fungicides significantly reduced field severity compared to the nontreated check, and Caramba was significantly better than Tilt or Stratego (Table 6). Stratego, BAS 500F + Folicur, Quadris + Benlate, and Caramba significantly reduced the percentage of Fusarium-damaged kernels compared to the nontreated check, and Caramba was significantly better than BAS 500F + Folicur and Quadris + Benlate. All fungicides significantly reduced the level of DON compared to the nontreated check, and Caramba was significantly better than all of the fungicides except Quadris + Benlate. Also, Caramba was the only fungicide to reduce DON levels below the threshold level of 2 ppm.

CONCLUSIONS

Caramba was the most effective fungicide against FHB among all of the fungicides ever tested in the uniform wheat fungicide trials. Caramba also was effective against foliar fungal diseases (data not shown) and therefore appears to have potential for cost-effective commercial use. Although Caramba is currently being used in Europe, the prospect for EPA registration on wheat in the United States and cost are not known at this time.

For the analyses without Caramba (Table 4), results were very similar to those reported from the 1999 analysis across locations (McMullen, et al. 1999). In 1999 and 2000, all fungicides tested significantly reduced field severity compared to the nontreated check, but none of the fungicides significantly reduced the percentage of Fusarium-damaged kernels or the level of DON.

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#	Fungicide	Rate/A	Adjuvant
1	Nontreated		
2	Folicur 3.6 F	4 fl oz	0.12% Induce
3	Tilt 3.6 EC	4 fl oz	0.06% Induce
4	Stratego 2.08 EC	14 fl oz	0.06% Induce
5	BAS 500F 2.09 EC	12.3 fl oz	1% Agri-Dex
6	BAS 500F + Folicur	6.2 + 2 fl oz	1% Agri-Dex
7	Quadris 2.08 F + Benlate 50 WP	9.2 fl oz + 0.25 lb	
7.5	Caramba 90 SL	13.5 fl oz	0.06% Induce

Table 3. Locations in the 2000 uniform trials that reported some level of FHB and provided data on field severity, Fusarium-damaged kernels, or DON level and the means across all seven treatments for those variables.

Location	Field severity ^a (%)	Fusarium-damaged kernels ^a (%)	DON ^a (ppm)
Fargo, ND #1	13.2	5.4	7.3
Fargo, ND #2	5.8	2.6	3.6
Carrington, ND #1	12.8	.	2.4
Carrington, ND #2	12.4	.	3.2
Langdon, ND	13.7	19.1	.
Crookston, MN	2.4	3.6	1.2
Beltsville, MD	14	24.6	.
Aurora, NY	.	7.6	11.8
East Lansing, MI #1	7.1	.	.
East Lansing, MI #2	12.6	.	.
East Lansing, MI #3	16.3	.	.
East Lansing, MI #4	11.9	.	.
Groton, SD	1	1.1	.
Brookings, SD #1	0.8	0.8	.
Brookings, SD #2	3.7	1.2	.
Fayetteville, AR	27.1	84.3	44

^aValues in bold font were used for analysis across locations. Other values were from locations where FHB levels were either too low or too high to be meaningful.

Table 4. Treatment means for field severity, Fusarium-damaged kernels, and DON level averaged across the locations indicated in Table 3.

Treatment	Field severity (%)	Fusarium-damaged kernels (%)	DON (ppm)
Nontreated	20.5	16.9	7.3
Folicur	9	14.5	4.6
Tilt	11.5	13.6	4.8
Stratego	12.7	12.7	6
BAS 500	8.6	14.4	5.7
BAS 500 + Folicur	10.5	14.1	6.3
Quadris + Benlate	11.2	12.9	5
LSD (P=0.05)	4.8	NS	NS

Table 5. Locations in the 2000 uniform trials that included the Caramba treatment and provided data on field severity, Fusarium-damaged kernels, or DON level and the means across all eight treatments for those variables.

Location	Field severity ^a	Fusarium-damaged kernels ^a	DON ^a
	(%)	(%)	(ppm)
Fargo, ND #1	12.1	5.1	6.8
Fargo, ND #2	5.3	2.6	3.4
Carrington, ND #1	11.9	.	2.2
Carrington, ND #2	11.7	.	3
Langdon, ND	12.6	18.4	.
Crookston, MN	2.4	3.3	1.1
Beltsville, MD	13.5	23.5	.

^aValues in bold font were used for analysis across locations. Other values were from locations where the level of FHB was too low to be meaningful.

Table 6. Treatment means for field severity, Fusarium-damaged kernels, and DON level averaged across the locations indicated in Table 5.

Treatment	Field severity	Fusarium-damaged kernels	DON
	(%)	(%)	(ppm)
Nontreated	24.9	19.8	6.4
Folicur	9.3	17.1	3.5
Tilt	12.2	16	3.6
Stratego	11.3	14	4.3
BAS 500F	8.6	16.9	3.5
BAS 500F + Folicur	8.5	15.5	4.1
Quadris + Benlate	9.3	15	3.4
Caramba	5.4	10.8	1.8
LSD (P=0.05)	4.3	3.9	1.6

USDA-ARS, OHIO STATE UNIVERSITY COOPERATIVE RESEARCH ON BIOLOGICALLY CONTROLLING FUSARIUM HEAD BLIGHT: FIELD TESTS OF ANTAGONISTS IN 2000

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OBJECTIVES

Determine the effect of culture media type on the efficacy of biocontrol agents against FHB in field tests located in Illinois and Ohio and to produce antagonists in a superior cultivation medium and field test antagonists in North Dakota. Results of field studies on reducing FHB on durum wheat using microbial antagonists is presented in our associated poster.

INTRODUCTION

Research on optimizing methods for selectively isolating, mass producing and utilizing microbial antagonists effective against FHB was initiated in 1997 at the NCAUR in Peoria, IL, in conjunction with The Ohio State University. Several biological control agents remain under consideration for commercial development (Table 1; Boehm et al., 1999; Khan et al., 1999). This update provides a summary of the results obtained from a portion of our 2000 field evaluation trials conducted on the soft red winter wheat cultivars Pioneer 2545 and Freedom in OH and IL and the hard red spring wheat cultivar Grandin in Langdon, ND.

MATERIALS AND METHODS

2000 field trials of FHB antagonists on soft red winter wheat in Peoria, IL and Wooster, OH

Inoculum of 6 microbial antagonists effective against FHB was produced using two semidefined liquid culture media that differed only in respect to their carbon to nitrogen ratios (C:N) which were set at C:N 6.5 and 11. These media were selected in part because of data obtained from a 2000 field trial on durum wheat in which antagonists produced in the C:N 6.5 medium were 18% more effective in reducing FHB than the same antagonists produced in the C:N 11 medium (Schisler et al. poster, this forum). The soft red winter wheat cultivars Pioneer 2545 (susceptible) and Freedom (moderately resistant) were used in both locations because of their widespread use throughout the Midwest and to test for possible integration of biocontrol and host genetic resistance for managing FHB. Biomass was harvested from Fernbach shake flasks as described previously and applied at the beginning of wheat flowering in aqueous suspensions containing a weak buffer and wetting agent (Schisler et al., 1999). Bacterial and yeast suspensions contained 25 % fully colonized broth (~5x10⁸ cfu/ml and ~2.5 x 10⁷ cfu/ml, respectively). Controls were untreated plants and

plants treated with buffer/wetting agent only. Pathogen inoculum was added to plots as colonized corn kernels (*F. graminearum* 3-93 and 6-93) scattered through plots (~25-40 kernels/m²) 2 wk prior to wheat flowering and mist irrigation provided for approximately one week after treatment application to promote FHB development. Plots were scored for disease severity and incidence. Randomized complete block designs were used in both trials ($n=4$ in Peoria; $n=6$ in Wooster).

2000 Langdon, ND, field trial of FHB antagonists on the hard red spring wheat Grandin

Biomass of the 6 FHB biocontrol agents listed in Table 1 was produced in the C:N 6.5 semidefined medium for the Langdon, ND field trial. Methods were as described above except that natural pathogen inoculum was used and the plots did not receive any mist irrigation. Controls were untreated plants and plants treated with buffer/wetting agent only. Plots were scored for disease severity and incidence. A randomized complete block design was used ($n=6$).

RESULTS AND DISCUSSION

All antagonists reduced FHB severity as compared to the buffer control in both Peoria and Wooster on Pioneer 2545 (Table 2). Yeast OH 182.9 reduced disease severity by 58% compared to the buffer control across both production media and test sites. Disease severity was very light on cultivar Freedom with few treatments differing from the controls at either field location. The C:N of the liquid culture media used to produce antagonist biomass did not have a consistent effect on the efficacy of antagonists in the field trials conducted in Peoria and Wooster (Table 2). In the Langdon, ND field trial, five of six antagonists decreased FHB severity compared to the untreated control (Table 3). Inexplicably, the buffer and wetting agent control treatment also had one of the lowest disease severity ratings, a result in contrast to our 1999 and other 2000 field trials where the buffer control had equivalent or higher disease ratings than the untreated control. As observed in previous field seasons, yeasts OH 181.1 and OH 182.9 were among the most effective antagonists.

We remain optimistic regarding the commercial development and FHB control potential of several of these antagonists. Further improvements in the level of control achieved using these antagonists will be sought via further research on optimizing media and formulations. Additional efficacy gains will also be attempted via the selection of fungicide insensitive variants of selected antagonists and their use in combination with fungicides registered for use against FHB. Lastly, improving the consistency of biological control will be sought by initiating studies to discover genes, regulatory mechanisms or other cellular processes responsible for biocontrol agent efficacy.

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Table 1. Antagonist strain designation and identification of bacteria and yeasts that reduce the severity of Fusarium head blight of wheat.

Antagonist	NRRL accession no. ¹	Identification
AS 43.3	B-30210	<i>Bacillus subtilus/amyloliquefaciens</i> ²
AS 43.4	B-30211	<i>Bacillus subtilus/amyloliquefaciens</i> ²
OH 71.4	Y-30213	<i>Cryptococcus</i> sp.(= <i>Torula aurea</i>) ³
OH 131.1	B-30212	<i>Bacillus subtilus</i> ⁵
OH 181.1	Y-30215	<i>Cryptococcus</i> sp. nov. 1 ³
OH 182.9	Y-30216	<i>Cryptococcus nodaensis</i> sp. nov ³

¹NRRL patent culture collection, National Center for Agricultural Utilization Research, Peoria, IL.

²Identification by DSMZ, Braunschweig, Germany, based on 16S rDNA sequence homologies and biochemical and physiological tests of taxonomic utility.

³Identification based on nucleotide sequence divergence in domain D1/D2 of large subunit 26S rDNA and on divergence in ITS 1/5.8/ITS2 rDNA. C.P. Kurtzman, personal communication.

⁴Yeast, not determined.

⁵Identification by MIDI Labs, Newark, DE, based on 16S rDNA sequence homologies and biochemical and physiological tests of taxonomic utility.

Table 2. Influence of six microbial antagonists produced in C:N 6.5 and 11 liquid media on Fusarium head blight on the soft red winter wheat

Treatment	Cultivar Pioneer 2545				Cultivar Freedom			
	Peoria, IL		Wooster, OH		Peoria, IL		Wooster, OH	
	Disease Severity, 1 %	Disease Incidence, %	Disease Severity, %	Disease Incidence, %	Disease Severity, %	Disease Incidence, %	Disease Severity, %	Disease Incidence, %
Untreated control	3.5 cd	23.8 bcd	4.2 c	26.4 cdefg	1.0 ab	10.8 a	2.7 de	25.8 cd
Buffer control	7.6 a	32.1 a	8.4 a	40.0 a	0.6 bc	7.9 ab	3.0 cde	24.7 d
AS 43.3 (6.5) ²	4.7 bcd	27.1 abc	4.0 cd	27.8 cdef	0.6 bc	7.5 ab	2.2 e	22.0 d
AS 43.4 (11)	4.3 bcd	26.7 abc	4.1 cd	30.3 bcd	0.9 ab	10.0 ab	4.0 abc	30.3 abc
AS 43.3 (6.5)	4.2 bcd	24.2 bcd	5.8 b	36.1 ab	0.4 c	5.0 b	3.8 bcd	31.1 abc
AS 43.4 (11)	4.4 bcd	24.6 abcd	4.2 c	28.3 cde	1.1 a	10.8 a	5.0 a	34.2 a
OH 71.4 (6.5)	3.9 bcd	23.8 bcd	2.9 d	22.5 efg	0.7 abc	7.9 ab	3.9 abc	28.1 abcd
OH 71.4 (11)	3.3 cd	21.2 cd	4.1 cd	24.2 defg	0.8 abc	10.0 ab	2.9 cde	25.8 cd
OH 131.1 (6.5)	4.6 bcd	29.6 ab	4.0 cd	25.3 defg	0.7 abc	8.8 ab	3.8 bcd	31.1 abc
OH 131.1 (11)	4.9 bc	26.2 abcd	3.9 cd	24.2 defg	0.8 abc	10.4 a	2.2 e	21.7 d
OH 181.1 (6.5)	5.3 b	26.7 abc	4.1 cd	28.6 cde	0.8 abc	10.8 a	4.4 ab	32.8 ab
OH 181.1 (11)	3.2 cd	18.8 d	5.6 b	32.5 bc	0.9 ab	9.6 ab	3.4 bcd	27.5 bcd
OH 182.9 (6.5)	4.1 bcd	27.1 abc	2.9 cd	21.4 g	0.9 ab	10.4 ab	3.0 cde	27.8 abcd
OH 182.9 (11)	3.0 d	20.8 dc	3.4 cd	22.0 fg	0.9 ab	11.7 a	2.7 de	25.3 cd

¹Within a column, means not followed by the same lower case letter are significantly different ($P \leq 0.05$, FPLSD).

²Value in parentheses indicates the carbon:nitrogen ratio of the medium used to produce biomass of the antagonist.

Table 3. Influence of six microbial antagonists produced in C:N 6.5 medium on Fusarium head blight on the hard red spring wheat cultivar Grandin in Langdon, North Dakota, 2000.

Treatment	Disease Severity, ¹ %	Incidence, %
Untreated control	21.8 a	52.8 a
Buffer control	12.9 de	28.1 e
AS 43.3 (6.5) ²	16.6 bc	39.7 c
AS 43.4 (6.5)	14.3 cd	42.2 bc
OH 71.4 (6.5)	18.9 ab	47.8 ab
OH 131.1 (6.5)	12.7 de	42.8 bc
OH 181.1 (6.5)	10.4 e	32.5 de
OH 182.9 (6.5)	12.3 de	37.2 cd

¹Within a column, means not followed by the same lower case letter are significantly different (P#0.05, FPLSD).

²Value in parentheses indicates the carbon:nitrogen ratio of the medium used to produce biomass of the antagonist.

Control of Fusarium Head Blight of Wheat with Foliar Fungicides

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INTRODUCTION AND OBJECTIVES

Scab, or Fusarium head blight, has been a difficult to control throughout the world. The shift to conservation tillage methods of crop production may be why Fusarium head blight has emerged as a major disease of wheat and barley through much of the US Corn Belt and the upper Great Plains (Bai and Shaner, 1994; Dill-Macky and Jones, 2000). No single traditional disease control option (disease resistant cultivars, crop rotations, tillage to destroy residues, and fungicides) is likely to control head blight because of lack of availability, excessive cost, or negative impacts on soil conservation (McMullen 1997). Bringing Fusarium head blight under control will require multiple disease management strategies (Parry et al. 1995, Bai and Shaner 1994), coupled with greater understanding of the epidemiology of the disease.

Fungicides would provide growers with a management option when susceptible cultivars are grown, and may help protect grain yield and quality of cultivars with partial resistance under conditions particularly favorable for disease. A few fungicides have shown some efficacy against Fusarium head blight, but results are often inconsistent over locations and years (Gilbert and Tekauz, 2000; McMullen et al., 1997; Parry et al., 1995; Shaner and Buechley, 1999). Some fungicides reduce DON contamination of grain, but others may cause an increased amount of DON (Gilbert and Tekauz, 2000; Shaner and Buechley, 1997; Shaner and Buechley, 1999).

We have been participants in a uniform fungicide trial. The purpose of this cooperative study is to compare a core set of fungicide treatments at several locations throughout the Corn Belt and upper Midwest for their efficacy against Fusarium head blight of wheat and barley.

MATERIALS AND METHODS

Wheat cultivar Clark was drilled at 7-in. row spacing into disked corn stalks at two locations in Indiana during 2000. Wheat at the Purdue Agronomy Research Center (ARC) near West Lafayette was planted on 13 Oct 99; wheat at the Southeast Purdue Agricultural Center (SEPAC), near North Vernon, was planted on 6 Oct 99. Before seeding at ARC, 300 lbs./A of 10-25-25 fertilizer was broadcast and incorporated. Plots at ARC were top-dressed in early spring with 315 lbs./A of 34-0-0. Prior to planting at SEPAC, the field was fertilized with 100 lbs. of 34-0-0 and 100 lbs. of 0-0-60. Top dressing was applied in early spring as 200 lbs. of 34-0-0. Mowing alleys both parallel and perpendicular to the direction of seeding created plots 25 ft long and 10 ft (ARC) or 15 ft (SEPAC) wide. Both experiments were set up as a randomized complete block with four replications.

Disease developed from natural inoculum. Fungicides were applied with a CO₂-pressurized sprayer that delivered 40 gpa at 40 psi, with a hand-held boom that had 8 (ARC) or 14 (SEPAC) TJ 60-8002 nozzles spaced at 14 in. Fungicides were applied at flag leaf emergence (GS 37) and beginning of anthesis (GS 61) at ARC and at early boot (GS 43) and full head emergence (GS 59) at SEPAC. Intensity of foliar disease was visually estimated on a whole-plot basis. Incidence of Fusarium head blight was estimated by counting the number of blighted heads in 10 arbitrarily selected 1-ft lengths of row and expressing these counts as a percentage of the average number of heads per foot of row.

Plots were combine-harvested on 23 June at ARC and on 29 June at SEPAC. Grain was dried to 13% moisture after which yield and test weight were measured. Approximately 1500 kernels from each plot from the ARC experiment were inspected for incidence of visible scab. Grain samples from each plot at both locations were sent to Dr. Pat Hart at Michigan State University for determination of DON content.

RESULTS AND DISCUSSION

Powdery mildew, leaf rust, and Septoria and Stagonospora leaf blotches were not severe at either location. Nonetheless, most treatments except the Cornell biological control agents reduced severity compared to the untreated control. Incidence of Fusarium head blight was low at SEPAC and moderate at ARC. No treatment at SEPAC reduced incidence compared to the untreated control. At ARC, Caramba, Stratego, and Folicur + Induce, when applied at GS 61, reduced head blight incidence significantly (Table 1). Four treatments at ARC reduced the percentage of scabby kernels compared to the untreated control. Five treatments also reduced the level of DON in grain, three of which were among those that reduced incidence of scabby kernels. At SEPAC, DON levels were generally lower than those at ARC, and no treatment had less DON than the untreated control. Several treatments, however, had significantly greater levels of DON than the untreated control. These were mostly treatments that included a strobilurin fungicide with no added Tilt. The correlation between visibly scabby kernels and DON level, calculated on a plot basis for the ARC experiment, was significant, but low ($R=0.41$). Percentage of scabby kernels was not calculated for the SEPAC trial because the incidence appeared to be very low.

Yields at SEPAC were high, but those at ARC were mediocre. The differences in severity of leaf rust and head blight incidence would not likely account for this difference. The range in average yields at ARC was 18.4 bu/A and at SEPAC was 14.8 bu/A. Only one treatment at ARC (Caramba) yielded significantly more than the untreated control. No treatment yielded significantly more than the untreated control at SEPAC. There was no correlation between yields for the same treatments at the two locations. However, plots that received the Caramba treatment had the highest average yield at ARC and the second highest yield at SEPAC. In contrast, the Cornell biological 1 treatment had the highest yield at SEPAC but a poor yield at ARC. There were no significant differences in test weight at either location.

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(See table 1 on next page)

Table 1. Fusarium head blight incidence, yield, and scab severity in Clark soft red winter wheat treated with various fungicides at the Purdue Agronomy Research Center (ARC) and the Southeast Purdue Agricultural Center (SEPAC) during 2000

Treatment, rate per acre, and growth stage	ARC				SEPAC		
	FHB ¹ %	Yield Bu/A	Scabby kernels %	DON ppm	FHB ¹ %	Yield Bu/A	DON ppm
BAS 500 9 fl oz + MSO 1%, GS 37	5.4	53.1	2.0	2.7	2.7	86.3	1.1
BAS 500 12 fl oz + MSO 1%, GS 37	6.7	52.4	2.8	2.2	3.2	81.0	0.8
BAS 500 9 fl oz + Agridex 1%, GS 61	4.9	56.9	2.6	2.8	4.1	83.3	1.7
BAS 500 12 fl oz + Agridex 1%, GS 61	5.2	52.9	1.9	2.4	2.2	83.6	1.3
BAS 500 6 fl oz + Folicur 3.6F 2 fl oz + Agridex 1%, GS 61	5.1	55.6	1.3*	1.2*	2.9	84.5	1.2
Caramba 13.5 fl oz, GS 61	2.7*	63.1*	0.9*	0.9*	3.2	93.4	0.5
Cornell biological 1, GS 61	5.4	48.9	2.0	1.6*	2.2	94.5	0.7
Cornell biological 2, GS 61	6.3	44.7	1.2*	1.9	2.9	81.9	0.9
Folicur 3.6 EC 4 fl oz + Induce 0.06 % v/v, GS 37	5.5	51.8	1.8	2.4	2.8	79.7	0.9
Folicur 3.6 EC 4 fl oz + Induce 0.06 % v/v, GS 61	3.3*	53.5	1.1*	1.4*	2.5	92.7	0.7
Quadris 2.08 EC 6 fl oz, GS 37	6.6	54.6	2.1	2.2	2.8	84.5	1.1
Quadris 2.08 SC 9 fl oz + Benlate 4 oz, GS 61	5.2	57.1	2.0	3.1	3.1	85.1	1.3
Stratego 2.1 EC 14 fl oz + Induce 0.06%, GS 61	4.0*	55.2	1.6	1.4*	3.1	93.1	0.7
Tilt 3.6 EC 4 fl oz, GS 37	5.7	58.4	2.2	2.1	2.3	84.4	0.6
Tilt 3.6 EC 4 fl oz + Induce 0.06, GS 37	5.2	52.2	2.3	2.1	2.1	93.8	0.8
Tilt 3.6 EC 4 fl oz + Induce 0.06 %, GS 61	4.9	55.8	1.5	1.8	2.2	85.1	0.7
Untreated	6.4	55.5	2.2	2.6	2.6	86.3	0.5
LSD (0.05)	1.5	7.5	0.7	0.8	1.5	12.4	0.5

Within a column, values followed by an asterisk are significantly ($P=0.05$) lower (FHB, scabby kernels, or DON) or higher (yield) than the value for the untreated control.

At ARC, GS 37 treatments were applied 28 April; GS 61 treatments were applied 10 May, except for the Cornell biological treatments, which were applied 12 May.

At SEPAC, "GS 37" treatments were actually applied at GS 43 (25 April) and "GS 61" treatments were actually applied at GS 59 (5 May).

¹FHB = Fusarium head blight. Disease incidence (percentage of spikes showing symptoms) was rated 5 Jun at SEPAC and 6 Jun at ARC.

IDENTIFICATION OF BIOPROTECTANTS FOR CONTROL OF *GIBBERELLA ZEAE*

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OBJECTIVES

To identify microbial bioprotectants effective in controlling *Gibberella zeae* when applied to cereal spikes, seed, or crop residue, and to evaluate bio-compatible chemicals for their ability to interfere with perithecial development or ascospore release when applied to crop residue.

INTRODUCTION

There is a need for safe, affordable and efficacious biological and bio-compatible protectants in the integrated management of Fusarium head blight (FHB) caused by the pathogen *G. zeae*.

Screening of microorganisms to control FHB was initiated in Brazil over a decade ago (Luz 1988). In a glasshouse trial, an endospore-forming bacterial isolate not only reduced FHB on wheat but also reduced by 10-fold the contamination of grain with the mycotoxin deoxynivalenol (DON) (Stockwell et al., 1997). DON reduction, from a New York producer viewpoint, is the most important potential effect of bioprotectants.

We have evaluated candidate biological and bio-compatible protectants for use as a protective spray at flowering-time, and as seed and residue treatments. By these studies we intend to advance biological control of FHB of wheat closer to the ultimate goal of commercial application.

MATERIALS AND METHODS

A culture collection was assembled with organisms isolated from environmental sources in New York and 19 elite Brazilian accessions provided by Dr. Wilmar Luz of Embrapa Trigo. Emphasis was placed upon the selection of organisms which are likely to be robust under harsh field conditions. This resulted in the isolation and preservation, in 15% glycerol at -80°C, of 120 candidate biocontrol organisms from 70 different sources.

In vitro and glasshouse evaluations - A substantial portion of the biocontrol culture collection has been screened for antibiosis to *G. zeae*. Several of the more promising isolates were tested in a series of glasshouse experiments. Two of these experiments were designed to determine the optimal conditions for *G. zeae* infection and efficacy of the elite biocontrol isolate, TrigoCor 1448. The goal of additional glasshouse experiments was to evaluate other promising isolates for their ability to reduce FHB under controlled environmental conditions. Wheat heads in anthesis were sprayed to run-off with water (control) or a bacterial suspen-

sion (72 hour-old culture grown in nutrient broth with yeast extract [NBYE]) and allowed to air dry for 24 hours before inoculation with a conidial suspension (10^5 cfu/ml) of *G. zeae* macroconidia. Plants were incubated for 24, 48 or 72 hours under constant mist before being transferred to the glasshouse where they were rated for disease incidence 6 to 10 days after inoculation. Treatments were replicated 5 times.

Anthesis-time spray - Three elite biocontrol bacteria and one bio-compatible fungicide were added as treatments to the Uniform Fungicide Trial conducted at Aurora, NY. In this same trial, TrigoCor 1448 was combined with tebuconazole (4 fl oz Folicur) to determine if the combination would give enhanced FHB control over either treatment alone. Bacteria were grown for 24 hours in NBYE plus manganese ($2-4 \times 10^8$ cfu/ml) and diluted 50% for application. Treatments were applied at flowering (Feekes 10.5.1) using a tractor-mounted sprayer. Pathogen inoculum was from natural sources supplemented by ascospores released from *G. zeae*-colonized maize kernels scattered between the plots 3 weeks prior to flowering. Plots were assessed for disease incidence, % Fusarium damaged kernels (fdk), test weight, yield and DON content. Treatments were replicated 4 times and arranged in a randomized block design.

In order to evaluate additional promising biocontrol isolates under field conditions, 10 organisms and three binary combinations were applied to small plots (two adjacent rows 36 inch in length) in a separate 'mini-plot' experiment. Treatments, applied at anthesis using hand sprayers, were replicated 5 times and arranged in a randomized block design.

Seed treatment - The bioprotectant TrigoCor 1448 was included at three concentrations (10^9 , 10^{10} and 10^{11} cfu/100 lbs seed) as part of a larger field trial on the effect of seed treatments on seedling emergence, seedling weight, and grain yield of hard red winter wheat (cv. 'Crimson') with 25-32% incidence of seed infected with *G. zeae*. The bioprotectant was grown in broth (NBYE) with agitation, centrifuged and the pellet resuspended in 15 ml water. The treatments were coated onto the seeds and allowed to air-dry. Treatments were replicated four times and arranged in a randomized block design.

Debris treatment - Fourteen treatments (9 candidate biocontrol organisms, three bio-compatible chemicals, and an nontreated control) were applied to artificially-infested maize stalks and grain by immersing the plant tissue in the bacterial suspensions or chemical solutions for 3 minutes with constant agitation. Treated material was allowed to air-dry. In December 1999, the samples were placed in nylon pouches, arranged on the ground in a randomized design and allowed to overwinter under ambient field conditions. Samples were collected in Spring 2000 and were kept frozen until evaluated. Plant tissue was placed on moistened filter paper covered by an inverted plate of Komada's medium and incubated at room temperature under fluorescent UV lights for a maximum of 14 days. Perithecia were counted completely or with the use of a sampling grid. Ascospore discharge was determined by the presence of colonies of *G. zeae* on the surface of the agar.

RESULTS AND DISCUSSION

Glasshouse evaluation of biocontrol isolates

a) Timing - Incubation of wheat plants for 48 hours in a mist chamber following inoculation with a spore suspension (10^5 cfu/ml) of *G. zeae* macroconidia resulted in high incidence of infection (75% and 96%) and adequate seed set. Treatment of the wheat with TrigoCor 1448, 24 hours prior to inoculation with the spore suspension, reduced FHB incidence by 15 and 22% in the two experiments and increased 100-seed weight by 13 and 45%. TrigoCor 1448 was included as a benchmark in subsequent glasshouse evaluations of other candidate biological control organisms.

b) Isolate evaluation - Treatment with TrigoCor 1448 (average of five experiments) resulted in a 33.5% decrease in disease incidence and a 29.5% increase in 100-seed weight when compared to the nontreated control. Treatment with TrigoCor 4712 gave a 93% increase in 100-seed weight (average of two experiments). Several other isolates showed promising results as well. In the one glasshouse experiment analyzed for the presence DON toxin, treatment with TrigoCor 1448 and TrigoCor 4712 reduced the toxin content of the seed by 27 and 71%, respectively, compared to the nontreated control.

Anthesis-time spray - Two of the three bacterial isolates (TrigoCor 1448 and TrigoCor 4712) tested in the Uniform Fungicide trial at the New York location gave slight reductions in the % incidence of scabby heads and % fdk, although test weight and yield were not significantly different from nontreated. TrigoCor 1448 reduced FHB incidence and DON content 17 and 23% respectively, compared to the nontreated control. When Folicur (4 fl oz) was combined with TrigoCor 1448, % incidence of scabby heads and % fdk was the lowest and test weight the highest of any of the 12 treatments included in the trial. This combination reduced FHB incidence by 38% and DON contamination by 25% compared to nontreated wheat.

Significant effects of biocontrol agents on FHB were not demonstrated in the 'mini-plot' experiment, possibly due to the restricted plot size.

Seed treatment - TrigoCor 1448 at 10^{11} cfu/ 100 lbs seed increased seedling emergence but to a lesser extent than Raxil-Thiram. No treatment of scabby seed had a significant effect on seedling weight, test weight or grain yield.

Debris treatment - In nodal tissue of maize, only treatment with acetic acid (5% v/v) resulted in the complete absence of perithecia. All other treatments resulted in higher numbers of perithecia and ascospore discharge than the control (water). Internode stem pieces and kernels which were collected from the field site at later dates were in more advanced states of decomposition, but those treated with acetic acid also did not produce any perithecia when incubated under favorable laboratory conditions.

DISCUSSION

The modest success of biocontrol of FHB in the glasshouse and the field with the elite bioprotectant TrigoCor 1448 suggests the potential of bioprotectants as a component in an

integrative approach to FHB control. Combination of the bioprotectant with a chemical fungicide gave promising results. At the same time, despite excellent spray coverage and nearly perfect spray timing, no treatment reduced FHB or DON levels as dramatically as required for agriculture application. Reduction in disease incidence in biocontrol field tests falls within the range reported by other groups (Boehm et al., 1999; Luo and Bleakley, 1999). Future efforts will be focussed to gain a better understanding of the FHB biocontrol process and to improve efficacy of elite bioprotectants alone or in combination with chemical fungicides.

The complete inhibition of perithecial production by acetic acid (5% v/v) has redirected the search for effective products to control perithecial development and ascospore discharge. In future trials, infested maize debris will be treated with organic acids in a range of concentrations.

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