SESSION 4:

FHB MANAGEMENT

Chairperson: Gary Bergstrom

EFFECTS OF WHEAT GENOTYPES AND INOCULATION TIMINGS ON FUSARIUM HEAD BLIGHT (FHB) SEVERITY AND DEOXYNEVALENOL (DON) PRODUCTION IN THE FIELD.

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ABSTRACT

Fusarium head blight (scab), caused primarily by *Fusarium graminearum* (teleomorph: *Gibberella zeae*), is an important disease of wheat and other cereals worldwide. The disease affects both yield and quality due to contamination of grains with various mycotoxins. Since 1993, the disease has caused billions of dollars loss to the wheat industry in the USA. Due to lack of effective resistant cultivars, FHB is managed through fungicide applications and cultural practices. New fungicides such as 'Proline' are effective in FHB management and DON reduction. It has been hypothesized that all wheat cultivars do not respond to fungicide applications in similar manner for DON production and yield increase. Research is in progress to make the FHB forecasting system more accurate. Information on wheat cultivars with various levels of resistance to FHB and their responses to the disease development are important parameters of accurate disease forecasting system.

The main objectives of this study were to determine the effects of three hard red spring wheat cultivars, Glenn (FHB resistant), Steel-ND (moderately susceptible) and Trooper (susceptible), and two inoculation timings on FHB development, and to examine the correlation between FHB severity and DON production under field conditions. Wheat cultivars were planted on May 4 and May 14, 2007 at North Dakota State University Experimental Station, Fargo. The experiment was planted as a split-split plot design with 3 replications. Planting date (early and late), wheat cultivars (Glenn, Steel-ND, and Trooper), and inoculation timing (no inoculation, inoculation at early flowering, and inoculation at mid flowering) were assigned in main plot, sub-plot, and sub-sub plot, respectively. Plants were spray-inoculated with *F. graminearum* (~100,000 spores/ml). Two hundred-twenty-five heads from each sub-plot were examined for FHB incidence and severity, and 20-40 heads with disease severity of 0%, 7-21%, 22-50%, 51-79%, and 80-100% in each sub-plot were tagged at dough stage (Feekes GS 11.2). Wheat ear heads with each disease severity category were collected separately to estimate DON, and correlation between FHB severity and DON production. The cultivars differed significantly in FHB severity, but not in disease incidence and DON production. The resistant wheat cultivar Glenn has the lowest severity (20.6%) while the susceptible cultivar Trooper has the highest disease severity (28.12%). Inoculation timings also had significant effect on FHB incidence, severity, and DON production. All three disease components incidence (12.75%), severity (41%), and DON (2.45 ppm) were higher when the cultivars were inoculated at mid flowering stage (GS 10.52). A positive correlation ($r = 0.98$) was observed between FHB severity and DON concentration in all three cultivars. As expected, the susceptible cultivar Trooper had higher DON concentration in all five disease severity categories (ranged from 1.06 to 75.68 ppm) as compared to Steel-ND (1.39 to 56.86 ppm) and Glenn (0.91 to 64.63 ppm). The samples with high DON concentration also had with high amount of 3-ADON. Our results indicate that infection at mid flowering growth stage is crucial in FHB incidence, severity, and DON production.

AEROBIOLOGY OF *GIBBERELLA ZEAE*: WHENCE COME THE SPORES FOR FUSARIUM HEAD BLIGHT?

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ABSTRACT

Gibberella zeae (*Fusarium graminearum sensu stricto*) is the principal causal agent in North America of Fusarium head blight (FHB) of wheat and barley and in several regions is the predominant causal agent of stalk rot and ear rot of corn. Research done primarily in New York over the past decade on the aerobiology, epidemiology, and population biology of *G. zeae* was summarized in terms of its implications for the regional management of FHB. The fungus survives between crop seasons as a saprophyte in infected crop debris, especially in corn stalks and small grain residues on which it sporulates (both conidia and ascospores) profusely during warm, moist conditions. Viable spores of *G. zeae* rely on atmospheric motion systems for transport to the florets of wheat and barley where they initiate FHB. Ascospore liberation into turbulent air currents is favored by spore release during daylight hours when peak discharge from perithecia on corn residues also occurs. Ascospore survival on the surface of wheat spikes is on the order of hours to days. Using unmanned aerial vehicles (UAVs), we documented the abundance of viable spores of *G. zeae* 60 m above the surface of the earth at all times of the day and night under a broad range of meteorological conditions. Viable spores were deposited across cereal fields and other landscape areas by gravitational settling mainly at random and predominantly at night. The temporal uncoupling of peak spore release and deposition suggests that inoculum in cereal fields may originate from distant as well as within-field sources. Genotypic diversity was extremely high in atmospheric populations of *G. zeae* collected in central New York over a four-year period. The predominant trichothecene mycotoxin genotype of *G. zeae* found in New York in both infected grain and in atmospheric populations is one that produces deoxynivalenol (DON) plus smaller amounts of 15-acetyl-DON. Our findings suggest that atmospheric populations of *G. zeae* are an abundant, well-mixed, and diverse source of inoculum for regional epidemics of FHB. Model computations with the atmospheric transport model HYSPLIT suggest that ascospores of *Gz* may be dispersed kilometer distances from area sources of inoculum in a matter of minutes. The ability to predict the regional transport of *Gz* from local inoculum sources may help refine risk models for FHB.

It is generally considered (but not proven) that airborne ascospores of *G. zeae*, constitute the principle inoculum for infection of wheat and barley florets. We provided evidence that viable ascospores are potentially transported at least kilometer distances from their site of discharge. Yet the conventional opinion among FHB researchers from Chester in 1890 to the present is that inoculum sources for FHB are mainly local and that long-distance dissemination of inoculum is of minor significance. For example, FHB risk forecasting models that predict local inoculum levels based on previous local weather are built, in part, on the assumption that local inoculum is derived exclusively or largely from nearby sources. This assumption awaits validation. Significant long-range dispersal would suggest that local management of overwintered inoculum (e.g., tillage, spraying of debris, etc.) may have negligible impact on the development of FHB in nearby cereal crops unless performed over extensive production areas. Long-range dispersal also implies that genotypes of *G. zeae* with novel toxin or virulence profiles could be rapidly disseminated across broad geographic regions. Published studies suggest that most rain-splash dispersal of spores to spikes occurs within 5 meters or less from inoculum sources on the

soil surface and that disease severity follows a similar gradient with distance from those inoculum sources. Various researchers have attempted to delineate spore dispersal gradients or disease gradients at linear distances from area sources of inoculum. Observations of 50% reduction in spore concentration or disease have ranged from 1 to 50 m distances from area inoculum sources with most studies indicating sharp gradients within 10 m of sources. In almost every study conducted, the background level of spores or disease has been at 50% or greater proportion of the level at the source area. Ascospores actively discharged from perithecia or even conidia caught in turbulent air may be deposited on local wheat spikes at a potentially much greater distance from debris than splash-dispersal. Spores may also escape the crop canopy, mix with spores over a wide area, and be transported in the atmosphere at least kilometer distances. Field survey-based studies of DON in grain have generally revealed that cereal cultivar and seasonal meteorological conditions were better quantitative predictors of toxin content than previous crop or tillage practice, strongly suggesting that regional inoculum plays a critical role in FHB epidemics. Based on several field studies with cereal debris level and crop sequence, within-field inoculum, where present, appears to be a significant source for local FHB, but regional inoculum appears to play an even greater role.

There are no reliable estimates of the relative contributions of within-field, local inocula to spike infection compared to other airborne sources. In New York and Virginia, we are utilizing a marked isolate, release-recapture experimental approach to assess relative contribution of localized clonal inoculum present in corn stalks to infection of wheat heads at varying distances from area sources of inoculum. Preliminary evidence from the first year of experimentation suggests that within-field sources of *G. zeae* provided a minor fraction of FHB inoculum compared to background atmospheric sources in a non-epidemic situation in New York and in a moderate epidemic situation in Virginia.

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2007 UNIFORM FUNGICIDE TRIALS ON SOFT WHITE WINTER WHEAT IN MICHIGAN.

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ABSTRACT

The objective of this project was to evaluate the effectiveness of commercially available and experimental fungicides for the control of Fusarium head blight (FHB) and reduction of deoxynivalenol (DON) in white winter wheat in Michigan. Four trial locations were planted in with Caledonia white winter wheat during 8 – 31 Oct 2006. Plots at the East Lansing and Clarksville locations were artificially inoculated with *F. graminearum* infested corn kernels at a rate of 0.2 oz/sq. ft. a week prior to heading. Irrigation was begun after inoculation was completed. Plots were irrigated four times /day for 20 minute intervals beginning the week prior to flowering until two weeks after flowering. The remaining two sites in Sandusky and Saginaw relied on natural inoculum and rainfall. All treatments were applied at early flowering (Feekes 10.5.1) at 25gpa and 40 psi using a CO_2-pressurized R&D tractor mounted spray boom with XR11003VS nozzles positioned forward and backward. Treatments consisted of: 1) Folicur (tebuconazole) 4 fl oz/a; 2) Proline (prothioconazole) 5 fl oz/a; 3) Caramba (metconazole) 13.5 fl oz/a; 4) Topguard (flutriafol) 14 fl oz/a; 5) Punch (flusiazole) 6 fl oz/a; 6) Proline 3 fl oz/a + Folicur 3 fl oz/a; and 7) untreated control.

Disease pressure (FHB and foliar diseases) in Michigan was generally very low in 2007. Plots were rated for foliar diseases 7 days after treatment and again at the soft dough stage (Feekes 11.2). Saginaw did not develop sufficient foliar disease for rating. FHB incidence, severity and index were rated at the soft dough stage. Only the Clarksville and East Lansing (both inoculated and irrigated) locations developed sufficient FHB for field ratings. Yield, test weight, percent moisture, *Fusarium* damaged kernels (FDK) and thousand grain weights were determined post harvest. Sub samples from each plot were sent to the University of Minnesota for DON analysis.

At the East Lansing location, FHB severity for all treatments was significantly lower (3.2-4.9%) than the control (21.9%). For FHB incidence, Punch (7.8%) was not significantly different from the control or other treatments, but other treatments (5.7-5.9%) were significantly lower than the control (21.9%). FHB index for all treatments (2.0-3.5%) was lower than the untreated control (21.9%). There were no significant differences in DON levels (1.1-1.9 ppm) among any treatments. Yields ranged from 72.1-87.9 bu/a. Proline + Folicur (87.9 bu/a) and Caramba (87.6 bu/a) were significantly higher than for Folicur alone (72.1 bu/a), but no treatments were significantly higher than the untreated control (73.7 bu/a). There were no significant differences among test weights or 1000 grain weights at the East Lansing location.

At the Clarksville location, there were no significant differences in FHB incidence (55.0-98.1%), severity (12.9-26.4%) or index (8.7-26.3%). There were no significant differences among treatments for test weight, 1000 grain weights, or yield (73.9-85.4 bu/a). Average DON levels ranged from 3.6 -10.1 ppm, but none was significantly different from the untreated control (6.8 ppm). All the treatments resulted in significantly less stagonospora and leaf rust than the untreated control. No phytotoxicity was observed in any of the treatments at any of the sites.

DURATION OF POST-FLOWERING MOISTURE AND INFECTION TIMING AFFECT ON FHB AND DON IN WHEAT.

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ABSTRACT

Our understanding of how environmental and host genetic influences interact to determine DON concentrations in small-grain spikes is incomplete. High levels of DON have sometimes been observed in the absence of abundant disease symptoms. This multi-year experiment explored the influences of post-flowering moisture duration, infection timing, and cultivar resistance differences on FHB and DON in winter wheat. The experiment had a split-plot design. Whole plots were four durations (0, 10, 20, or 30 days) of post-anthesis misting. Sub-plots were soft red winter wheat cultivars, of which one (2005) or two (2006 and 2007) were susceptible to FHB and six were moderately resistant. There were two plots of each cultivar under each duration of irrigation: one inoculated at anthesis with a backpack sprayer, and one in which individual funnel-isolated spikes were chosen at random and inoculated with a spray bottle at specific post-flowering intervals in order to study the effect of late infection. Inoculations utilized *F. graminearum* spore suspensions of 10^4 (2005) or 10^5 (2006 and 2007) macroconidia/ml. All treatments were replicated three times. In the backpack-inoculated plots, disease incidence and severity were assessed prior to the onset of senescence, and a DON time-course study was performed by collecting spike samples six times at 10-day intervals starting two weeks after flowering. Samples of all treatments were assayed for *Fusarium*-damaged kernels (FDK), percent infected kernels (using Komada's medium), and DON concentration. Assays of *F. graminearum* DNA by tissue type (kernel, rachis, or glume) were performed on a limited sample in 2005, using real-time PCR, and these assays are being conducted for all treatments in 2006 and 2007.

Preliminary results:

1) Under conditions conducive to disease (2006 and 2007), FHB incidence and severity and grain DON concentrations increased with increasing duration of post-flowering moisture (*P* d" 0.05). Cultivar grain DON rankings changed under longer moisture durations, suggesting that resistance to post-flowering moisture may be a distinct trait.

2) In 2006, spikes inoculated 10 days after flowering contained significantly less grain DON at harvest time than those inoculated at flowering ($P < 0.0001$). Spikes inoculated 20 days after flowering had still less harvest-time grain DON than those inoculated 10 days after flowering ($P < 0.0001$), and had the same level of grain DON as noninoculated spikes (*P* d" 0.48). Changes in DON rankings suggested that resistance to late infection may also be a distinct trait. (Data from 2007 not yet available.)

3) In 2006, for spikes inoculated 10 days after flowering, 0 and 10 days of post-flowering mist resulted in mean harvest-time grain DON levels of 0.6 and 1.2 ppm, respectively, while 20 and 30 days of post-flowering mist resulted in harvest-time grain DON levels of 2.0 and 2.4 ppm, respectively. At the same time, the percentage of FDK from spikes inoculated 10 days after flowering was significantly lower than that from spikes inoculated at flowering ($P < 0.0001$), and not significantly different from the FDK percentage from

noninoculated spikes ($P = 0.40$). Thus, late infections coupled with extended post-flowering moisture may be one scenario accounting for observations of visually healthy grain with excessive DON at harvest. (Data from 2007 not yet available.)

4) In 2005 and 2006, the time-course study results showed a significant decline in grain DON between mid-May and early June, which is normal harvest time. In 2006, DON progression was evaluated under varying durations of post-flowering moisture. Prolonged moisture delayed the DON decline, and raised the DON levels from which decline commenced, but DON levels continued dropping in those treatments during the three weeks after normal harvest time. This suggests that DON may actually be reduced by delaying harvest if DON levels are high early in grain-fill.

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DISCLAIMER

Any opinions, findings, conclusions, or recommendations expressed in this publication are those of the author and do not necessarily reflect the view of the U.S. Department of Agriculture.

EFFECT OF POST INOCULATION MOISTURE ON DEOXYNIVALENOL ACCUMULATION IN *FUSARIUM GRAMINEARUM*-INFECTED WHEAT.

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OBJECTIVE

The objective of this study was to examine factors; including host genetics, pathogen aggressiveness and environmental moisture, affecting the production and accumulation of deoxynivalenol (DON) in wheat.

INTRODUCTION

Fusarium head blight (FHB) or scab of wheat and other cereals, primarily caused by *Fusarium graminearum*, has reemerged as a devastating disease in the United States. The disease causes yield losses both from reduced grain number and grain weight, including the formation of tombstones (shriveled kernels with a chalky appearance). FHB also affects quality of the grain through the production of a range of mycotoxins, among which DON is the most commonly present and that which is regulated in the grain trade. Though the production and accumulation of DON in infected grain is generally positively correlated to FHB severity, the correlation is not consistent and predicting the DON content of grain in commercial wheat and barley crops or breeding nurseries based on visual disease assessments prior to harvest is generally unreliable. The production and accumulation of DON in infected grain is not well understood and likely results from the complex interactions of host and pathogen genetics which is modified by the prevailing environmental conditions. The objective of this multi-year study was to identify factors affecting the production and accumulation of DON in wheat.

MATERIALS AND METHODS

Experiments were conducted at the St. Paul Experimental Station of University of Minnesota in 2006 and 2007 as split-split plot designs, each with five replications. Main plots were days of mist-irrigation after inoculation (14, 21, 28 and 35 days after inoculation [DAI]), sub-plots were wheat genetics and sub-sub plots were individual *F. graminearum* isolates (49-3, B63A, Butte86-ADA11, 81-2, B45A) differing in their relative aggressiveness and a mock-inoculated (water) control. Two row plots (each 1.8 m long) of three wheat varieties; Alsen (moderately resistant, resistance source Sumai 3), 2375 (moderately resistant, unknown resistance source) and Wheaton (susceptible) were planted in mid-April each year of the study. All plots were inoculated at the anthesis (mid-June) and 3 days later with macroconidial inoculum (1×10^6 macroconidia ml^{-1}) using a CO_2-powered backpack sprayer dispensing inoculum at the rate of 30 ml per meter of row. Mist-irrigation was started immediately following the first inoculation. Disease was assessed visually 21 DAI by counting total infected spikelets in 20 arbitrarily selected heads in each plot (10 heads per plot row). Grain was harvested at maturity (late July), machine threshed and dried for 10 d at 95^0 C. The percentage of visually scabby kernels (VSK) analysis was assessed on a 25 g sub-sample of harvested grain following the procedures of Jones and Mirocha (1999). Following the assessment of VSK the sub-samples were analyzed for DON at the University of Minnesota's Mycotoxin Laboratory. Data were analyzed by ANOVA and LSD tests and correlations performed using SAS.

RESULTS AND DISCUSSION

In 2006, the average FHB severity was 22.3%, the average VSK was 4.9% and the average DON accumulation was 0.62 ppm. Overall, FHB severity, VSK and DON was higher in 2007 than 2006. In 2007, the average FHB severity was 37.5%, and the average VSK was 27.8% while the average DON concentration was 10.5 ppm. The effects of *F. graminearum*

isolate, wheat genetics and mist-irrigation were significant in both years of the study. While the isolate 49-3 generated the highest FHB severities, VSK and DON production in 2006, B63A had the lowest DON levels despite inciting high FHB severities and VSK. In 2007, isolates B63A and 49-3 produced the highest levels of DON and were associated also with higher FHB severities and VSK. Isolates 81-2 and Butte86-ADA11 generally were associated with lower FHB severities, VSK and DON.

In 2006 the severity of FHB and the percent VSK was significantly higher in Wheaton (42.5% and 11.5%, respectively) than the other two varieties (FHB severity < 15.5%, VSK < 2.9%). The DON concentration of Wheaton, across all isolates, was significantly higher (1.2 ppm) than for the other two wheat varieties tested (< 0.4 ppm).

FHB severity and VSK was significantly lower in the treatments receiving the least amount of mist-irrigation (14 DAI; FHB severity 19%; VSK 4%) than longer mist-irrigation treatments (FHB severity 22.6-25.4%: VSK > 5%). The DON concentration was however significantly lower in the longest mist-irrigation treatment (35 DAI; DON 0.5 ppm) than in treatments where the mist-irrigation was applied for shorter periods of time (DON 0.6 – 1 ppm). The Spearman's rank correlations of DON with FHB severity and VSK were 0.78 ($P < 0.0001$) and 0.85 ($P < 0.0001$), respectively.

Similarly, in 2007 Wheaton had significantly higher FHB severity (59%) VSK (53.87%) and DON (17.64 ppm), than the other wheat genotypes examined (FHB severity < 27.51%; VSK < 19.4%; DON < 7.45 ppm). The severity of FHB was highest in mist-irrigation treatments applying supplemental water for 28 DAI (40.3%) than the other mist-irrigation treatments (36.1-36.8%). VSK readings were significantly higher (37.7%) for the longest mist-irrigation treatment (35 DAI) than the others (19-33.2%). DON was significantly lower (7.95 ppm) in the 35 DAI mist-irrigation treatment compared to the other irrigation treatments (9.9-13.3 ppm). The Spearman's rank correlations of DON with FHB severity and VSK were 0.78 ($P < 0.0001$) and 0.78 ($P < 0.0001$), respectively.

Our results show that FHB severity, VSK and DON level increases in more susceptible wheat cultivars. It also varies with the fungal isolates aggressiveness with respect to disease and DON production. DON level increased with mist-irrigation applied till 28 DAI but was reduced in the 35 DAI irrigation treatments. These results are in concordance with several researches which reported a reduction in DON levels with the long irrigation treatments. Similarly, it has been reported that DON accumulation in *Fusarium*-infected tissues peaks approximately six weeks after infection and then declines prior to harvest. In our case, the peak DON level was observed between four and five weeks after inoculation. Since DON is water soluble, the decline in DON levels might have been accelerated by the mist-irrigation, perhaps from leaching of the DON. The observed increase in DON levels despite mist-irrigation until 28 DAI was likely due ongoing production of DON as the fungus continues to grow and infect new tissues under conditions favorable for the pathogen. Thus, any DON leached by mist-irrigation water before 28 DAI would likely have been replaced by that produced by the spreading fungus. As the plant begins senescence, growth of the fungus and the production of DON may be reduced or even stop, and thus the leaching effect of irrigation on reduction of DON was readily detectable. Based on our results it may be concluded that longer durations of wetting, from either mist-irrigation or rainfall, after infection will increase the severity of FHB and VSK and thus the damage to grain, although DON concentrations may be reduced.

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DISCLAIMER

Any opinions, findings, conclusions, or recommendations expressed in this publication are those of the authors and do not necessarily reflect the view of the U.S. Department of Agriculture.

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PROSARO® – A NEW FUNGICIDE FOR CONTROL OF *FUSARIUM* AND MYCOTOXINS IN CEREALS.

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ABSTRACT

Fusarium head blight (FHB) and mycotoxins can be major challenges in cereal production. FHB is a disease caused by different *Fusarium* species. Under field conditions, all the predominant *Fusarium* species may produce mycotoxins with the exception of *M. nivale*. Quality and quantity of grain harvest are affected by FHB.

Combatting FHB is demanding because many factors may influence the severity of the infection. Agricultural practices (cultivars, cropping methods, crop rotation etc.) and environmental conditions are all contributing factors to any epidemic. Chemical control with *Fusarium* active compounds such as the triazole fungicides Prothioconazole and Tebuconazole contribute significantly to a reduction of FHB and mycotoxin production.

Suppression of the mycotoxins with these fungicides can reach values of higher than 70%. Success of the fungicide treatment is dependent on optimal application timing and spray coverage. In general, smaller droplet sizes provide more effective *Fusarium* control.

With the mixture of Tebuconazole & Prothioconazole – Prosaro ®- the application window can be broadened due to the combination of preventive and curative strengths of the two active ingredients involved. In addition to FHB control, Prosaro controls a broad range of fungal pathogens in cereals and thus contributes to higher yield.

ADDITION OF ADJUVANT TO IMPROVE COVERAGE AND FUNGICIDE EFFICACY ON BARLEY, LANGDON 2006

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INTRODUCTION

Most fungicide application technology studies have focused on maximizing deposition on the entire small grain head. Observations in the field would indicate that the Fusarium head blight (FHB) ascospore can infect the awn part of the grain head but generally will not move down the awn and infect the other parts of the spike or kernel. The awns are a very effective structure for collecting both ascospores that cause the initial infection for FHB and unfortunately the fungicide solutions we apply to protect against FHB. Most fungicides applied to control FHB have a localized systemic type of activity meaning that there is little translocation within the grain head. In addition, most of the translocation is in an upward and outward movement from the initial point of deposition. Studies have been initiated to determine if we can increase the amount of fungicide solution collected on the whole head and reduce the percentage of fungicide solution collected on the awns relative to the spike. This study reports results of those efforts. It includes evaluation of the addition of several adjuvants, suggested by adjuvant manufacturers, and two adjuvant compounds previously reported to significantly increase coverage on the other parts of the spike or kernel portion of the grain head.

MATERIALS AND METHODS

A study was initiated in 2006 at the North Dakota State University Langdon Research Extension Center, Langdon North Dakota. The objective of the study was to determine if an adjuvant included with fungicide could increase deposition on the grain head as a whole, the developing kernel portion of the spike and subsequently reduce deposition on the awn portion of the head decreasing FHB disease incidence and deoxynivalenol concentration in the harvest sample. The study was designed as a randomized complete block with five replicates. A block of barley was planted with a double-disk type drill, rows spaced 7-inches apart to cultivar 'Stellar' in mid May. After emergence and after application of weed control, the block was divided into plots 12 x 30 ft. Prosaro fungicide, prothioconazole, was applied at 3.25 fl oz/ acre which is one-half the rate recommended by manufacturer Bayer CropScience. The one-half rate was used with expectation that we could maximize and measure the beneficial effect of the adjuvant. A food grade dye, FD&C blue #1, was mixed with each of the fungicide solutions at a rate of 44 grams per acre. The dye was included as an indirect type measurement to determine differences in coverage on the parts of the grain head. After delineation of the plots, a *Fusarium* inoculum was hand-broadcast on each plot to encourage development of disease. Fungicides were applied with a tractor using a side-mounted spray boom. The tractor traveled 6 mph delivering the solution at 10 GPA and 40 psi with Spraying Systems XR8002 nozzles angled 30 degrees downward from horizontal and oriented to spray forward or the same direction of travel as the tractor. The spray system was equipped with a CO_2 type delivery system instead of a standard pump. After applying the treatments, a sample of 10 heads were collected from each plot, deposited in Ziploc type bags and placed on ice. The awns of each head were individually clipped from the kernels. The awns and the remainder of the heads were deposited in separate 250 ml Erlenmeyer type flasks and sealed with a rubber stopper. A solution of 80 ml 95% ethyl alcohol was added to each flask and shaken for three minutes with a Burrell wrist-type action shaker. A sub sample of the solution was placed in a cuvette

and placed in a Jenway photospectrometer to determine the absorbance of the solution. Each absorbance reading was indirectly used to determine differences in the amount of dye collected on the grain head parts. A whole head sample was the sum of the parts. After the fungicide was applied, a sprinkler irrigation system was installed to modify the environment as needed and encourage the development of disease to determine differences among treatments. North Dakota State University Extension recommended production practices for barley in Northeast North Dakota were followed. A visual estimation was made from 20 samples per plot collected 20 days after fungicide application to estimate the incidence (number of spikes infected) and field severity (number of FHB infected kernels per head divided by total kernels per individual spike) of FHB in each plot. A rotary mower removed the front and back five feet from each plot prior to harvest to minimize any chance of interference by drift from the tractor when stopping or starting. Each plot was harvested with a Hege plot combine and the grain sample cleaned and processed for yield, protein, plump, and test weight. A sub sample was ground and analyzed for deoxynivalenol (DON) by North Dakota State University. Data was analyzed with the general linear model (GLM) in SAS. Fisher's protected least significant differences (LSD) were used to compare means at the 95% probability level.

DISCUSSION AND RESULTS

The environment at the LREC was warm and dry both before and after fungicide application in 2006. Fusarium head blight developed later in the season and may have negated some of the beneficial effects of the adjuvants and the fungicide. The fungicide reduced FHB field severity over the untreated but no differences were measured among treatments (Table 1). Although DON levels were reduced by more than 50% from the untreated by WECO 6065 and AG 6470, they were not statistically significant. Several adjuvants increased the deposition on the whole head and there were differences recorded among adjuvants. The most notable was the adjuvant In-Place which is an encapsulating compound that could be used with an additional adjuvant to further increase deposition, distribution on the head, and fungicide efficacy. Also of note was the low deposition value of the Silkin adjuvant. Silkin is an organosilicate type adjuvant. The results may be a rate related effect and may be improved with the addition of another type adjuvant. Syl-Tac is also a silicon type adjuvant that includes a penetrator and performed considerably better. No differences were measured on the kernel portion of the spike. Significant correlations were determined between FHB incidence and yield, test weight and deposition on the awns, and most notably DON levels with deposition on the spike, awns, and whole head indicating that these efforts are focusing in the right areas. Significant negative correlations were measured between coverage parameters and DON levels.

Table 1. FHB incidence and field severity, yield, test weight, plump, coverage, and deoxynivalenol concentration (DON) by treatment, Langdon 2006.

Treatment/ Adjuvant	Adjuvant Rate	FHB Incidence (%)	FHB Field Severity (%)	Yield (bu/a)	Test Weight (lb/bu)	Plump (%)	Absorbance Spike	Absorbance Awns	Absorbance Whole	DON (ppm)
Syl-Tac	0.5% v/v	100	10.4	132.5	46.7	97	.114	.336	.450	1.84
Untreated		100	14.8	116.4	47.2	98	.054	.143	.197	1.80
AG06038	0.5% v/v	100	11.9	126.7	47.0	98	.130	.321	.451	1.64
no adjuvant		100	10.5	130.9	47.3	98	.116	.347	.463	1.62
WECO5036-7	0.25% v/v	99	10.6	117.9	47.6	98	.105	.296	.401	1.34
Triton X405	0.25% v/v	99	10.1	131.7	47.0	98	.123	.335	.458	1.20
Silkin	0.25 pint/100	100	10.0	127.6	47.3	98	.089	.281	.370	1.12
Preference	0.25% v/v	99	11.1	124.4	47.4	98	.108	.339	.448	1.10
Alfonic 1412-80	0.25% v/v	100	11.2	128.7	47.5	98	.108	.406	.514	1.08
In-Place	1/4 (adj./fung)	100	10.0	121.4	46.9	98	.137	.413	.551	1.02
AG 5004	8 fl oz/a	100	10.2	124.4	47.2	98	.121	.379	.500	1.02
Induce	0.125% v/v	100	10.7	127.2	47.4	98	.120	.322	.441	0.96
WECO6065	0.25% v/v	99	10.6	124.3	47.6	98	.084	.297	.381	0.86
AG06470	1%v/v	100	10.8	126.6	47.3	98	.139	.301	.441	0.76
LSD(0.05)		NS	2.1	NS	NS	NS	NS	.079	.103	NS
% C.V.		1	15	9	1	1	36	19	19	65
Pr>F		0.6947	0.0056	0.5656	0.1053	0.5381	0.1193	<0.0001	<0.0001	0.5012

Table 2. Pearson correlation coefficients for FHB incidence and field severity, yield, test weight, plump, coverage, and deoxynivalenol concentration (DON) Langdon, 2006.

	Incidence	Field Severity	Yield	Test Weight	Plump	Absorbance Spike	Absorbance Awns	Absorbance Whole	DON
Incidence	1.00	0.124	-0.255	-0.133	-0.153	-0.045	-0.067	-0.068	0.048
		0.308	0.033	0.272	0.204	0.714	0.584	0.545	0.692
Field Severity		1.00	-0.025	0.138	-0.162	-0.106	-0.212	-0.204	0.221
			0.840	0.253	0.181	0.381	0.078	0.090	0.066
Yield			1.00	-0.124	-0.275	0.113	0.234	0.234	0.111
				0.306	0.021	0.352	0.051	0.063	0.361
Test Weight				1.00	0.193	-0.106	0.143	0.072	-0.256
					0.109	0.381	0.239	0.5560	0.032
Plump					1.00	-0.185	-0.143	-0.180	-0.123
						0.125	0.239	0.137	0.310
Spike						1.00	0.448	0.719	-0.315
							<0.001	<0.001	0.008
Awns							1.00	0.943	-0.235
								<0.001	0.050
Whole								1.00	-0.300
									0.012
DON									1.00

ASSESSMENT OF AIR STREAM SPEED WITH TWO NOZZLE TYPES AS A TOOL TO IMPROVE DEPOSITION OF FUNGICIDE FOR CONTROL OF FHB IN WHEAT.

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OBJECTIVES

To determine most effective air stream speed using two contrasting nozzle types to maximize deposition on the grain spike and improve efficacy of fungicide on hard red spring wheat.

MATERIALS AND METHODS

A field was selected near Esmond, North Dakota that was previously cropped corn. The field was planted to 'Alsen' spring wheat in an east/west direction with an air seeder with tramlines every 80 feet. The study was arranged as a factorial (nozzle type x air stream speed) in a randomized complete block design laid out in four replicated blocks, split into plots 40 x 500 ft. to accommodate one spray boom and the grower's combine straight cut header. Plots were arranged in an east/west direction between tramlines and the plot length measured with a global positioning unit mounted on an all terrain vehicle after all herbicide applications had been completed. The sprayer was a Hardi-Twin (Hardi, Davenport, IA 58206) modified to accommodate the tramlines and to spray one half of the area between the trams, 40 feet width, beginning at the center of the tractor. The sprayer contained a diaphragm type pump and traditional flat fan hydraulic nozzles. The spray nozzles are mounted to direct the spray into the air stream which carried the spray solution to the grain. Before the field trial were completed, the spray booms were equipped with Teejet XR11003 and TT11003 nozzles (Spraying Systems Co, Wheaton, IL 60189)on each boom, respectively and calibrated. The nozzles were directed to spray forward from vertical at the maximum of 30 degrees forward. Both nozzles were calibrated at 40 psi to determine the output of the specific nozzles. The air stream speed was determined by setting the rpm on the fan at 1800, 2400, or 2900. The air stream velocities were about 23, 35 and 50 mph respectively, measured at one-inch from the air stream orifice. This was measured with a 'Kestrel' 2000 wind velocity meter (Niche Retail, Sylvan Lake, MI 48302). The spray drop size measurement application parameters were characterized by mounting two water sensitive papers (WSP) 1" vertically x 30" horizontally" side by side on a piece of flat iron at canopy height and spraying across the WSP. Each combination of the respective factors was measured. Volume median diameter (VMD) of the spray drops formed on the WSP, spray volume, and % area coverage was completed with a WRK 'Droplet Scan analyzer' Cabot, Arkansas.

The fungicide was applied at Feekes growth stage 10.51. The fungicide solution included Folicur (tebuconazole) fungicide 4 fl oz/acre + Induce adjuvant at 0.125% v/v and a food-grade tracer dye (FD & C Blue #1) added at 44grams/acre. Folicur is manufactured by Bayer CropScience and Induce by Helena Chemical Co. Immediately after the fungicide was applied, ten heads were sampled from each plot in each of three locations and placed in 250 ml Erlenmeyer flasks, sealed with a stopper, and placed on ice for transport to the laboratory to measure the relative volume of solution collected for each of the treatments. Barley production recommendations from the North Dakota State University Extension Service for northeast North Dakota were followed.

Eighty ml of 95% ethyl alcohol was added to each flask and shaken for three minutes with a wrist-action mechanical shaker (Burrell Scientific Instruments and

Laboratory Supplies, Model BT, Pittsburgh, Pennsylvania 15219). A sub sample of the wash solution was measured with a Jenway spectrophotometer (Jenway, Model 6300, Dunmow, Essex CM6 3LB England) and an absorbance of the tracer dye recorded to determine differences among application parameters. The absorbance reading quantifies differences in the amount of tracer dye deposited on the grain spike (a larger absorbance value is the result of more tracer dye in the solution).

A visual estimation of disease incidence (number of spikes infected) and field severity was made from 20 heads per plot at early dough stage. Field severity rating is the number of FHB infected kernels per head divided by total kernels per individual spike. All plots were harvested with a Caterpillar Lexion combine on 7 August and weighed with a weigh wagon and a grain sample collected. The yield was determined from the grain collected from the harvested plot. The grain sample was cleaned and processed to determine plump, test weight, and protein. A sub sample was ground and analyzed for the toxin deoxynivalenol (DON) by North Dakota State University. Data was analyzed with the general linear model (GLM) in SAS. Fischer's protected least significant differences (LSD) were used to compare means at the 5% probability level.

RESULTS

The VMD measured with water sensitive paper describes the relative drop size of the spray deposits from each of the nozzles. Half of the spray volume is in drops larger than this size in microns and half of the spray volume is in drops smaller than this value. The XR11003 nozzles had a smaller VMD than the TT11003 nozzle. Increasing air speed increased the VMD by spreading the drop over a larger area. Adding air as a spray solution carrier increased the area of coverage on the cards and increasing air stream speed increased area of coverage further.

Deposition (Absorbance) on the grain heads were the same with both nozzles when no air was used to assist in deposition. The XR11003 nozzle deposited more tracer dye than the TT11003 nozzle. The 2900 rpm air stream speed was statistically the same as no air but greater than the 1800 and 2400 air stream speed when averaged across both nozzle types indicating a benefit to the high speed air stream.

A characterization of the nozzles (F025_110) supplied with the Hardi-Twin and operating parameters traditionally used by the grower, indicated a reduction in VMD when nozzle pressure was increased and a large reduction in coverage of the cards when nozzle pressure was 90 psi. This indicates that the sprayer may provide better consistency of fungicide application due to increased deposition on the wheat head when operated with lower nozzle pressure and increased air stream speed.

The growing season was very dry during flowering and no measurable disease developed. Only the disease levels on the untreated plots were assessed, FHB incidence and field severity were found to be 6.3 and 0.5 percent, respectively. There were no differences among treatments in yield, test weight, plump, or deoxynivalenol concentration.

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The authors wish to acknowledge the cooperation of Bill and Louis Arnold, Esmond, ND. This material is based upon work supported by the U.S. Department of Agriculture, under Agreement No. 59-0790-3-079. This is a cooperative project with the U.S. Wheat & Barley Scab Initiative.

DISCLAIMER

Any opinions, findings, conclusions, or recommendations expressed in this publication are those of the author(s) and do not necessarily reflect the view of the U.S. Department of Agriculture.

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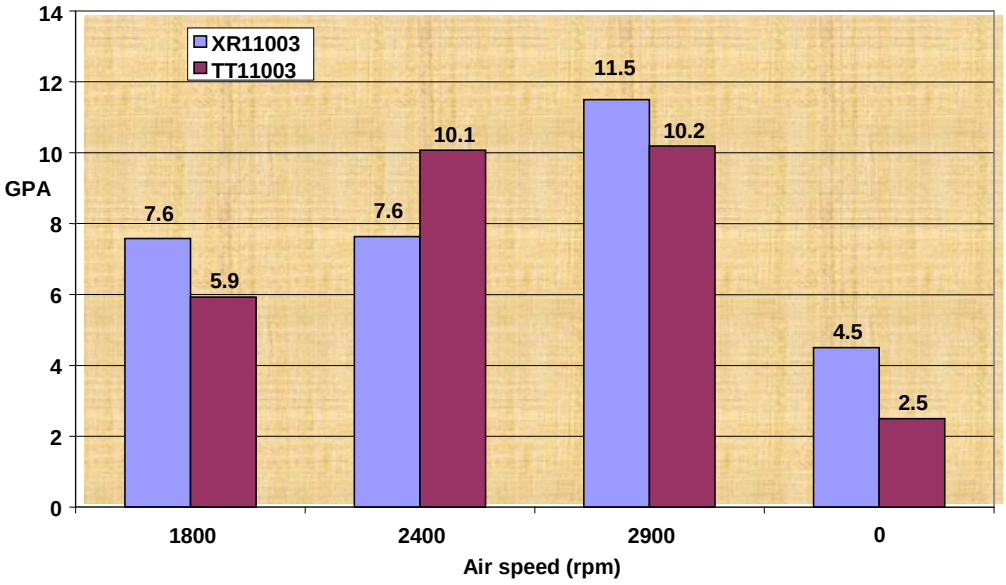

Figure 1. Volume Median Diameter (VMD) drop size produced using XR11003 and TT11003 nozzles at varying spray air delivery speeds.

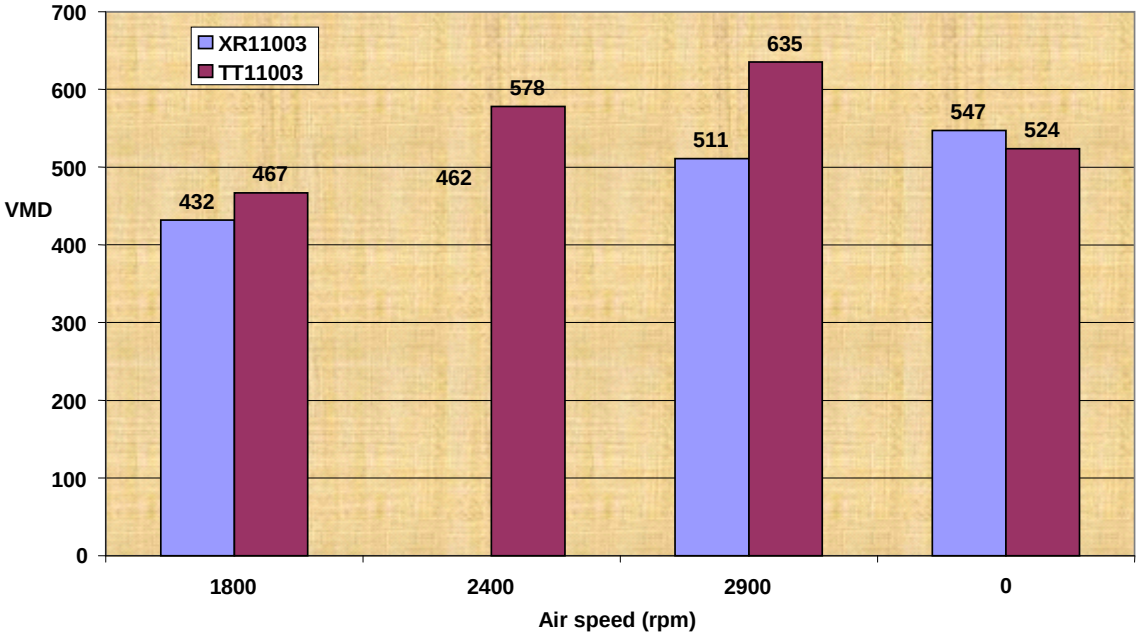

Figure 2. Estimated spray volume in GPA using XR11003 and TT11003 nozzles with varying spray air delivery speeds.

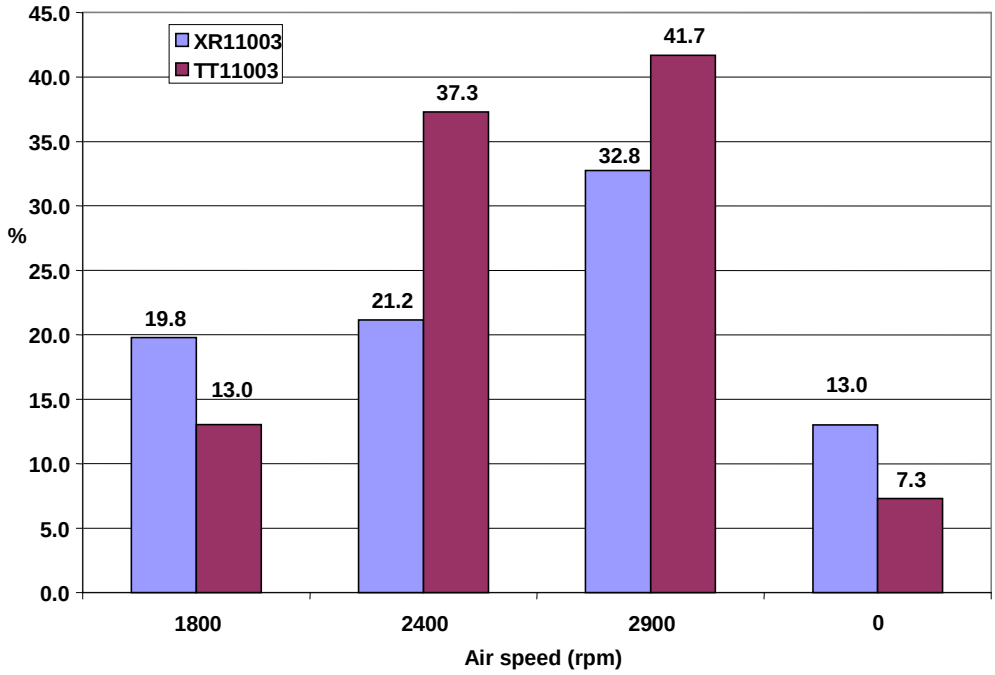

Figure 3. Relative percent area of water sensitive paper coverage using XR11003 and TT11003 nozzles with varying spray air delivery speeds.

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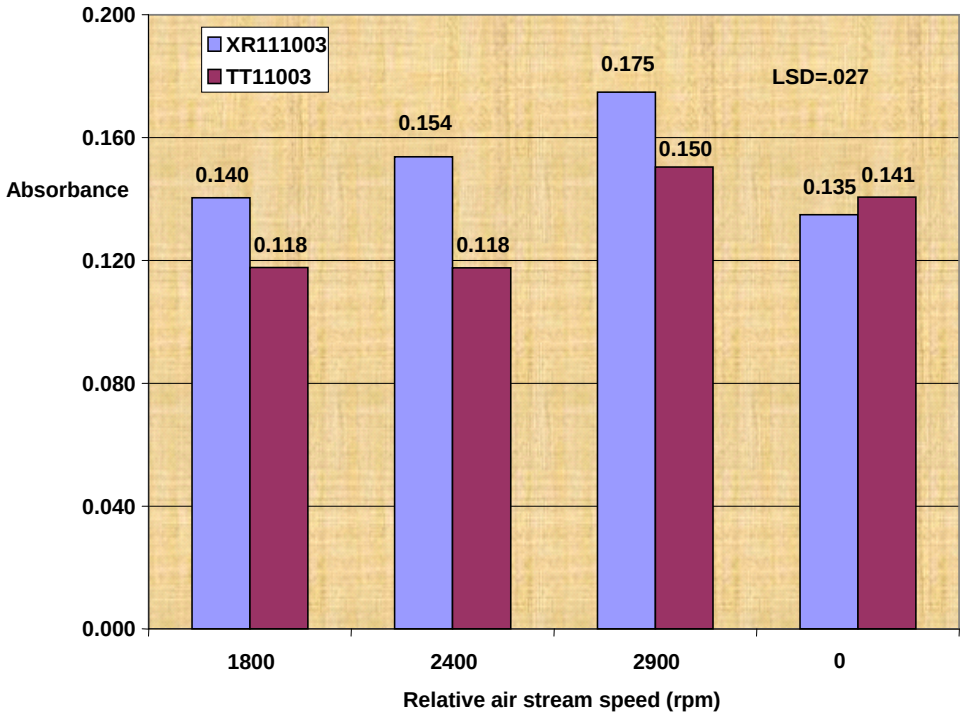

Figure 4. Relative deposition of spray on wheat heads using XR11003 and TT11003 nozzles in varying spray air delivery streams.

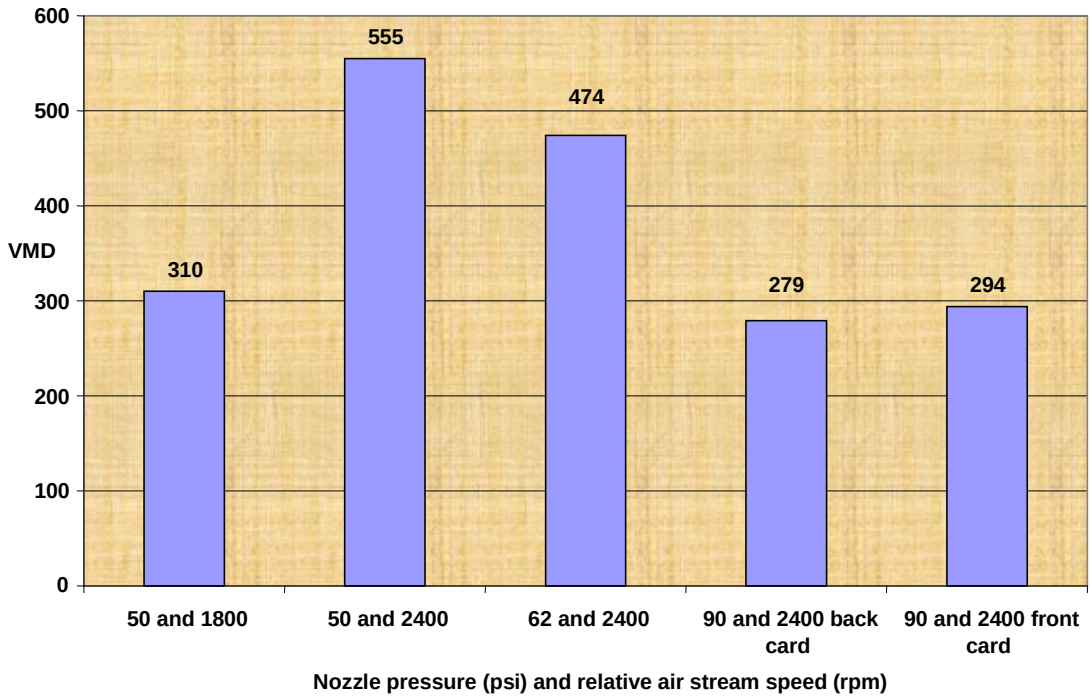

Figure 5. Volume Median Diameter (VMD) drop size measured with water sensitive paper using Hardi FO25_110 nozzles at varying pressures and spray air delivery speeds.

Table 1. Effects of fungicide on absorbance, yield, test weight and protein by nozzle and air stream speed, Esmond 2006.

Sources of Variation	Nozzle or Air Stream Speed	Absorbance	Yield (bu/a)	Test Weight (lb/bu)	Protein (%)
Nozzle		0.0444	0.1438	0.0867	0.1075
Air Stream Speed		0.0771	0.9207	0.6719	0.6693
Noz*Air		0.4275	0.7027	0.6749	0.8804
%C.V.		18	8	1	3
Nozzles averaged across air stream speeds					
	XR11003	.151	30.3	58.0	15.8
	TT11003	.132	28.8	57.4	16.1
$LSD_{(0.05)}$		.02	NS	00.6[Z]	NS
Air stream speeds averaged across nozzles					
	Fast	.163	29.0	57.4	16.1
	Medium	.136	29.5	57.7	16.0
	Slow	.129	29.2	57.7	15.8
	None	.138	29.7	57.9	15.9
$LSD_{(0.05)}$		.03[Z]	NS	NS	NS

[Z]Significant at 0.10 level.

CHARACTERIZING PARAMETERS OF AIR DELIVERY TYPE SPRAY SYSTEMS TO MAXIMIZE FUNGICIDE EFFICACY ON SMALL GRAIN.

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ABSTRACT

Several major manufacturers of ground application equipment (e.g. Hardi Spray Systems and Spray-Air Technologies Inc.), manufacture and sell sprayers that use an air stream to assist in delivering the spray solution to the plant canopy. These sprayers have been shown to offer several unique performance characteristics. First, the air stream minimizes spray drift by overpowering the ambient wind and carrying the smaller spray droplets to the target plant material. Second, the energy of the air stream tends to carry the small droplets (less than 200 microns) deeper into the plant canopy. Third, the turbulence of the air stream assists in more uniformly depositing the spray drops in the hard-to-reach areas of the canopy. The second and third characteristics would be important in controlling foliar diseases. The air stream, depending on velocity, also would be able to alter the orientation of the grain head and change potential deposition. Our objective is to characterize the effects of varying the speed of the air stream, drop sizes and application angles for improved fungicide efficiency to control Fusarium head blight on spring barley and hard red spring wheat (HRSW). The two studies were randomized complete block designs with factorial arrangements and replication. Factors included three drop sizes, three air speeds, and three spray angles. Prosaro fungicide and Induce adjuvant were applied at 6.5 fl. oz/acre and 0.125% v/v to control FHB.

RESULTS

Fungicide coverages were different among sprayer factor combinations on HRSW but not barley indicating the uniqueness of architecture of the individual crop. Fungicide applied with a 'large' fine drop at 60º angle had the lowest incidence and field severity on the HRSW. HRSW yield was greatest when a median air speed was used, 55.8 vs 52.4 and 51.3 bu/acre. On barley a smaller yield was measured when a coarse drop was used in combination with near vertical orientation and minimum air speed. Several sprayer configurations increased plump. The untreated control was included in the trials but was not included in the statistical calculations because it did not fit with the factorial arrangement.

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DISCLAIMER

Any opinions, findings, conclusions, or recommendations expressed in this publication are those of the author(s) and do not necessarily reflect the view of the U.S. Department of Agriculture.

EVALUATION OF FUNGICIDE FOR CONTROL OF FUSARIUM HEAD BLIGHT WITH AERIAL APPLICATION TECHNOLOGY.

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INTRODUCTION

Fusarium head blight (FHB) has been a major problem for cereal grain producers during the past decade. To combat this disease, growers have applied fungicide by both aerial and ground application. About 50% of the small grains acreage sprayed with fungicide in the Dakota-Minnesota region of the Great Plains is applied with spray planes. Aerial application has several advantages over ground application. The planes travel at speeds greater than 100 mph so large acreages can be sprayed in relatively short periods of time, the planes can make applications when surface conditions do not permit use of ground application equipment, and there is less damage to the crop due to tracking. Most aerial applications are applied at 3 to 7 GPA depending on the fungicide label. Applicators use a variety of nozzle types which often include deflectors to discharge spray perpendicular to the air stream. Spray discharge angles perpendicular to the air stream create a smaller drop size than nozzles directed parallel to the air stream. Faster travel speeds will decrease drop size and increasing liquid operating pressure will increase drop size. Spray volumes can be increased by increasing orifice size or by adding additional orifices along the spray boom. Aerial application spray drop size is determined by orifice size, nozzle orientation to the air stream, operating pressure and flying speed.

MATERIALS AND METHODS

An aerial application study was conducted near Esmond, North Dakota in 2006 to evaluate fungicide application for control of FHB on 'Tradition' cultivar barley. A site was selected on the Bill and Louis Arnold farm. The study team included Bill Arnold, farm operator, Dakota Aviation, Don Hutson-owner/pilot Grafton, ND, Vern Hofman and Scott Halley-North Dakota State University, Extension Engineer and Crop Protection Scientist, respectively. Several additional summer staff completed the team. The study was designed as a randomized complete block with four replicates. The plots were 150 ft wide (three application passes) by 450 to 850 ft long. Plots in blocks for replicate one and two were north/south and replicates three and four east/west. The treatments included Folicur 3.6 F (tebuconazole) fungicide (Bayer CropScience manufacturer) at 4 fl oz/acre applied with spray volumes of 3 or 7 GPA applied with a fine and a 'small' medium size drop and one volume of 5 GPA applied with a 'small' medium size drop (the 5 GPA treatment is a typical application standard of commercial aerial applicators). The applications were applied to heading barley (greater than 50% of main stem heads fully extended from the boot). The fungicide was applied with a fixed-wing Cessna Ag Truck aircraft equipped with CP-03 nozzles flying at 125 mph with an operating pressure of 40 psi. The different spray volumes were obtained by changing orifice size across the spray boom and the drop size adjustment was made by using the 30 or 90 degree deflector, large and smaller drop size, respectively. The treatments were applied on 30 June between 10:00 a.m. to 2:00 pm after the dew had dried from the plants. Wind conditions were WNW at speeds of 8.5 to 10.4 mph. This is a typical wind speed for the region at this time of year. The fungicide was applied with Induce adjuvant (Helena Chemical Co.) at 0.125% v/v and F D&C Blue #1 dye added at 44 grams per acre. The dye is a food grade type used in coloring food products. Water sensitive cards were placed on stands at grain head height in the center of each plot to replicate a head. The most commonly used method to evaluate spray technology is the use of water and oil sensitive paper (WSP Spraying Systems Co. ®, Wheaton, Illi-

nois 60189). Cards, 26 x76 mm, were placed at grain head height on stands (Panneton, 2002). One card was placed horizontal (Wolf and Caldwell, 2004). Applied stain size was determined with WRK DropletScan system (WRK, Cabot, Arkansas 72023) and presented as volume median diameter (VMD) which indicates that ½ of the spray volume is in drops smaller than this drop size and ½ of the spray volume is in drops larger than this size. The area of coverage is presented as percent of the card area analyzed.

Three 50 ft spray passes were made side by side (150 ft.) on each plot. All data were collected from the center of the plot. Additionally, three samples of ten heads were collected at 3 points across the center swath and placed in glass Erlenmeyer flasks for determination of head coverage of the spray solution. The collected heads were stored on ice until they could be measured for dye coverage. The spray coverage of the heads was determined by washing the dye from the heads by wrist action shaking for three minutes with 80 ml of 95% ethyl alcohol and determining the absorbance with a Jenway spectrophotometer (model 6300). Differences among treatments were determined by a visual assessment of FHB and foliar disease at mid dough growth stage by assessing twenty heads per plot and determining the incidence of the disease (present or not) and the severity of the individual head. The summation of the incidence times the severity of the twenty heads gave a field severity per plot. Foliar disease differences were determined by estimating the infected area on five leaves at two locations. The field was harvested on 5 August. One pass of the combine was made through the center of each plot with a Caterpillar Lexion combine with a straight cut header. The grain from the harvested area of each plot was measured with a weigh wagon and a sub sample saved to determine yield, test weight, protein, plump and deoxynivalenol (DON) from the processed grain sample. Data were analyzed with the general linear model (GLM) in SAS. Fisher's protected least significant differences (LSD) were used to compare means at the 5% probability level.

RESULTS AND DISCUSSION

The environmental conditions were in contrast in 2006 compared to 2005 when a duplicate trial was conducted. The crop in 2005 was devastated with FHB. In 2006 low relative humidity levels and little precipitation kept both FHB and foliar diseases from causing an economic loss. The limited available soil water also limited yields and reduced test weight and plump to levels so that malting barley standards were not met. No differences were determined among yield, test weight, protein and absorbance. Plump was increased 10% with a 3 GPA spray volume. The benefit was a result of increased amount of fungicide active ingredient collected on the spike th the larger fungicide concentration of the spray solution. Some fungicies extend the growing period of the plant before senescence and it is the authors' perspective that this may have occurred. A trend was established showing greater deposition with the finer type drop size. The awns of the barley are efficient collectors of fine drops and also spores. This trend is different from a typical application with ground equipment where a drop size of 300 to 350 microns will deposit in greater quantities than a fine drop size. The ASABE standard S-572 spray drop classification system for the two applications should have been about 240 and 300 microns. The WSP card showed a larger stain size than the reported limits of the technology. The differences show a drop size difference of about 50 microns between the two stain sizes and show one of the limitations of using WSP and field spray applications. The untreated in each replicate was not included in the statistical analysis because of the lack of fit with the factorial arrangement used to compare the mean volumes and drop sizes but is presented as a reference to overall fungicide efficacy.

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DISCLAIMER

Any opinions, findings, conclusions, or recommendations expressed in this publication are those of the author(s) and do not necessarily reflect the view of the U.S. Department of Agriculture.

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Table 1. Yield, Test Weight, Plump, Protein, and Head Coverage (Absorbance) by Spray Volume and Drop Size Esmond 2006.

Spray Volume	Drop Size	Yield (bu/ac)	Test Weight (lb/bu)	Plump (%)	Protein (%)	Absorbance
untreated		66.3	45.1	48.9	11.8	0.057
		Spray volume averaged across drop sizes				
3		62.0	45.8	58.3	11.8	0.255
7		64.8	45.4	52.8	12.7	0.263
LSD (0.05)		NS	NS	5.3	NS	NS
		Drop size averaged across spray volumes				
	Fine	62.3	45.6	55.3	11.9	0.289
	Medium	66.6	45.6	55.9	11.9	0.229
3	Fine	58.5	46.0	58.6	11.8	0.286
	Medium	65.4	45.5	58.0	11.9	0.225
7	Fine	61.9	45.2	51.9	12.1	0.292
	Medium	67.7	45.6	53.8	12.0	0.235
Sources of variation						
Rep		0.0474	<0.0001	0.0019	0.1765	0.0431
Volume		0.8153	0.1592	0.0430	0.3871	0.8911
Drop Size		0.2256	0.8025	0.7976	0.9459	0.3390
Vol*Drop		0.6634	0.1146	0.6099	0.5453	0.9735
%C.V.		10	1	9	3	45

Table 2. Volume Median Diameter (VMD), GPA, and Coverage determined Spray Solution Collected on Horizontal Placed Water Sensitive Cards, Esmond 2006.

Spray Volume	Drop Size	VMD	GPA	Coverage (%)
3	Fine	360	4.2	10.1
3	Medium	404	2.0	4.7
5	Medium	370	4.9	12.0
7	Fine	401	7.5	17.8
7	Medium	451	4.7	11.9
Untreated		200	0.07	0.2

RELATIONSHIPS BETWEEN YIELD, GRAIN QUALITY VARIABLES, AND FUSARIUM HEAD BLIGHT INTENSITY IN WINTER WHEAT.

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ABSTRACT

Fusarium head blight (FHB) of wheat, caused by *Fusarium graminearum*, can cause significant losses resulting from yield reduction, kernel damage, and presence of deoxynivalenol (DON), an important mycotoxin with serious food safety implications. In 2007, two experiments were conducted to identify relationships between (i) yield, grain quality variables, and FHB intensity and (ii) visual assessments of FHB and DON. In the first experiment, three winter wheat varieties (Jagalene, Harry and 2137) were planted on two planting dates, 5 and 27 October 2006. Plots were inoculated with conidia and ascospores of *F. graminearum* (1×10^5 spores/ml) at early and mid anthesis, or were not inoculated. Experimental design was a split-split-plot in randomized complete blocks with three replications. Planting date was the main plot, variety the subplot, and inoculation timing the sub-subplot. FHB severity was determined 21 and 25 days after inoculation on 20 heads in each of five arbitrarily selected locations in each plot. There was a significant positive correlation ($0.48 \leq r \leq 0.76$, $P \leq 0.05$) between FHB incidence and FHB severity in each variety (N = 18), first planting date (N = 27), and all varieties and planting dates combined (N = 54). Correlation between FHB index and yield was negative but not significant at $P = 0.05$. Correlation between FHB index and 1000 kernel weight was significant for the first planting date ($r = 0.45$, $P = 0.0191$) but positive, contrary to what as was expected. Correlation between FHB index and *Fusarium* damaged kernels (FDK) was significant for the first planting date ($r = -0.47$, $P = 0.0132$) but negative, contrary to what was expected. Correlation between FHB index and DON was not significant at $P = 0.05$. Correlation between FDK and 1000 kernel weight was not significant at $P = 0.05$ for Jagalene and Harry, but was significant for 2137 ($r = -0.47$, $P = 0.0507$), first planting date ($r = -0.65$, $P = 0.0002$), second planting date ($r = -0.63$, $P = 0.0005$), and all varieties and planting dates combined ($r = -0.64$, $P < 0.0001$). Correlation between FDK and DON was not significant at $P = 0.05$, as was correlation between FDK and yield. In the second experiment, two varieties (Harry and 2137) were planted on 9 October 2006. Plots were inoculated at early anthesis as described above. Varieties were arranged in randomized complete blocks with three replications. In mid June 2007, 20 heads were randomly tagged in each of 11 disease severity categories in each plot: 0, 5, 10, 15, 20, 25, 30, 35, 40, 45, and 50%. There was a significant positive correlation between FHB severity in the 11 severity categories and DON for both Harry ($r = 0.74$. $P = 0.0092$) and 2137 ($r = 0.70$, $P = 0.0157$). However, DON levels were higher in Harry than in 2137. The results from this study indicate that (i) relationships between yield, grain quality variables, and FHB intensity may not be clear cut, (ii) there is a positive association between DON levels and FHB severity, and (iii) wheat varieties differ in the levels of DON they accumulate.

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This material is based upon work supported by the U.S. Department of Agriculture, under Agreement No. 59-0790-7-080. This is a cooperative project with the U.S. Wheat & Barley Scab Initiative. Any opinions, findings, conclusions, or recommendations expressed in this publication are those of the author(s) and do not necessarily reflect the view of the U.S. Department of Agriculture.

OUTCOMES OF USING INTEGRATED FHB MANAGEMENT STRATEGIES ON MALTING BARLEY CULTIVARS AND GERMPLASM IN MINNESOTA.

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ABSTRACT

The objective of our trial was to determine grain and malt quality responses from four commercially-available malting cultivars and four advanced malting germplasm lines following exposure to four Fusarium head blight (FHB) disease management strategies. The experiment was planted on 9 May 2007 into soybean residue and was situated in a commercial production field within Marshall County, in northwest Minnesota. Barley entries included Tradition, a 2004 Busch Agricultural Resources, Inc. (BARI) release; Legacy, a 2000 BARI release; Drummond, a 2000 North Dakota State University (NDSU) release; B2218 and B2513, BARI advanced germplasm lines; ND20448, NDSU advanced germplasm line; and M122, University of Minnesota advanced germplasm line. Treatments were replicated four times and exposed to one of four fungicide strategies (Table 1). The test area was neither misted nor inoculated.

Environmental conditions did not support normal plant growth or stand establishment. Frequent rain events caused soil saturation for an extended period of time prior to Feekes growth stage (FGS) 10.5 (early heading). This resulted in plant stress which caused severe plant stunting, tiller abortion, and low yield (entry means for yield ranged from 21 bu/a to 37 bu/a). Split-plot analyses from PROC GLM in SAS were conducted where fungicide treatment represents whole plots and entry represents subplots. FHB symptoms were assessed at approximately FGS 14. Barley entry and fungicide treatment were significant for FHB incidence, while barley entry was also significant for FHB severity and FHB index ($P<0.05$). While FHB symptom expression was relatively low (entry means for FHB index ranged from 0.2% to 1.8%), resistant germplasm lines (B2218, M122, and B2513) had significantly lower index values than current commercial cultivars (Legacy, Tradition, and Robust). Deoxynivalenol (DON) levels in grain were miniscule to below detectable limits with an overall test mean of 0.1 ppm. The nontreated control fungicide treatment was not different from tebuconazole (4 fl. oz./a), but had significantly higher DON levels compared to either rate of Prosaro ($P<0.05$).

Three replicates of grain samples from treatments #1 and #4 (Table 1) were micro-malted in the BARI Seed Research Quality Lab located at Fort Collins, CO. Resulting malt was analyzed for alpha amylase, beta glucan, diastatic power, free amine nitrogen, percent fine extract, predicted extract, malt protein, wort protein, and turbidity. Differences between barley entries were significant across all quality traits ($P<0.05$). Responses of malt to fungicide were significant for alpha amylase ($P=0.03$). Treatment #4 resulted in larger levels (75.0) of alpha amylase than the nontreated control (69.7). There were no significant fungicide*entry interactions at $P<0.05$.

Data produced from a single growing season in northwest Minnesota indicate that interactions between fungicide treatment and barley entries generally did not influence FHB disease symptoms, grain yield, or kernel and malt quality traits. However, additional data is needed from a typical growing season before further conclusions can be drawn.

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The authors would like to thank Busch Agricultural Resources, Inc. for supporting this research; Bayer CropScience for supplying fungicide products; UM and NDSU breeders for providing germplasm; Busch Agricultural Resources, Inc. Seed Research Quality Lab and the University of Minnesota Mycotoxin Lab for providing malt quality analyses and DON results, respectively.

Table 1. Fusarium head blight disease management strategies tested on eight malting barley entries near Warren in the northwest Minnesota Red River Valley. Fungicide applications were made at Feekes growth stage 10.5 (early heading).

Trt	Product	Active ingredient	Rate* (fl. oz./a)
1	Nontreated control...		
2	Folicur.....	tebuconazole	4.0
3	Prosaro.....	tebuconazole and prothioconazole	6.5
4	Prosaro.....	tebuconazole and prothioconazole	8.2

*Treatments 2 through 4 included 0.125% Induce, a nonionic surfactant.

UNDERSTANDING PRACTICAL OUTCOMES FROM IMPLEMENTING FHB MANAGEMENT STRATEGIES ON SPRING WHEAT.

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ABSTRACT

The objective of our trial was to determine grain yield and quality responses, as well as economic outcomes from 13 hard red spring wheat cultivars when exposed to six different disease management strategies. This research represents Minnesota's participation in the multi-state, multi-year integrated disease management cooperative research which is meant to identify the most practical means in managing Fusarium head blight (FHB) across states and wheat classes.

The test included four replicates at each of two experiment locations. Planted into soybean residue, a site was located near Oklee in northwest Minnesota and another was near Fergus Falls in west central Minnesota. The Oklee site was planted on 27 April 2007 and the Fergus Falls site on 2 May 2007. Spring wheat cultivars included Ada, Alsen, Banton, Bigg Red, Briggs, Freyr, Glenn, Knudson, Oklee, Samson, Steele-ND, Ulen, and Walworth which were exposed to one of six disease management strategies (Table 1). The test areas were neither misted nor inoculated.

Environmental conditions varied substantially between locations. Split spilt-plot analyses using PROC GLM in SAS were made where 'location' represented the whole plot factor, 'fungicide' the subplot factor, and 'cultivar' the sub-subplot factor. Transformations were conducted on data identified with non-normal distributions. Fergus Falls had lower test weights, kernel protein, and FHB index ratings than the Oklee site ($P<0.05$). Deoxynivalenol (DON) levels in grain were miniscule to below detectable limits at Fergus Falls (d"0.13 ppm). Oklee location DON results are not yet available. Cultivar and disease management strategy were both significant for net revenue, yield, test weight, protein, and FHB incidence, while FHB severity and FHB index were significant for cultivar ($P<0.05$). Knudson (77.8 bu/a), Samson (77.6 bu/a), and Steele-ND (74.9 bu/a) had the largest yields, while Bigg Red (62.4 bu/a) and Alsen (63.3 bu/a) had the smallest. Cultivars Samson, Ulen, Steele-ND, and Oklee had the highest ratings for FHB incidence, FHB severity, and FHB index while Bigg Red, Alsen, Glenn, and Knudson had the lowest. Knudson ($611.27/a), Samson ($608.26/a), and Steele-ND ($591.48/a) had the greatest net return while Bigg Red ($482.89/a) and Alsen ($497.13/a) returned the least ($P<0.05$). Across all cultivars, yield, protein, and test weight were significantly increased with disease management strategies #3, #4, and #5, compared with strategy #1 (Table 1). Strategy #4 resulted in the largest net return and strategies #1, #2, #5, #6 the least returns ($P<0.05$).

Disease-associated limitations to yield were offset by timely fungicide application. Cultivars known for susceptibility to disease responded well to the growing environment, producing excellent yields of high quality grain. Fungicide application increased net returns compared with no fungicide even during a year of relatively low disease pressure. Economically-speaking, spring wheat growers in the Minnesota Red River Valley who benefited the most during 2007 grew cultivars that were moderately susceptible to FHB.

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Table 1. Disease management strategies tested on 13 cultivars of hard red spring wheat at two locations in the Minnesota Red River Valley.

			Application	
Strategy	Product	Active ingredient	Rate*	Timing**
1	Nontreated control..			
2	Dividend Extreme..	difenoconazole and mefenoxam	3 fl. oz./100 lbs.	Seed applied preplant
3	Headline.....	pyraclostrobin	3 fl. oz./a	FGS 2
	Folicur/Proline.....	tebuconazole & prothioconazole	3 + 3 fl. oz./a	FGS 10.51
4	Dividend Extreme..	difenoconazole & mefenoxam	3 fl. oz./100 lbs.	Seed applied
	Headline.....	pyraclostrobin	3 fl. oz./a	FGS 2
	Folicur/Proline.....	tebuconazole/prothioconazole	3 + 3 fl. oz./a	FGS 10.51
5	Dividend Extreme..	difenoconazole & mefenoxam	3 fl. oz./100 lbs.	Seed applied
	Folicur/Proline.....	tebuconazole & prothioconazole	3 + 3 fl. oz./a	FGS 10.51
6	Folicur/Proline.....	tebuconazole & prothioconazole	3 + 3 fl. oz./a	FGS 10.51

*Treatments 3 through 6 included 0.125% Induce, a nonionic surfactant.

** Feekes growth stage (FGS) 2 = 4 to 5 leaf, and FGS 10.51 = early anthesis.

CONTRIBUTION OF WITHIN-FIELD INOCULUM SOURCES TO FUSARIUM HEAD BLIGHT IN WHEAT.

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ABSTRACT

Knowledge of the relative contribution of within-field inoculum sources of *Gibberella zeae* to infection of local wheat and barley is important for developing and/or excluding strategies for managing Fusarium head blight (FHB). Our research is based on the hypothesis that spores of *G. zeae* that are deposited on wheat spikes and that result in Fusarium head blight come primarily from well-mixed, atmospheric populations in an area. Our experimental objective was to determine the relative contribution of within-field, clonal inoculum sources of *G. zeae* to FHB in susceptible wheat cultivars. In 2007, corn stalks and corn kernels infested with clonal, fingerprinted isolates of *G. zeae* containing rare alleles (relative to background populations) were released in replicated 1 m diameter circular plots in single wheat fields in New York and Virginia. We collected mature wheat spikes at the inoculum source, at a radius of 10 feet from the source, at a radius of 20 feet from the source, and in more distant parts of each field. We used amplified fragment length polymorphisms (AFLPs) to genotype isolates recovered from these spikes and to determine the contribution of released isolates to FHB at various distances from those sources. Since our inoculum sources contained clonal isolates that have unique AFLP haplotypes, we were able to observe these clones in a mixed/diverse background population containing numerous AFLP haplotypes. Nearly 500 isolates of *G. zeae* were collected and single-spored from NY and VA. Preliminary AFLP data from the first year of experimentation suggests that within-field sources of *G. zeae* provided a minor fraction of FHB inoculum compared to background atmospheric sources in a non-epidemic environment in New York and in a moderate epidemic environment in Virginia.

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TIME OF FLOWERING IN WHEAT FOR MANAGING FUSARIUM HEAD BLIGHT.

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ABSTRACT

Efficacy of management is increasingly being timed based on crop developmental stage and consideration of crop developmental physiology. In the case of Fusarium head blight, it has often been managed by one pesticide application timed to the developmental stage of anthesis (i.e., flowering). However, flowering is controlled by the interaction of genotype, environment, and management and can occur over an extended period of time, confounding when to make the application. The objective of this talk is to discuss wheat development to provide information for improving the current management practice and exploring alternative management options. The presentation will first present a brief overview of wheat development and highlight when the various yield components are being formed. Wheat development follows a few general principles beginning with development being an orderly and predictable process. The genetics provides the "blueprint" for the orderly sequence of events leading to flowering. Temperature, reflecting thermal time, is then used to predict when flowering will occur. Sources of variation in flowering time are identified including a) within a shoot, b) among shoots on a plant, c) among plants within small areas/plots, and d) across landscapes. Other sources of variation exist among genotypes and variable planting/emergence dates. Management options to reduce the period of flowering are discussed, along with the risks of doing so

DIFFERENTIAL EFFECTS OF INFECTION TIMING ON FUSARIUM HEAD BLIGHT AND ON DON AND DON DERIVATIVES IN THREE SPRING GRAINS.

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ABSTRACT

The effect of infection timing on the development of Fusarium head blight (FHB) and DON and DON derivatives was evaluated under a controlled greenhouse environment. Test plants were susceptible and resistant or moderately resistant lines of three spring grains - hard red spring wheat (HRSW) ('Grandin' and 'Glenn'), durum wheat ('Munich' and 'Divide'), and six-rowed spring barley ('Robust' and ND20448 [a line from R. Horsley's breeding program]). Infection timings included head half-emerged (Feekes 10.3), full head emergence (Feekes 10.5), anthesis in wheat (Feekes 10.51), and kernel watery ripe (Feekes 10.54), or dual infections at the two later growth stages. Infections were initiated by atomizing a mixture of four isolates of *Fusarium graminearum*, at 20,000 spores/ml, 20 ml/pot, with a DeVilbiss atomizer, followed by 24 hours of misting. FHB incidence and severity were determined at 21-25 days after inoculation. At maturity, kernels were hand threshed for subsequent mycotoxin analysis. DON, 3ADON, 15ADON and nivalenol (NIV) analyses were done using gas chromatography and electron capture techniques. FHB indices [(incidence x severity)/100] and mycotoxin levels (ppm) indicated that differential responses to single infection timings occurred among spring grain classes: a) in barley, values of these parameters were highest with infection at the watery ripe stage; b) in HRSW, at anthesis; and 3) in durum, about equally high at anthesis or watery ripe stage infections. In all three crops, infections at head half-emerged resulted in the lowest FHB severities and DON levels. The dual infections at the two latter growth stages, generally resulted in the highest FHB index and DON values in all grain classes. DON, 15ADON and 3ADON accumulations were highly correlated with FHB index in all spring grain classes. 15ADON and 3ADON levels also were highly correlated with DON levels. 15ADON was more frequently recovered and at higher ppm than 3ADON (highest average 15ADON was 4.5 ppm in barley, vs 1.1 ppm 3ADON in barley). 3ADON generally was detected only when the average DON levels were high: 22 ppm in barley, 45 ppm in HRSW and 37 ppm in durum, with average 3ADON levels well under 1.0 ppm. Resistant lines generally had much lower DON levels than susceptible cultivars, across all infection timings. 3ADON was not detected in the resistant HRSW or the moderately resistant durum cultivars. NIV was not detected in any of the grain classes.

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EFFECTS OF FUNGICIDE TIMING ON FUSARIUM HEAD BLIGHT AND ON DON AND DON DERIVATIVES IN THREE SPRING GRAINS.

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ABSTRACT

The effects of fungicide timing on the reduction of Fusarium head blight (FHB) and mycotoxins were evaluated under a controlled greenhouse environment. Fungicides were tested on susceptible and moderately resistant to resistant lines of three spring grains - hard red spring wheat (HRSW), durum wheat, and six-rowed spring barley. Fungicide timings included head half-emerged (Feekes 10.3), full head emergence (Feekes 10.5), anthesis in wheat (Feekes 10.51), and kernel watery ripe (Feekes 10.54) or a dual treatment at full head emergence in barley or anthesis in wheat, followed by treatment at kernel watery ripe. Fungicide was applied with a greenhouse track sprayer with XR8001 forward and backward flat fan nozzles, 18 gpa, at appropriate growth stages. Treatments were either Prosaro (tebuconazole + prothioconazole) at 6.5 fl oz/A, or Proline (prothioconazole) at 5 fl oz/A. Fungicide treatments were applied 4 hours after infection initiations. Infections were initiated by atomizing a mixture of four isolates of *Fusarium graminearum*, at 20,000 spores/ml, 20 ml/pot, with a DeVilbiss atomizer, followed by 24 hours of misting. FHB incidence and severity were determined at 21-25 days after treatment. At plant maturity, kernels were hand threshed for subsequent mycotoxin analysis. DON, 3ADON, 15ADON and nivalenol (NIV) analyses were done using gas chromatography and electron capture techniques. FHB indices [(incidence x severity)/100] and DON values (ppm) were significantly reduced by fungicide treatments in all grain classes and cultivars. In the most susceptible lines of all 3 grain classes, a single fungicide treatment, applied at optimal growth stage for infection, resulted in 90-98% reductions of FHB indices and DON levels (example: 24.4 ppm DON in durum wheat infected at anthesis, vs 0.56 ppm DON with fungicide treatment added at anthesis). With the dual timings of application, FHB indices and DON levels were reduced by 86 to 97%. Similar percent reductions were observed in the more resistant lines, but overall FHB and DON levels were lower in the more resistant lines. Fungicide treatment at any of the tested timings in HRSW and durum resulted in zero detection of 15ADON and 3ADON. In barley, fungicide treatment resulted in 100% reduction of 3ADON, and 91-94.4% reduction of 15A DON. These fungicide treatments were very effective in reducing FHB, DON and DON derivatives under greenhouse conditions.

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EXPERIENCES IN REDUCING DISEASE AND DON THROUGH COMPONENTS OF FHB MANAGEMENT.

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ABSTRACT

Few would argue that a favorable climate during vulnerable crop growth stages often is the key factor resulting in severe Fusarium head blight (FHB). However, as unfavorable weather is hard to avoid, researchers and producers have looked for implementable strategies for managing FHB. Champeil et al., 2004 (*Fusarium head blight: epidemiological origin of the effects of cultural practices on head blight attacks and the production of mycotoxins by Fusarium in wheat grains. Plant Science 166:1389-1415)* provided an extensive review of studies of cultural practices that may affect FHB severity and mycotoxin production, and more recent papers also have been published. Key strategies that have been extensively researched include: crop rotation, tolerant cultivars, and fungicide use.

From 2003-2005, various regions in the US had severe FHB outbreaks, and individual FHB management strategies used alone did not necessarily reduce disease severity and DON to levels required by the grain industry. Several 2005 research trials in eastern ND provided quantitative evidence that a combination of crop rotation, variety choice, and fungicide treatment reduced FHB severity and DON levels in an additive manner, ie 10 ppm DON levels in spring wheat with no strategy; 5 ppm DON with soybean rotation added; 2.0 ppm with soybean + resistant variety; and 1.2 ppm with soybean + resistant variety + fungicide.

Members of the management group of the US Wheat and Barley Scab Initiative (USWBSI) met in 2006 and decided to implement studies, across multiple states and grain classes, to quantify the value of additive strategies for FHB and DON management. These cropping systems studies were to be done under natural field conditions and the objectives were to:

1) demonstrate that integrated management is the most effective means of reducing losses to FHB/DON; and

2) increase grower adoption of integrated strategies by demonstration of their effectiveness in a wide range of environments.

Funded USWBSI cropping system studies were in place in 2007, a year in which some locations again had FHB. The Forum's presentation for this abstract will provide examples of 2007 results from these studies. Dr. Pierce Paul has statistically analyzed results from these cropping system studies, and his poster will be presented at the 2007 FHB Forum. Others also may be presenting their individual state's data. ND results will be published in the 2007 *Proceedings of the 5th Canadian Fusarium Head Blight Workshop, Winnipeg. Nov. 27-30*.

ACKNOWLEDGEMENT AND DISCLAIMER

This material is based upon work supported by the U.S. Department of Agriculture, under Agreement No. 59-0790-4-114. This is a cooperative project with the U.S. Wheat & Barley Scab Initiative. Any opinions, findings, conclusions, or recommendations expressed in this publication are those of the author(s) and do not necessarily reflect the view of the U.S. Department of Agriculture.

COMPARISON OF FUNGICIDES AND NOZZLE TYPES AGAINST FHB IN WHEAT AT FARM APPLICATION.

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ABSTRACT

In 2006 the Turbo FloodJet and the TeeJet XR nozzles were compared in three winter wheat cultivars Petur (MR), Kapos (MS) and Miska (S). In 2007 the AIC TeeJet and Turbo TeeJet Duo nozzles were added to have a wider spectrum of comparison. All fungicides were run at 250 L/ha water and 8 km/hr speed. 17 m wide boom was used, on both sides mounted with a different nozzle type. A plot was 7 m wide and 300 m long. Technologies were evaluated across cultivars and fungicides, the fungicides were rated across cultivars and technologies. In both years natural epidemic occurred. In cv Miska, the most susceptible genotype, about 30 infected heads/m^2 were recorded.

In 2006 eight, in 2008 10 different fungicides were used:

Prospect (200 g/L carbendazim, 80 g/L propiconazole) 1.5 L/ha;
Falcon 460 EC (167 g/L tebuconazole 250 g/L spiroxamine 43 g/L triadimenol) 0.8 L/ha;
Prosaro (125 g/L prothioconazole, 125 g/L tebuconazole) 1.0 L/ha;
Tango Star (84 g/L epoxyconazole, 250 g/L fenpropimorph) 1.0 L/ha;
Eminent 125 SL (125 g/L tetraconazole) 1.0 L/ha;
Amistar Xtra (200 g/L azoxystrobin, 80 g/L ciproconazole) 1.0 L/ha;
Coronet (Nativo) (200 g/L tebuconazole és 100 g trifloxystrobin) 1.0 L/ha;
Artea 330 EC (250 g/L propiconazole, 80 g/L ciproconazole) 1.0 L/ha; and
Juwel (125 g/L epoxyconazole, 125 g/L krezoxim-metil) 1.0 L/ha.

In 2006 the mean efficacy of the TeeJet XR nozzle across fungicides was 44 %. The TurboFloodJet nozzles gave 58.60 % reduction of the natural head infection. At the Turbo FloodJet nozzle the lowest efficacy was measured for Eminent (16 %) and 91.5 % for Prosaro. At the traditional nozzles 14.42 and 79 % were the corresponding values. For DON the mean efficacy for TeeJet XR nozzles was 51 %, the reduction for Turbo FloodJet 65 %. In 2007 four nozzles were compared, the two nozzles types from 2006 were supplemented by AIC TeeJet and the Turbo TeeJet Duo nozzles as the Turbo FloodJet nozzles need very uniform soil level to keep the boom constantly at 20-30 cm above stand. This is not always at hand. The data for the FHB data showed a 60.7 % efficacy for AIC TeeJet and 62.8 % for TeeJet XR across fungicides. The Turbo TeeJet Duo reached 70.2 % and the Turbo FloodJet finished at 79.2 %. Prosaro was again the best with 95 % efficacy at the Turbo FloodJet nozzle type. The results provide several important conclusions. The traditional nozzles can reach with the best fungicides up to 70 % reduction when technology, timing is optimal. The difference is very large between fungicides, in this test 42 % for Eminent and 92 for Prosaro across technologies. We believe therefore that the successful chemical control needs both better technology and better fungicides.

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FIELD AND LABORATORY STUDIES TO MONITOR CELL POPULATIONS, LIPOPEPTIDES AND LIPOPEPTIDE GENES OF *BACILLUS* 1BA, A BIOCONTROL AGENT. ACTIVE AGAINST FUSARIUM HEAD BLIGHT.

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ABSTRACT

Fusarium graminearum causes Fusarium Head Blight (FHB) on wheat, barley and other small grains. Biocontrol agents (BCAs), such as certain *Bacillus* sp., can be used to control FHB and/or reduce deoxynivalenol (DON) levels in grain. Population studies of the BCAs on inoculated grain heads show how BCA numbers fluctuate over time, and if there are differences in BCA populations between wheat and barley. In previous work to count populations of *Bacillus* 1BA in the field, we found that this bacterium is thermotolerant and salt tolerant, able to grow on Tryptic Soy Agar (TSA) at elevated salt and temperature conditions that inhibit most micrflora native to the grain heads. Field plot treatments in 2007 were 1BA by itself; and 1BA + Prosaro (a fungicide) + Induce NIS (nonionic surfactant). Population counts were done over a 20 day period on both the wheat and barley. Endospore formation was examined by pasteurizing the samples at 85°C for 10 minutes, then plating. 1BA was isolated using TSA containing 10% NaCl, with incubation at 50°C for 48 hours. Controls for wheat and barley that did not receive 1BA inoculation gave low cell counts throughout the experiment, no higher than about 3.5 X 10^2 CFU/g fresh plant weight. For wheat, in the treatment with 1BA alone, vegetative cell counts peaked around day 10, then declined sharply, followed by a second increase in numbers by day 20, with a concurrent increase in endospore numbers. Compared to the treatment with 1BA alone, in the wheat treatment combining 1BA with Prosaro and Induce NIS, the population peaks shifted in time. Peak numbers of vegetative cells occurred at about day 6 then declined, with a smaller second peak on day 17. As vegetative numbers of 1BA in this treatment declined, endospore numbers increased. Numbers of 1BA on inoculated barley were much lower than on wheat, not being much different from the uninoculated control and not fluctuating much over time. It appears that 1BA, originally isolated from wheat material, is much better able to colonize and grow on grain heads of wheat than barley. The biocontrol effect of 1BA may be due to its production of lipopeptides including surfactin. Attempts to directly detect presence of *Bacillus* lipopeptides in extracts from inoculated grain heads were not successful, but efforts to detect these molecules directly on inoculated plant material will continue. Direct or indirect detection of surfactin or other lipopeptide genes/products on inoculated grain heads will be checked by methods including PCR. As a step leading to this, DNA of 1BA was isolated, then PCR was performed using primers for the surfactin production genetic locus (sfp). Good yield of PCR product was observed on an agarose gel, verifying that the primers and PCR method may be useful in detecting lipopeptide genes on inoculated grain heads.

EFFECTS OF SOLAR RADIATION ON THE VIABILITY OF *GIBBERELLA ZEAE* ASCOSPORES.

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ABSTRACT

Ascospores of *Gibberella zeae* are considered to be an epidemiologically important type of inoculum for Fusarium head blight of wheat and barley. The objectives of this study were to evaluate the effects of solar radiation, temperature, and relative humidity on the survival of ascospores in the environment at Rock Springs, PA and Manhattan, KS. In each experiment, ascospores of *G. zeae* were collected by inverting cultures containing mature perithecia over glass cover slips for several hours. After deposition, the ascospores were placed on glass Petri dishes and exposed to solar radiation or shaded conditions for predetermined lengths of time. The temperature of the exposed and shaded ascospores was kept constant by allowing the dishes to contact a circulating source of water. Total solar radiation, UV radiation, air temperature, relative humidity, and temperature of the water in the spore exposure apparatus were recorded during each experiment. Following exposure, the spores were washed from the cover glass, placed on water agar and incubated for 8-10 h. The percentage of germinating spores in five sub-samples of 100 ascospores was recorded for each exposure period, and germination was expressed as a ratio of the initial germination of ascospores for that experimental run. The preliminary results of these experiments indicate that the mean initial germination rate of the ascospores produced by isolates considered in this study was 55.6% with a standard deviation of nearly 20%. Regression analysis suggests that total solar radiation and the dose of UV radiation significantly impacted spore survival, but that temperature and relative humidity may also be important variables. The dose of solar radiation resulting in 100% ascospore mortality was 19.8 MJ/m^2 at the KS location, but was significantly greater at the PA location. Differences between the locations may be explained in part by differences in the isolate considered and range of temperatures experienced during the exposure periods.

MECHANISTIC SIMULATION MODELS FOR FUSARIUM HEAD BLIGHT AND DEOXYNIVALENOL.

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ABSTRACT

An empirical model with a single mathematical equation is commonly used as a part of disease forecasting systems. A web-based Fusarium Head Blight (FHB) forecasting tool (http://www.wheatscab.psu.edu/) is one of example of this approach to disease forecasting. These models are relatively easy to implement, and provide reasonably accurate forecasts in many cases. However, it may be difficult to derive biological interpretation from these empirical models. Other modeling approaches, such as mechanistic modeling, can be considered as an alternative. In this study, an object-oriented language STELLA (isee systems, Lebanon, NH) was utilized to create a mechanistic simulation models for FHB and deoxynivalenol (DON) based on the results of past studies on disease development and pathogen biology. Several candidate models have been developed with different scopes of interests, and one of candidate models will be discussed this presentation. This model estimates a distribution of *Fusarium* damaged kernels among heads of wheat and DON accumulation of the grain based on environmental conditions. Major steps in the disease cycle, such as perithecia development by *Gibberella zeae* and infection events were expressed as differential equations that use environmental conditions as input variables. These equations were connected in a logical manner, and using past weather data, a theoretical disease cycle of FHB was simulated over time. The model development process, preliminary results, as well as potential usage of this modeling approach as a tool for hypothesis testing for future studies will be discussed.

SPORE LOAD, DISEASE, AND DON: A FOUR YEAR VARIETY BY RESIDUE STUDY FOR FHB MANAGEMENT.

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OBJECTIVES

A four year field study was established near Brookings, SD in the years 2003 through 2006 to examine the impact of maize residues, host susceptibility and timing of planting on disease and mycotoxin accumulation in hard red spring wheat due to Fusarium head blight. These factors were also examined to determine their impact on inoculum concentrations on spikes from emergence through mid-milk stage. Finally, inoculum concentration was compared with DON in grain at harvest to determine if any consistent relationships existed.

INTRODUCTION

Fusarium head blight, caused primarily by *Gibberella zeae* (Schwein.) Petch (anamorph: *Fusarium graminearum* Schwabe) in the Northern Great Plains (including South Dakota), is usually the greatest threat to wheat quality and production in the humid areas of the region. The fungus survives and over-winters in plant tissue residues including small grain stems and roots as well as maize stalks and ear pieces (Sutton 1982). Recent research suggests that for South Dakota, ascospores and conidia of *G. zeae* are somewhat ubiquitous in the air, thought to be a result of extensive acreage of spore-bearing residues in the region coupled with extensive air mixing and medium to long-distance spore movement (Osborne 2006). Management of FHB in South Dakota involves the use of alternative rotations (wheat not to follow maize), fungicide application at flowering, and the use of resistant varieties to reduce risk. Over the past five years, several moderately resistant varieties have been released which are adapted to South Dakota growing conditions. In 2003, however, only one variety, 'Alsen', was both a recommended variety for SD and classified as 'moderately resistant' to FHB (Hall et al., 2003). This variety, along with 'Norm', a HRSW with high susceptibility to FHB, were used in this study to investigate the interaction of host resistance and 'local' maize residues in a region with abundant airborne inoculum.

METHODS

Field plots were established in Brookings, SD in 2003 through 2006. Each year, two planting dates (PD1 and PD2), timed 10-13 days apart, were utilized creating two identical studies upon which all treatments were applied and all measurements were collected. Planting date 1 represented the typical planting time for area wheat producers, whereas PD2 represented late-planted fields. In general, plots consisted of residue treatment (0, 30, and 80% soil coverage, by line-transect method) to generate corresponding low, medium and high levels of 'local' inoculum (local inoculum being defined as that produced from within the plot area, in contrast to inoculum produced outside the plot area). The medium level was discontinued after 2004. Sub-plots consisted of spring wheat varieties 'Alsen' and 'Norm'. Plot size varied slightly from year to year due to space restrictions; however, final plot disease measurements were collected from areas no smaller that 3.1m by 4.6m. In each year, whole-plots (residue treatments) were buffered on all sides by 8m of a tall wheat variety to mitigate inter-plot interference. The study was dependant on inoculum formed locally (i.e. within or beneath the crop canopy in plant tissues or residues), or externally (i.e. from adjacent fields with corn or small grain residue) and received no additional inoculum in the form of spore suspension or colonized grain (for ascospore spawn). No environmental modification was implemented to alter the conditions for disease development.

Within all sub-plots, a designated area was sampled daily after spike emergence by collecting five spikes per sub-plot for enumeration of spike-borne inoculum as described by Francl, et al. (1999). At three weeks post-flowering, disease assessments were performed on all sub-plots as described by Stack and McMullen, (1995) and included incidence and severity estimates on 100 spikes. Incidence is defined as proportion of 100 rated spikes exhibiting disease symptoms. Severity is the mean severity per infected spike. Disease index, often called 'field severity', is the product of incidence and severity and represents the overall 'amount' of disease in a given area. Harvest data collected included plot yield, test weight and moisture content. Harvested grain was sent to the NDSU Veterinary Toxicology Lab for assessment of mycotoxin concentration in grain following Tacke and Caspers (1996). A Burkard volumetric spore collector was placed for daily monitoring of airborne inoculum in the study area. Weather data was collected using a Campbell Scientific CR10X data logger and peripheral sensors. Parameters measured included temperatures and relative humidity in and above the crop canopy, wind, solar radiation, precipitation, soil temperature, soil wetness and leaf wetness estimations.

RESULTS AND DISCUSSION

The growing seasons 2003 through 2006 in eastern South Dakota were each distinct in terms of FHB levels on spring wheat across East-Central and Northeastern South Dakota. These differences were mirrored in the overall disease levels observed within this study each year, summarized in Table 1. Years within this study represented four distinct categories: very low disease (2006), low disease (2003), moderate disease (2005), and high disease (2004) seasons. The variation across years and PDs in this study established a range of environments that allowed for a more broad examination of treatment effects and disease parameters than if environments had been consistently favorable or unfavorable for disease over years.

Impact of Maize Residue Treatments on Spike-borne inoculum. The intermediate objective in placing maize residues in the study area was to establish three (or two) distinct levels of local inoculum intensity. Treatments were compared in each of eight year-PD environments to determine the effect of residue level on spike-borne inoculum for this study. Table 2 shows the average cfu's per day washed from heads in each treatment. Residue treatments significantly affected cumulative inoculum load within two of the environments for 'Alsen' (2004-PD1 and 2004-PD2), and only one for 'Norm' (2004-PD2). In each case there was a significantly higher level of inoculum on heads within the 80% residue treatment compared to 0% or 30% residue treatments. In all other environments, residue treatments had no significant effect on inoculum load. Therefore, it cannot be said that distinct levels of spike-borne inoculum resulted from maize residue treatments. The whole-plot area may have been too small to overcome problems of fetch (upwind distance from major confounding inoculum sources) and edge-effects. These problems could have led to inter-plot interference. The 2004 experiments give the best example of local inoculum dynamics under a high disease-pressure environment. In 2004, local inoculum apparently did contribute significantly to total inoculum concentration on spikes and therefore the influence of maize residue under the wheat canopy cannot be ignored. However, their influence under most environments is less obvious because of the relatively high level of 'external' inoculum entering the system concurrently.

Impact of Maize Residue, Variety, and Late Planting on FHB and DON. Residue treatments produced no significant effects on any of the visual disease estimates in any environment (data not shown), however DON was significantly increased by the 80% residue treatment for one environment (2004-PD1). Table 3 presents average DON concentration in grain for all treatments under each environment. There was also a trend toward higher DON in grain with increased maize residue for several environments, though it was not significant. These results mirror closely the average CFU/head data (Table 2). It is hypothesized that inoculum on spikes and final DON in grain may have a strong relationship. This is being investigated further.

The two varieties in the study exhibited different levels of disease and DON under all environments and treatments. As expected, 'Norm' was significantly higher

in FHB and DON than 'Alsen' in nearly all cases. The most noticeable and most significant differences were in DON estimates, where 'Norm' accumulated toxin to a much higher level than 'Alsen' under comparable environments (Table 3). As overall FHB disease pressure increased (greater inoculum, higher overall disease) across environments, the differences between 'Norm' and 'Alsen' for all response variables increased, indicating a strong variety by environment interaction, which is also represented in the analysis of variance (Table 4). This suggests that even some level of resistance in the host to FHB was made relatively more valuable as disease pressure increased due to environment.

As mentioned, planting dates each year resulted in somewhat unique environments. The effect of late planting can be seen in the higher levels of FHB each year of this study and in the analysis of variance (Table 4). Though the effect of late planting could be inverted under certain environmental conditions such as a dramatic drought or cool period at or just prior to anthesis for a late planted crop, in general, late plantings will experience higher temperatures and greater stress on plants at anthesis.

Based on the higher degree of variability in disease and DON across environments than within any one environment, the overall influence of the weather component of the system was probably the largest factor in any of the disease estimates or in toxin accumulation. The environmental variability coupled with the earlier mentioned confounding effects on inoculum load (interplot or 'external' inoculum interference) probably masked the relatively subtle effects of the residue treatments. Thus, for this set of experiments, maize residue on the soil surface was not a good predictor of disease development risk. This is contrary to the generally accepted etiological models of FHB which place high risk on corn and small grains residue beneath a susceptible crop (Parry et al. 1995; McMullen et al. 1997; Sutton, 1982; Andersen, 1948; Bai and Shaner 1994; Pererya et al. 2004). The impact of maize residue in this set of studies was much less important than the impact of the environment on disease development. Apparently, non-local inoculum was a significant portion of total inoculum load in this study. Compared to plots in this study, residue-based inoculum in farm-scale fields would likely be more influential on disease development. Furthermore, a degree of host resistance will be made relatively more valuable under high disease pressure situations, potentially reducing disease and DON compared to more susceptible varieties. With varieties under development and in the first few years of production generally having higher levels of FHB resistance than in earlier decades, overall impact of FHB will likely begin to decline. The risk of severe epidemics such as in the Northern Great Plains in the early 1990's will be lessened by the widespread adoption of resistant varieties.

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Table 1. Mean amount (index[1]) of FHB (%) by variety and year across all residue treatments.

Year	Variety		Average of both varieties	Disease Category[2]
	'Alsen'	'Norm'		
2003	1.8	5.2	3.5	Low
2004	19.1	48.9	34.0	High
2005	7.0	17.4	12.2	Moderate
2006	0.6	1.5	1.1	Very Low

Differences in disease values were highly significant between varieties each year, and within each variety over years.

[1]index=disease incidence*severity; an indicator of disease level within a population.

Table 2. Average Daily Inoculum Load on Heads by Residue Treatment.

	Inoculum load (cfu) per head per day (average)[1]							
	2003		2004		2005		2006	
'ALSEN'	[2]PD 1	PD 2	PD 1	PD 2	PD 1	PD 2	PD 1	PD 2
0% Residue	[3]--	--	388[a]	1408[a]	529[a]	579[a]	29[a]	84[a]
30% Residue	--	--	380[a]	1523[a]	--	--	--	--
80% Residue	--	--	493[b]	2133[b]	475[a]	662[a]	39[a]	43[a]

	Inoculum load (cfu) per head per day (average)							
	2003		2004		2005		2006	
'NORM'	PD 1	PD 2	PD 1	PD 2	PD 1	PD 2	PD 1	PD 2
0% Residue	88[a]	167[a]	472[a]	1654[a]	639[a]	754[a]	34[a]	52[a]
30% Residue	102[a]	282[a]	355[a]	1686[a]	--	--	--	--
80% Residue	146[a]	200[a]	534[a]	1990[b]	693[a]	781[a]	38[a]	39[a]

[1] letters after mean values indicated significant differences among the means (within an environment only)
[2] PD=planting date
[3] 'Alsen' was not sampled in 2003, per collaborators protocol.

Table 3. Average DON In Grain for each environment

	DON in grain, ppm (average)[1]							
	2003		2004		2005		2006	
'ALSEN'	[2]PD 1	PD 2	PD 1	PD 2	PD 1	PD 2	PD 1	PD 2
0% Residue	0.25	0.25	0.53	3.75	0.25	0.66	0.25	0.25
30% Residue	0.25	0.25	0.59	4.95	--	--	--	--
80% Residue	0.25	0.25	1.53	7.38	0.51	1.00	0.25	0.25

	DON in grain, ppm (average)							
	2003		2004		2005		2006	
'NORM'	PD 1	PD 2	PD 1	PD 2	PD 1	PD 2	PD 1	PD 2
0% Residue	0.60	0.78	4.43	17.18	1.75	2.15	0.25	0.25
30% Residue	0.98	0.87	4.63	17.30	--	--	--	--
80% Residue	0.40	1.28	7.93	18.23	2.05	2.43	0.25	0.25

[1] limit of detection = 0.5ppm, samples below detection limits were assigned the value 0.25ppm for calculations
[2] PD=planting date

No significant differences were detected within an environment among residue levels for the same variety except for 2004-PD1 (80% treatment yielded higher levels of DON in grain for both 'Alsen' and 'Norm'.

In all environments except 2006-PD1 and PD2, 'Norm' contained significantly higher DON in grain than 'Alsen'

Table 4. ANOVA for Disease Parameters and Toxin Levels in Grain across environments (2003-2005 only).

[1]Analysis of Variance

	[2]F-values			
source of variation	DON	INC	SEV	INDX
planting-date(year)	294.12***	5.24**	20.99***	19.78***
variety	389.43***	36.18***	109.19***	125.91***
planting-date(year)*variety	60.38***	1.6	15.06***	11.91***

[1]PROC Mixed, SAS 9.1, SAS Inc. Cary, NC
[2]Prob.>F values indicated by asterisk: *<0.05; **<0.01; ***<0.001

SPORE LOAD, DISEASE, AND DON: AN INOCULUM GRADIENT STUDY USING SISTER WHEAT LINES.

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ABSTRACT

A greenhouse study was conducted utilizing a range of *Gibberella zeae* inoculum concentrations applied to differentially Fusarium head blight (FHB)-susceptible sister lines of hard red spring wheat. The study was designed to evaluate the relationship between spore concentration on wheat spikes and deoxynivalenol (DON) in grain. Prior observations from several years of field research indicated that high inoculum density on spikes often was associated with high levels of DON in grain, even when disease levels were not well correlated with either DON or inoculum level. The present study utilized aqueous inoculum treatments at seven concentrations from 100 to 100,000 cfu/ml plus a control treatment applied to two sister wheat lines, SD3851 and SD3854. The lines are similar in agronomic characteristics but differ in susceptibility to FHB. Both lines were derived from the same population however line SD3851 possesses resistance conferred by the *Fhb1* QTL while SD3854 does not. Inoculated spikes were incubated for 72 hours under 100% RH, then left under ambient GH conditions for 12 additional days. At 15 days post-inoculation, disease assessments were completed and sub-samples of either whole spikes or grain only were ground for mycotoxin analysis. As expected, line SD3851 had less disease and accumulated less DON than SD3854. Both lines tended to accumulate higher levels of DON as inoculum concentration increased, but the susceptible line showed a greater response in both grain and whole-head sub-samples. Whole-head samples contained 4 to 10 times as much DON as grain-only samples, suggesting that chaff tissues might serve as a source of DON which could move to grain under certain environmental conditions. Furthermore, the FHB-susceptible line accumulated about twice as much DON as the FHB-resistant line over the range of inoculum densities comparable to field levels in SD (approximately 125 to 1250 cfu/spike). This further supports the idea that the *Fhb1* QTL or any type of host resistance is a crucial part of the USWBSI-stated mission for developing "...control measures that minimize the threat of Fusarium head blight (scab), including the reduction of mycotoxins...". This study also lends support to the idea that inoculum incident on spikes may be a useful predictor of DON in grain when accompanied by information about host resistance.

A QUANTITATIVE SYNTHESIS OF THE RELATIVE EFFICACY OF TRIAZOLE-BASED FUNGICIDES FOR FHB AND DON CONTROL IN WHEAT.

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ABSTRACT

Fungicide efficacy against FHB and DON in wheat has been highly inconsistent. Of the classes of fungicides most widely tested, triazoles have been the most effective; however, even among triazoles, the results have been highly variable. A recent quantitative synthesis of the results from over 100 Uniform Fungicide Trials (UFTs), showed that the efficacy of 38.7% tebuconazole (Folicur 3.6F), the longstanding industry standard for FHB and DON control, varied among individual studies and was generally higher in spring wheat than winter wheat. Besides tebuconazole, other triazole-based fungicides have been tested against FHB and DON, with some showing numerically (if not always statistically) superior efficacy relative to tebuconazole. One such fungicide is the recently-registered 41% prothioconazole (Proline 480 SC). In some individual studies, when used as a solo active ingredient or in combination with tebuconazole (either as a premix or a tank mix), this product contributed to significantly greater reduction in FHB and DON than tebuconazole alone. In other studies, however, the differences in efficacy were merely numerical. Similar results were observed in comparisons between metconazole and tebuconazole, suggesting that study- or environment-specific factors were probably influencing the performance of these fungicides. Using data collected from 10 years of UFTs, a multivariate meta-analysis was performed to evaluate the overall and relative efficacy of triazole-based fungicides against FHB and DON, and to determine whether efficacy was consistent across wheat types. Propiconazole (PROP), prothioconazole (PROT), tebuconazole (TEBU), metconazole (METC) and prothioconazole+tebuconazole (PROT+TEBU) fungicides were applied at flowering and disease index and DON concentration were quantified. Based on percent control (*C*), all fungicides led to a reduction in FHB and DON relative to the untreated check. PROT+TEBU was the most effective product against index, with an overall mean *C* of 52%, followed by METC (50%), PROT (48%), TEBU (40%), and PROP (32%). For DON, METC was the most effective, with a mean *C* of 45%; PROT+TEBU and PROT were of equal efficacy, with a *C* of 42%; whereas TEBU and PROP were the least effective, with mean *Cs* of 23 and 12%, respectively. In general, fungicide efficacy was higher in spring wheat than in winter wheat studies. When considering the efficacy of PROP+TEBU, METC, PROT and PROP relative to TEBU, that is, using TEBU as the reference for comparison in the meta-analysis instead of the untreated check, all products, with the exception of PROP, were significantly more effective than TEBU against both index and DON. Using the estimated mean efficacy of each fungicide against IND and DON and the estimated between-study variability in efficacy form the meta-analysis, the probability of each fungicide achieving > 50 (p_{50}) percent control in a new (single), randomly-selected study (conducted in a way similar to the UFTs) was estimated. For spring wheat, METC had the highest p_{50} values for both index (0.64) and DON (0.56), followed closely by TEBU+PROT and PROT. For winter wheat, TEBU+PROT had the highest p_{50} value for index (0.42), followed by PROT (0.36),

and METC (0.31), whereas for DON, PROT, TEBU+PROT, and METC had comparable p_{50} values (0.31, 0.27, and 0.26).

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This material is based upon work supported by the U.S. Department of Agriculture, under Agreement No. 59-0790-4-112. This is a cooperative project with the U.S. Wheat & Barley Scab Initiative. Any opinions, findings, conclusions, or recommendations expressed in this publication are those of the author(s) and do not necessarily reflect the view of the U.S. Department of Agriculture.

AN INTEGRATED APPROACH TO MANAGING FHB AND DON IN WHEAT: UNIFORM TRIALS 2007.

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OBJECTIVES

1) Evaluate the integrated effects of multiple strategies for FHB and DON management under a range of environmental conditions; and 2) increase grower adoption of multiple strategies by demonstrating that integrated management is the most effective means of reducing losses due to FHB/DON.

INTRODUCTION

Fusarium Head Blight (FHB) and the associated toxin (deoxynevalenol, DON) produced by its causal agent, *Fusarium graminearum*, continues to be a concern in every sector of the wheat and barley industries. Through years of research funded by the US Wheat and Barley Scab Initiative (USWBSI), several chemical, biological, and cultural management approaches have been evaluated and shown to contribute to FHB and DON reduction. However, when used individually, none of these approaches have been fully effective against FHB and DON. The effects of fungicide application, genetic resistance, and residue management (through crop rotation or tillage) are highly variable and strongly influenced by the environment. Under favorable weather conditions, moderately resistant varieties may become infected and DON contamination may exceed critical threshold levels. In the case of fungicides, efficacy varies from one trial to another, with overall mean percent control between 40 and 60% for index and between 30 and 50% for DON (for the most effective fungicides). Fungicides are generally most effective at reducing FHB and DON under moderate disease pressure and when applied to moderately resistant varieties than to susceptible varieties. In 2006, members of the then CBCC RAC of the UWBSI met with researcher and established protocols for conducting integrated FHB management trials. In 2007, these trials were implemented for the first time across multiple states and grain classes. The results of these trials are summarized herein.

MATERIALS AND METHODS

Field experiments were conducted to investigate the integrated effects of multiple management strategies on FHB and DON accumulation in wheat under natural conditions. The standard experimental design was a split plot with 3 to 6 replicate blocks. Wheat variety and fungicide application served as the whole-plot and sub-plot factors, respectively. In some individual trials, biological control agents and cropping sequence were used as additional treatment factors.

Plot dimensions and planting and cropping practices varied somewhat from trial to trial (see individual trial reports for details). In general, between three and six locally adapted and commonly cultivated varieties, with a range of susceptibility to FHB, were planted. There were two adjacent plots of each variety in each block. Sub-plot treatments were established by applying Proline + Folicur (as a tank mix of 3 fl. oz of each) or Prosaro (6.5 fl. oz/A) to one plot of each variety at the flowering date (Feekes' growth stage 10.5.1) of

the variety and leaving the other plot untreated. A non-ionic surfactant was added to the treatment at a rate of 0.125% v/v, and applications were made using CO_2-pressurized sprayers, equipped with Twinjet XR8002 nozzles or paired XR8001 nozzles, mounted at an angle (30 or 60°) forward and backward.

In each plot, percent FHB incidence (INC), diseased-head severity (SEV), index (IND; also known as field or plot severity), and *Fusarium*-damaged kernels (FDK) were measured. Plots were harvested and yield and test weight determined. Milled grain samples from each plot were sent to one of the USWBSI-funded DON Testing Laboratories for DON analysis.

Analysis of variance (linear mixed model) was used to evaluate the effects of variety, fungicide and their interaction on FHB intensity and DON content at each location. Percent control of FHB and DON was estimated for each treatment and treatment combination by using the level of disease and DON in the untreated plot of the most susceptible variety as the reference.

RESULTS AND DISCUSSION

A total of 15 trials were conducted in eight states (Table 1). FHB intensity and DON varied from one location to another, with some trials having zero or nominal disease development and DON contamination.

Kansas – Plots in this trial were artificially inoculated with *F. graminearum*-infested corn kernels and mist-irrigated to enhance disease development. As a result, FHB intensity and DON contamination were high, with mean index and DON ranging from 2 to 95% and 7.5 to 30.2 ppm, respectively. The effects of fungicide, variety, and their interaction on FHB index were statistically significant ($P < 0.05$). For DON, only the main effect of fungicide was statistically significant. Averaged across the three varieties, mean index was 43% in Prosaro-treated plots compared to 70% in the untreated check. For the three varieties evaluated (Harry, Jagalene, and Pioneer 2137) treated plots had significantly lower levels of disease than untreated plots. Untreated plots of Jagalene had the highest level of disease, with a mean index of 87.5%, whereas treated plot of Harry had the lowest level of disease with a mean index of 6.7% (Table 2). For DON, Prosaro-treated plots had significantly lower mean DON (16.7 ppm) than untreated plots (20.14), averaged across varieties. Among the varieties, mean DON content was highest in Jagalene and lowest in Pioneer 2137, however, this difference was only numerical (Table 2).

Kentucky – Due to dry conditions, FHB intensity and DON contamination were very low in this trial. Mean index and DON ranged from 0.03 to 2% and 0.05 to 1.3 ppm, respectively. For index, the main effects of fungicide and variety were statistically significant, whereas for DON, only the main effect of variety was statistically significant.

Missouri – Two trials were conducted in Missouri to evaluate fungicide and variety effects on FHB and DON. In the first (MO1), plots were planted no-till into corn residue and in the second (MO2), no-till into soybean residue. In MO1, mean FHB and DON levels ranged from 0.12 to 38% and 0.25 to 5.6 ppm, respectively, whereas in MO2, the corresponding ranges were 0 to 8, and 0.25 to 2, respectively. For index, all main and interaction effects were statistically significant in MO1 and only variety and variety x fungicide effects were significant in MO2. In both trials, the effects of fungicide and variety x fungicide interaction on DON were not statistically significant. In MO1, averaged across varieties, mean index in Proline 3+3-treated plots was 7.1% compared to 11.7% in the untreated check. Among the varieties, averaged across fungicide treatments, Elkhart has the highest level of disease (23.8%), followed by Pioneer 25R47 (18.2%), whereas Bess had the lowest mean level of disease (0.32%). Overall, the highest and lowest levels of disease occurred in untreated plots of Elkhart and treated plots of Bess, respectively (Table 2).

Nebraska – Mean FHB index ranged from 2.8 to 47.7% in this trial, with very similar mean levels of disease occurring in fungicide-treated and untreated plots, averaged across varieties. Among the varieties, Pioneer 2137 had the highest mean index (17.5%), followed by Jagalene (16.8%), and Harry (12.8%). The main and interaction effects of variety and fungicide treatment on index were not statistically significant.

North Dakota - Six FHB integrated management trials were conducted in North Dakota - three HRWW, two HRSW and one Durum. Disease and DON levels were not reported for two of the winter wheat trials. For the Durum and one of the HRSW trials, previous crop was used as an additional treatment factor (along with fungicide and variety). For the other spring wheat trial and the third winter wheat trial, 20 different varieties were planted.

For the Durum wheat trial (ND_D), the three-way interaction effect of previous crop, variety and fungicide on index was not statistically significant. However, the main effects of variety and fungicide and the interaction effects of previous crop x variety and fungicide x variety were significant. Mean index (averaged across variety and cropping sequence) in Prosaro-treated plots (8.54%) was significantly lower than mean index in untreated plots (20.04%). Averaged across cropping sequence and fungicide treatment, Monroe and Divide had the highest and lowest mean levels of disease, respectively. Overall, untreated plots of Monroe planted following HRSW (as the previous crop) had the highest mean index (36.54%). For DON, only the main effects of fungicide and previous crop were statistically significant. The highest mean level of DON occurred in untreated plots of Monroe and Lebsock (3 ppm) planted after HRSW and the lowest mean level occurred in treated plots of Grenora (0.97 ppm) planted after canola.

CONCLUSIONS

Percent control was estimated for a few of the trials to evaluate the efficacy of individual treatments and treatment combinations against index and DON. The results for three trials with the highest levels of disease are presented in Table 2. Trials with nominal levels of disease and DON were not included in the table, because percent control tends to be highly variable at low index and DON levels. In general, moderately resistant variety x fungicide treatment combination resulted in the highest percent control. For the trials with cropping sequence as a treatment factor, none-host crop + moderately resistant variety + fungicide generally resulted in the highest percent control (Table 2).

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Table 1. States, principal investigator, institution, wheat class and number of FHB Integrated management trials conducted in 2007.

State	PI	Institution	Wheat class	No. trials
KS	Bill Bockus	Kansas State Univ.	HRWW	1
NE	Stephen Wegulo	Univ. of Nebraska - Lincoln	HRWW	1
MO	Laura Sweets	Univ. of Missouri	SRWW	2
KY	Don Hershman	Univ. of Kentucky	SRWW	1
ND	Marcia McMullen	North Dakota State Univ.	HRSW	6
	Joel Ransom		Durum	
	Scott Halley		HRWW	
	Kent McKay			
NY	Gary Bergstrom[a]	Cornell Univ.	SRWW	2
SD	Kay Ruden[a]	South Dakota State Univ.	HRSW	1
OH	Pierce Paul[a]	Ohio State Univ.	SRWW	1

[a] Fusarium head blight did not develop.

Table 2. Mean FHB index and DON and estimated percent control for different treatments and treatment combinations.

	Index					DON				
Trial	Previous Crop	Variety	Treatment	Mean Index (%)	Percent Control	Previous Crop	Variety	Treatment	Mean Index (%)	Percent Control
Kansas	…	Jagalene	Untreated	87.50	0.00	…	Jagalene	Untreated	20.08	0.00
	…		Prosaro	67.50	22.86	…		Prosaro	18.03	10.21
	…	Harry	Untreated	46.00	47.43	…	Harry	Untreated	22.53	-12.20
	…		Prosaro	6.66	92.39	…		Prosaro	14.68	26.89
	…	P2137	Untreated	77.83	11.05	…	P2137	Untreated	17.82	11.25
	…		Prosaro	55.00	37.14	…		Prosaro	17.60	12.35
Missouri	…	Elkhart B	Untreated	29.38	0.00	…	…	…	…	…
MO1	…		Proline 3+3	18.23	37.95	…	…	…	…	…
	…	25R47 C	Untreated	23.68	19.40	…	…	…	…	…
	…		Proline 3+3	12.66	56.91	…	…	…	…	…
	…	25R54 D	Untreated	2.02	93.12	…	…	…	…	…
	…		Proline 3+3	1.92	93.46	…	…	…	…	…
	…	Roane E	Untreated	3.13	89.35	…	…	…	…	…
	…		Proline 3+3	2.66	90.95	…	…	…	…	…
	…	Bess A	Untreated	0.42	98.57	…	…	…	…	…
	…		Proline 3+3	0.22	99.25	…	…	…	…	…

Table 2. cont.

North Dakota ND_D	HRSW	Monroe	Untreated	36.54	0.00	HRSW	Monroe	Untreated	2.98	0.00
			Prosaro	9.79	73.21			Prosaro	1.40	53.02
		Lebsock	Untreated	20.42	44.12		Lebsock	Untreated	3.00	-0.67
			Prosaro	8.31	77.26			Prosaro	1.98	33.56
		Grenora	Untreated	12.41	66.04		Grenora	Untreated	2.80	6.04
			Prosaro	6.29	82.79			Prosaro	2.15	27.85
		Divide	Untreated	14.74	59.66		Divide	Untreated	2.32	22.15
			Prosaro	11.89	67.46			Prosaro	2.10	29.53
	Canola	Monroe	Untreated	25.22	30.98	Canola	Monroe	Untreated	2.25	24.50
			Prosaro	4.97	86.40			Prosaro	1.08	63.76
		Lebsock	Untreated	19.4	46.91		Lebsock	Untreated	2.05	31.21
			Prosaro	12.26	66.45			Prosaro	1.27	57.38
		Grenora	Untreated	23.9	34.59		Grenora	Untreated	2.00	32.89
			Prosaro	9.37	74.36			Prosaro	0.97	67.45
		Divide	Untreated	7.66	79.04		Divide	Untreated	1.93	35.23
			Prosaro	5.49	84.98			Prosaro	1.15	61.41

Trials with nominal levels of disease and DON were not included in the table.

FUNGICIDE EFFECTS ON FHB AND DON IN WHEAT ACROSS MULTIPLE LOCATIONS AND WHEAT CLASSES: UNIFORM FUNGICIDE TRIALS 2007.

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OBJECTIVE

Evaluate foliar fungicides for effectiveness against Fusarium head blight (FHB) and deoxynivalenol (DON) accumulation in wheat across multiple trials and different wheat classes.

INTRODUCTION

Fusarium head blight (FHB), caused predominantly by *Fusarium graminearum*, continues to impact every sector of the wheat and barley industries, causing substantial yield and quality losses. *F. graminearum* produces a mycotoxin called deoxynivalenol (DON) (among other toxins) which may accumulate to unacceptable levels in harvested grain. DON levels above 2 ppm may render grain and their by-products unfit for commercialization and consumption. Efforts to minimize the impact of FHB and DON have been based on the use of management strategies such as host resistance, crop rotation, tillage, and fungicide application. Through collaborative research involving scientists from multiple states, representing various wheat-growing regions, Uniform Fungicide Trials (UFTs) have been conducted annually since 1998 to evaluate fungicide efficacy against FHB and DON. The 2007 results from 23 UFTs across 6 states are presented herein.

MATERIALS AND METHODS

In general, each trial consisted of six core fungicide treatments and an untreated control in a randomized complete block design, with four replicate blocks (one trial had 3, two had 5, and another had 8 blocks). The core treatments were:

- Non-treated control;
- Folicur at 4.0 fl oz/A;
- Prosaro at 6.5 fl oz/A;
- Caramba at 13.5 fl oz/A;
- Topguard at 14 fl oz/A;
  Proline at 5 fl oz/A;
- Tilt at 4 fl oz/a;

Other treatments evaluated in separate individual trials were Proline at 3 fl. oz + at Folicur 3 fl. oz; Caramba at 10 fl. oz/A; Caramba at 8.2 fl. oz/A; Punch at 6 fl. oz/A; Proline at 3 fl. oz/A; Topguard at 10 fl. oz/A; Stratego at 10 fl. oz/A; Quadris 8 fl. oz/A; Dithane at 2 fl. oz/A; Quilt at 14 fl. oz/A; Folicur at 2 fl. oz/A + Topguard at 8 fl. oz/A; Headline at 8 fl. oz/A; and Folicur at 2 fl. oz/A. All treatments were applied at Feekes 10.5.1. A non-ionic surfactant was added to each treatment at a rate of 0.125% v/v, and applications were made using CO_2-pressurized sprayers, equipped with Twinjet XR8002 nozzles or paired XR8001 nozzles, mounted at an angle (30 or 60^o) forward and backward.

Planting and crop production practices varied somewhat from trial to trial. See individual trial reports for details. Most plots were planted with a susceptible cultivar. To enhance disease development, plots were either planted into corn or wheat residue and/or artifi-

cially inoculated with *F. graminearum*-infested kernels. Many plots were mist-irrigated as a means of enhancing production of, and infection by fungal inocula. In each plot of each trial, percent FHB incidence (INC), diseased-head severity (SEV), index (IND; also known as plot or field severity), and *Fusarium*-damaged kernels (FDK) were measured as previously described (McMullen, et al., 1999). DON accumulation was measured at one of the USWBSI-funded DON Testing Laboratories.

Each trial was analyzed separately using a mixed effect model in PROC MIXED of SAS to determine treatment effect on FHB, DON, yield (bu/ac) and test weight (lb/bu). Linear contrasts were used to make pair-wise comparisons between treatment means or means across groups of treatments. Studies with zero or nominal levels of disease and DON were not analyzed.

RESULTS AND DISCUSSION

Weather conditions in both winter wheat and spring wheat areas were generally unfavorable (hot and dry during flowering) for FHB development. In addition, adverse weather conditions (floods in some areas and cold temperatures in others) caused plots to be lost and the results to be highly variable in some trials. Consequently, non-irrigated trials and a few irrigated trials had nominal disease development. Mean and maximum FHB index across all replicates of the untreated check plots ranged from 0 to 26.28 and 0 to 55.00%, respectively (Table 1). In 15 of the 23 trials, mean index in the untreated check was less than 1% and less than 2% in 18 of the 23 trials. FHB intensity was highest in the Clarksville and East Lansing trials, with mean index of 26.3 and 21.9%, respectively.

In three (Urbana, IL, East Lansing, MI, and Fargo, ND) of the five trials with mean index in the untreated check greater than 2%, fungicide treatment had a significant ($P < 0.05$) effect on FHB index (Table 1). Based on pair-wise differences between each treatment and the check, Caramba at 13.6 fl.oz/A was the most effective treatment in the Urbana trial; Proline at 3 fl.oz/A was the most effective treatment in the East Lansing trial, and Proline at 5 fl.oz/A was the most effective treatment in the Fargo trial (Table 1). The corresponding percent controls (Hershman and Milus, 2003, Paul et al 2007) resulting from these treatments in their respective trials were 97, 91, and 81%. Although the Clarksville trial had the highest level of disease and the greatest difference in mean index between the Topguard treatment (14 fl. oz/A) and the check (17%), this difference was not statistically significant ($P = 1.00$). This was probably because of the high variability observed in this trial. Similar results were observed for other measures of FHB intensity (SEV, INC, and FDK). Since index is a direct function of INC and SEV (see Paul et al., 2005), only the results for IND are summarized herein.

DON content of the grain was reported in 13 of the 23 trials. In nine of these trials, DON levels in the untreated check were below 1 ppm. Trials conducted in Browntown, IL; Clarksville, MI; East Lansing, MI; and Langdon3, ND were the only trials with DON levels in the check close to or above 2 ppm (Table 2). In these trials, Prosaro at 6.5 fl.oz/A, Punch at 6 fl.oz/A, Proline at 5 fl. oz/A, and Caramba at 14 fl. oz/A, respectively, were the most effective treatments. However, the difference in mean DON between fungicide-treated plots and the untreated check was not significantly different from zero in the Clarksville and East Lansing trials (Table 2). Punch resulted in a 47% reduction in DON relative to the check in the Clarksville trial; however, the mean level of DON in Punch-treated plots still exceeded the critical threshold level of 2 ppm.

CONCLUSION

In summary, in the trials with some level of disease, fungicide treatments reduced FHB intensity and DON accumulation relative to the untreated check (based mainly on data from four locations). Fungicide efficacy varied among the trials, with percent control ranging from 67 to 97% for index and 41 to 60% for DON. However, since the overall levels of disease and DON were very low, these results should be interpreted with caution. Paul et al. (2007a, 2007b) showed that FHB and DON responses to fungicide treatments were most variable at low levels of disease (index < 2%) and DON (< 1 ppm) than at intermediate or high levels. In general, the overall levels of disease and DON in 2007

were too low for us to make broad conclusions regarding the treatments evaluated.

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Table 1. Fungicide effect on Fusarium head blight index – 2007 UFT.

Trial		Wheat Type	Most effective Treatment[a]				Index (%) Check	
State/PI	Location		Treat	IND (%)	Percent Control	*P*	Mean	Max
IL/Adee	Browntown	W	…	…	…	…	0.00	0.00
	Monmouth	W	…	…	…	…	1.23	3.11
IL/Bradley	Urbana	W	Caramba 13.5 fl.oz	0.17	97	0.006	5.90	12.73
LA/Padgett	Crowley 1	W	…	…	…	…	1.20	1.44
	Crowley 2	W	…	…	…	…	0.34	0.55
	Macon Ridge 1	W	Prosaro 6.5 fl oz	0.51	78	0.189	2.37	3.60
MI/Brown-Rytlewski	Clarksville	W	Topguard 14 floz	8.73	67	0.100	26.28	55.00
	East Lansing	W	Proline 3 fl. oz	2.00	91	<0.01	21.90	32.50
	Saginaw	W	…	…	…	…	0.00	0.00
	Sandusy	W	…	…	…	…	0.00	0.00
MO/Sweets	Columbia 1	W	…	…	…	…	1.00	1.60
	Columbia 2	W	…	…	…	…	0.48	0.80
ND/McMullen	Fargo	S	Proline 5 fl. oz	0.55	81	<0.01	2.85	3.30
	Langdon 1	S	…	…	…	…	0.40	0.50
	Langdon 2	S	…	…	…	…	0.20	0.50
	Langdon 3	S/D	…	…	…	…	0.75	1.00
SD/Draper	Brookings 1	S	…	…	…	…	0.73	1.32
	Brookings 2	S	…	…	…	…	0.74	1.94
	Groton 1	S	…	…	…	…	0.00	0.00
	Groton 2	S	…	…	…	…	0.29	1.00
	Watertown 1	S	…	…	…	…	0.00	0.00
	Watertown 2	S	…	…	…	…	0.00	0.00

[a] Treat = the most effective treatment (s) within each trial based on the pair-wise difference between mean index for each treatment and the check; IND (%) = mean index across plots receiving the most effective treatment; *P* = level of significance from *t* test of the difference between mean IND across plots receiving the most effective treatment and the untreated check ($P < 0.05$ ➔ significant different). All tests of significance were done using arcsine-transformed IND. … = Trials with zero or nominal levels of disease.

Table 2. Fungicide effect on DON – 2007 UFT.

Trial		Wheat Type	Most effective Treatment[a]				Index (%) Check	
State/PI	Location		Treat	DON (ppm)	% Reduction	*P*	Mean	Max
IL/Adee	Browntown	W	Prosaro 6.5 fl.oz	0.76	60	<0.01	1.91	3.50
	Monmouth	W	Topguard 14 fl.oz	0.17	58	0.16	0.40	0.67
IL/Bradley	Urbana	W	Prosaro 6.5 fl.oz	0.21	52	0.02	0.43	0.62
MI/Brown-Rytlewski	Clarksville	W	Punch 6 fl.oz	3.60	47	0.15	6.80	8.40
	East Lansing	W	Proline 5 fl.oz	1.10	41	0.17	1.85	2.30
	Saginaw	W	…	…	…	…	0.00	0.00
	Sandusy	W	…	…	…	…	0.05	0.10
ND/McMullen	Fargo	S	Proline 5 fl oz	0.38	58	0.02	0.90	1.10
	Langdon 1	S	…	...	…	…	0.83	0.90
	Langdon 2	S	…	…	…	…	0.17	0.50
	Langdon 3	S/D	Caramba 13.5 fl.oz	1.40	53	0.01	2.97	3.80

[a]DON data were not available for some trials or available but equally low (below 1 ppm) for all treatments.
[b]Treat = the most effective treatment within each trial based on the pair-wise difference between mean DON for each treatment and the check; DON (ppm = mean DON across plots receiving the most effective treatment; % reduction = percent reduction in DON; *P* value = level of significance from *t* test of the difference between mean DON across plots receiving the most effective treatment and the untreated check ($P < 0.05$ ➔ significant difference). All tests of significance were done using log-transformed.
… = Trials with zero or nominal levels of DON.

INFLUENCE OF SRWW, HRSW, AND HRWW VARIETIES ON THE RELATIONSHIP BETWEEN FHB AND DON.

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ABSTRACT

The relationship between visual estimates of Fusarium head blight (FHB) intensity and deoxynivalenol (DON) content of wheat is of interest to both researchers and producers because visual symptoms often are used as an indication of DON contamination of grain. In general, there is a significant positive relationship between FHB and DON, however, this relationship may vary among studies, and in some instances, fairly high levels of DON may accumulate in the absence of visual symptoms of FHB, or conversely, relatively high levels of visual symptoms may be associated with disproportionately low levels of DON contamination. The association between FHB and DON may be influenced by weather conditions, fungicide treatment, pathogen aggressiveness and DON producing ability, and variety resistance to FHB and DON. Field experiments were conducted in Nebraska, North Dakota and Ohio to evaluate the influence of variety resistance on the relationship between FHB and DON. At each location, locally adapted varieties with different levels of resistance to FHB (based on visual symptoms) were planted in a randomized complete block design, with three replicate blocks. The varieties evaluated were HRWW varieties Harry and Pioneer 2137, in Nebraska; HRSW varieties Trooper, Steel-ND, and Glenn, in North Dakota; and SRWW varieties Cooper, Hopewell, and Truman, in Ohio. Plots were inoculated at anthesis, and in each plot of each variety, diseased spikes in different severity categories were tagged. Tagged spikes were hand-harvested, cleaned, and a sample of grain from each disease category was analyzed for DON. DON content varied among varieties in each disease category in the three wheat classes. In all cases, DON generally increased with increase in disease intensity. Of the two HRWW varieties evaluated, Pioneer 2137 had lower mean DON contamination than Harry at all FHB severity levels. Among the SRWW varieties, Hopewell had the highest and Truman the lowest mean levels of DON in all disease categories. In general, Cooper, the susceptible SRWW variety, had DON content comparable to that of Truman, the moderately resistant SRWW variety. Among the spring wheat varieties, Trooper had higher mean DON content than Glenn and Steel-ND at all severity levels. Between Glenn, the moderately resistant HRSW variety, and Steel-ND, the moderately susceptible HRSW variety, the levels of DON contamination were similar in most cases.

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DONCAST: SEVEN YEARS OF PREDICTING DON INWHEAT ON A COMMERCIAL SCALE.

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ABSTRACT

Accurate predictions of mycotoxins in harvested grain are useful to help prevent entry of toxins into the food chain. DONcast is an empirical model for predicting mycotoxins in mature wheat grain, mainly as a decision support tool. To our knowledge, DONcast is the only mycotoxin prediction tool that is published and deployed commercially in the world; it was deployed in Ontario (Canada) in 2000, Uruguay (South America) in 2002, and has been undergoing a validation/calibration process in France since 2004. DONcast is attractive to the industry because: 1) prediction accuracies of over 85% have been demonstrated across diverse environments for making decisions on whether or not to apply a fungicide at heading, and 2) of the efficient platform for hosting the model and access to accurate input variables (mainly weather) that result in the following outputs: a) field- or site-specific DON predictions (Site-Specific DONcast – SSD), or b) regional-scaled (map format) outputs, all of which are conveniently managed through Weather INnovations Incorporated or WIN www.weatherinnovations.com (Chatham, ON, Canada). However, after seven years of commercial deployment, users need constant reminders on the limitations of both versions, learn how to interpret predictions toward management decisions, and be warned about unrealistic expectations of model-based predictions especially when they are derived from unrealistic weather or agronomic (or lack thereof) inputs. It has been well documented by others that FHB infection and DON accumulation is highly responsive to weather (mainly around heading), varietal susceptibility, and to the management of crop residue on the soil surface; therefore, these inputs should not be ignored. We will demonstrate that the accuracy of predictions on a regional-scale (i.e., map format) may be less than acceptable because input variables such as weather, wheat variety, crop rotation, and tillage effects tend to be over-generalized; all of these inputs are used in the Site-Specific Calculator. Although prediction maps produced on a regional scale are useful for establishing warnings or trends and are popular amongst growers, the most accurate predictions are derived from input variables that are both accurate and representative of individual fields. Both the input variable database and deployment of these predictions on a field-scale effort are enormous, considering the database is updated daily and the platform must be easily accessible to agribusiness and growers through the internet. The platforms and experiences that have evolved over seven years of commercial use will be presented in more detail.

EFFECTS OF FUNGICIDES ON FHB CONTROL AND YIELD OF WINTER WHEAT CULTIVARS IN NORTH DAKOTA.

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ABSTRACT

Research was conducted during three years (2005-2007) to determine the benefits of applying registered fungicides on a range of adapted winter wheat cultivars in North Dakota (ND). Experiments consisted of a factorial combination of fungicides and cultivars laid out in a RCBD with a split plot arrangement. The fungicide treatments served as the main plots and consisted of no fungicide or applying tebuconazole in 2005-06 or tebuconazole and prothioconazole in 2007 at early flowering. Cultivars served as the subplots and consisted of 12-18 cultivars commonly grown in ND or cultivars recently released by breeding programs in the region. Fungicide consistently improved yield and grain quality. Yield increases were associated with the control of leaf spots and especially leaf rust, and in two environments with the control of FHB. Fungicides were profitable when applied to the most disease resistant cultivars when disease pressure was high, but were not beneficial when disease pressure was low. Most of the added value from the use of fungicides resulted from increases in grain yield, but in environments with significant disease pressure, improved test weight also contributed to an increase in the crop's value. Reductions in DON levels did not improve the grain's value in the years that it was measured as they were below the threshold where discounts apply. In the least fungicide-responsive environment the return from fungicides did not exceed the cost of the application when applied to the most disease resistant cultivars as they did not produce a yield improvement. In the environment with the greatest disease pressure, however, the most disease resistant types tended to be the ones with the greatest return from fungicides. This indicates the potential value of using disease prediction models if resistant cultivars are used. With the more susceptible cultivars, fungicides should probably always be applied in eastern ND. The value of using resistant varieties, even when planning to use fungicides, was illustrated in the high disease pressure years; the combination of resistance variety and fungicide resulted in the highest yield and generally the greatest return to the fungicide and the highest overall returns.

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EFFECTS OF FUSARIUM HEAD BLIGHT ON YIELD AND QUALITY PARAMETERS OF WINTER WHEAT.

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OBJECTIVES

To assess Fusarium Head Blight impact on winter wheat yield reduction, deoxynivalenol (DON) accumulation, sinking of DON content after grading, milling and baking. These parameters are discussed both from the view of susceptible and medium resistant varieties and by application of different fungicide treatment.

INTRODUCTION

Food safety is nowadays priority for cereal producers and grain-processing industry. Fusarium head blight causes severe yield losses and decreases baking and food quality (Mesterházy, 2003). The most frequent species in Europe are now *F. graminearum* and *F. culmorum* (Logrieco et Bottalico, 2001; Mesterházy, 2003), both of which produce mycotoxins (Joffé 1986, Abramson 1998, Chelkowski 1998). The basic toxins are deoxynivalenol (DON), zearalenone and nivalenol (Logrieco et al., 2003). These substances are highly heat and chemically stable. They may be entering the food chain by direct consumption of contaminated foodstuffs, implicitly through feedstuffs and consequently through animal products.

Most of registered winter wheat varieties are middle or high susceptible to FHB. The results of many researches show us that it is difficult to reach high resistance level and simultaneously high yield and necessary food quality (Mesterházy, 2003).

MATERIALS AND METHODS

We used six winter wheat varieties differed into 2 groups: a) tolerant group (with medium resistant varieties – Sakura, Simila, Petrus), b) susceptible group (Darwin, Mladka, Sulamit). The project was sown in 3 replications and 4 various fungicide treatments: 1) control – without artificial infection and fungicidal treatment, 2) infection – with artificial *Fusarium* infection, without fungicide, 3) infection + fungicide, 4) infection + targeted fungicide. Variant infection + fungicide was sprayed with Tango Super (1l/ha, active substances epoxiconazole 84g/ha and fenpropimorph 250g/ha) in growing stage DC 37 – 39. In the variant infection + targeted fungicide was used Tango Super fungicide in DC 37-39 and targeted fungicide Caramba 24 hours before *Fusarium* infection (1l/ha, active substance metconazole 60g/ha). The experiment was based by small parcel sowing machine type Hege. Final parcel area was $10m^2$.

Inoculum with spore concentrations of 6-7 x 10^6 spores/ml was prepared and each parcel was infected with 1 liter of inoculum. Infections run up in full flowering period according to each variety term. Symptomatic evaluation was carried in 21st day after the infection. The experiment was harvested by plot harvester. The grain was analyzed; mycotoxins assessment in grain, flour, bread and bran was determined immunochemically using ELISA.

RESULTS AND DISCUSSION

Symptomatic evaluation **-** The results are average of three years (2005-2007). Head blight symptoms were evaluated on a 1-9 scale (9 - without symptoms, 1 - 100% disease development). Tolerant varieties have with strong infectious pressure significantly lower occurrence of pathogen then susceptible varieties. The difference between infection and non-targeted fungicide is not significant, while targeted fungicide lead to the less presence of symptoms (the evaluation was about 1 point better).

Yield reduction – Targeted treatment was significantly effective in susceptible varieties, which increased their yield about 16% compared to infection variant. Targeted treatment was less significant in tolerant varieties; their yield was higher about 3%. In the susceptible varieties was the lowest yield reduction in targeted treatment (16%), the highest yield reduction was in untreated variant (32%). Yield reduction in tolerant varieties was 12% by targeted treatment, 15% by infection. These results clearly advert to importance of variety tolerence. The active fungicide protection is questionable. Fungicide must be used preventively before symptoms appearance respectively in the right time. ¨

DON content – In the chart 3 is deoxynivalenol content by 4 fungicide treatments. European Commission devised the limits for DON 1.25 ppm for raw wheat and 0.5 ppm for bread. Tolerant varieties contain about 2/3 less DON than susceptible ones. Targeted fungicide treatment takes positive effect both in tolerant and susceptible varieties and reduces the DON content about more than 50%.

Grain processing and DON content – Figure 4 represent infection variant of 4 varieties, 2 susceptible (Darwin, Sulamit) and 2 medium resistant (Sakura, Simila). Once again is perceived significantly lower DON content in resistant varieties. It is possible to lower DON content just by grading, and that was between 30 – 50%. 70 – 80% of DON proceeds from grain to flour. Due to the high heat stability is the DON occurrence in bread approximately the same as in flour.

Development of tolerant varieties is the most effective protection against FHB infection and mycotoxin accumulation. Targeted fungicidal treatment highly influences mycotoxin accumulation and yield in susceptible varieties. However the application date in this work was accurately determined (24 hours after infection), estimation of the application time is doubtful in practice. Non-targeted fungicidal treatment is not explicit. Grading on the 2,2mm sieve causes reduction of the DON content up to 50%. Further manipulation as milling or baking has not so significant influence and major part of DON proceeds to the bread.

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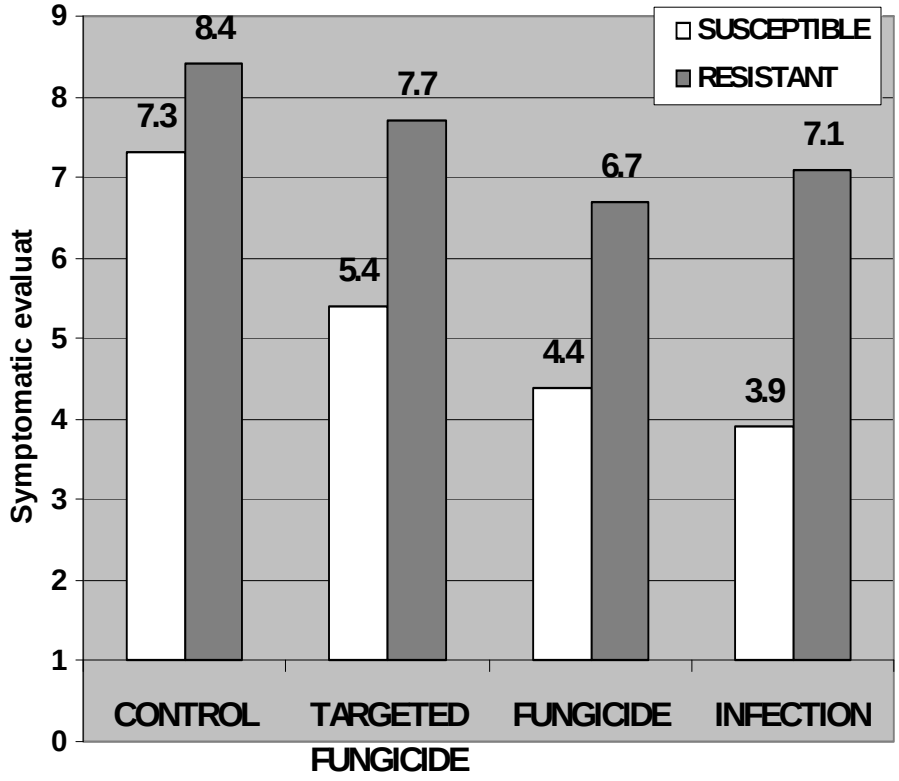

Fig. 1: Symptomatic Evaluation (2005-2007)

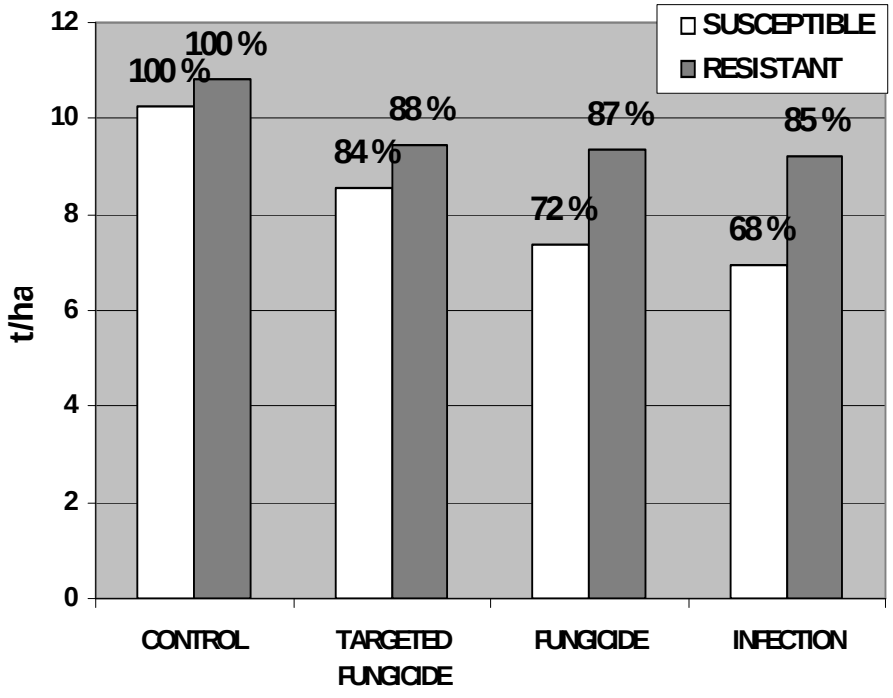

Fig. 2: Yield in Different Treatment (2005-2007)

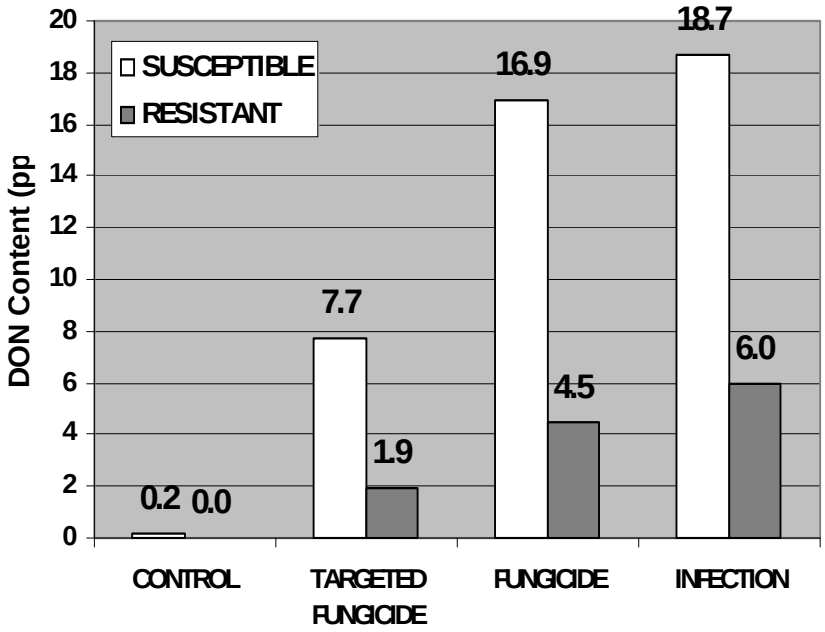

Fig. 3: DON Content (2005-2006)

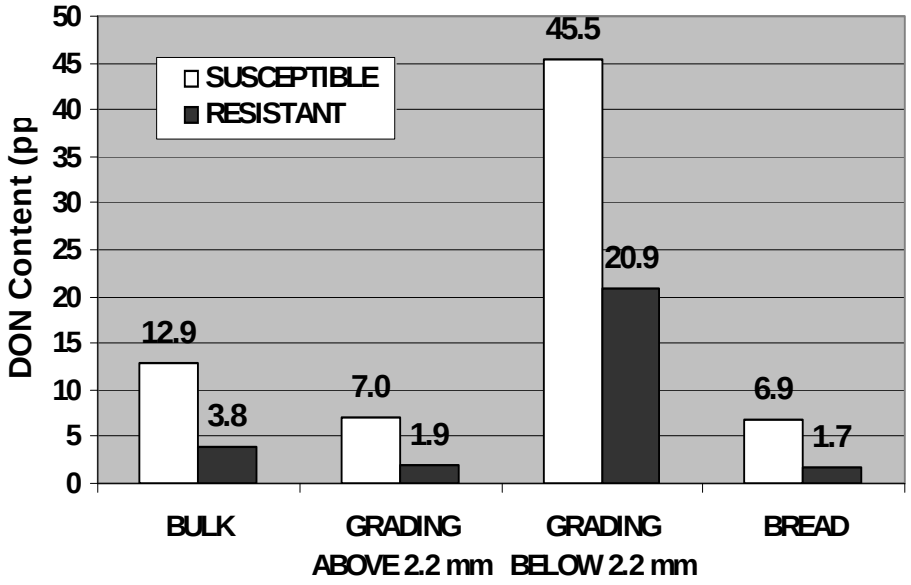

Fig. 4: Grain Processing (2005-2006)

2007 UNIFORM FUNGICIDE PERFORMANCE TRIALS FOR THE SUPPRESSION OF FUSARIUM HEAD BLIGHT IN SOUTH DAKOTA.

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ABSTRACT

Fusarium head blight (FHB – scab) has been a serious concern for wheat and barley producers in South Dakota for ten years and a serious epidemic impacted the state's wheat and barley crop in 2005. The objective of this study was to continue to evaluate the efficacy of various fungicides and fungicide combinations for the suppression of Fusarium head blight and other wheat diseases. Two hard red spring wheat cultivars, 'Briggs' and 'Forge', were planted at three South Dakota locations (Brookings, Groton, and South Shore/ Watertown) and Robust barley was planted at Brookings. Studies at two of these sites were conducted under ambient conditions. At the Brookings site, both the barley and the spring wheat trials received supplemental mist irrigation. Trial treatments from the Uniform Fungicide Trial treatments list for the suppression of FHB included an untreated check, Folicur (tebuconazole) applied at 4.0 fl oz/A, Prosaro (a premix of prothioconazole and tebuconazole) applied at 6.5 fl oz/A, Caramba (metconazole) applied at 13.5 fl oz/A, Topguard (flutriafol) applied at 14 fl oz/A, Proline (prothioconazole) applied at 5 fl oz/A and Tilt (propiconazole) applied at 4 fl oz/ A. All treatments included Induce, a non-ionic surfactant, applied at 0.125% v/v. Spring wheat trials were planted in a factorial randomized complete block design with six replications. The barley trial included four replications. Trial treatments were applied at anthesis (Feekes growth stage 10.51). The spring wheat and barley plots at the Brookings location were inoculated by spreading *Fusarium graminearum* (isolate Fg4) inoculated corn (*Zea mays*) grain throughout the field and providing overhead mist irrigation applied from 6:00 pm until 8:00 am each day for two weeks following anthesis. Other sites had natural inoculum from corn stalk residue and natural moisture conditions. Twenty-one days following treatment, plots were evaluated for leaf diseases, FHB incidence, FHB head severity, and FHB field severity. Samples were collected for *Fusarium* damaged kernels (FDK), deoxynivalenol (DON), grain yield, and test weight. Spring wheat under dryland conditions at South Shore/Watertown and Groton had negligible FHB. No significant differences resulted from the barley trial. On spring wheat in the Brookings trial, Prosaro, Caramba and Proline significantly reduced FHB Incidence, FHB Severity and FHB Index. All products except Folicur significantly increased grain yields with increases ranging from 35-42%. Total leaf disease pressure was very significant, as was leaf rust pressure which occurred late in the season. Control of the leaf diseases likely had a larger effect on yield than FHB control. Data is not yet available for DON.

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2007 UNIFORM TRIALS FOR THE PERFORMANCE OF BIOLOGICAL CONTROL AGENTS IN THE SUPPRESSION OF FUSARIUM HEAD BLIGHT IN SOUTH DAKOTA.

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ABSTRACT

Fusarium head blight (FHB – scab) has been a serious concern for wheat and barley producers in South Dakota for ten years and was very severe in parts of SD in 2005. The objective of this study was to continue to evaluate the efficacy of various fungicides and fungicide combinations for the suppression of Fusarium head blight and other wheat diseases under SD conditions. Briggs hard red spring wheat and Robust barley were planted at Brookings, South Dakota. Trial treatments included an untreated check; Prosaro (a premix of prothioconazole and tebuconazole) applied at 6.5 fl oz/A; TrigoCor 1448 (*Bacillus* sp.) from Cornell University, Ithaca, NY; and TrigoCor 1448 + Prosaro coapplied; 1BA (*Bacillus subtilus*) from South Dakota State University, Brookings, SD; 1BA + Prosaro coapplied, C3 (*Lysobacter enzymogenes*) from University of Nebraska, Lincoln, NE; C3 + Prosaro coapplied. The treatments were applied at anthesis. Plots were inoculated by spreading *Fusarium graminearum* (isolate Fg4) inoculated corn (*Zea mays*) grain throughout the field and providing overhead mist irrigation applied from 6:00 pm until 8:00 am each day for two weeks following anthesis. Twenty-one days following treatment, plots were evaluated for FHB incidence, FHB head severity, and FHB field severity. Plots were harvested for yield and test weight and samples were collected for *Fusarium* damaged kernels (FDK) and deoxynivalenol (DON).

Even with amending the environment in 2007, significant drought severely limited disease development. In the preliminary analysis, the assessments of FHB Severity and FHB Index disease components indicate a possible treatment effect in the barley study.

In the wheat study, all treatments showed a non significant increase in yield as compared to the untreated check. There were no differences among the treatments for the components of FHB.

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CHARACTERIZATION OF DON ACCUMULATION IN SRWW CULTIVARS WITH DIFFERENT LEVELS OF TYPE II RESISTANCE TO FHB.

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ABSTRACT

Under favorable conditions, deoxynivalenol (DON), a mycotoxin produced by *Fusarium graminearum*, may accumulate to unacceptable levels in harvested grain, making the grain and their by-products unfit for commercialization and consumption. The use of cultivars with resistance to FHB and DON is a widely recommended management approach for reducing the impact of this disease. However, resistance to *F. graminearum* is complex, with several different types (I, II, III, IV and V) reported, but not completely characterized. While it is clear that there is a positive association between FHB development and DON accumulation, the association between Type II (resistance to disease spread within the spike) and Type III resistance (resistance to DON accumulation) is unclear. It is quite possible for cultivars with similar levels of resistance to FHB to have different levels of resistance to DON accumulation. Some speculate that differential accumulation of DON among cultivars may be the result of differential fungal colonization of grain or the ability of some cultivars to detoxify DON. To quantitatively characterize the associations among FHB, fungal colonization, and DON accumulation in SRWW cultivars with different levels of Type II resistance to FHB, inoculated field trials were conducted at the Ohio Agricultural Research and Development Center, Wooster, during the 2007 growing season. Two experiments (1 and 2) were established, with three wheat cultivars (Cooper, susceptible; Hopewell, moderate susceptible; and Truman, moderately resistant) planted in a randomized complete block design with three replicate blocks. Plots were spray inoculated at early anthesis with a spore suspension (10^5 spores/ml) containing an equal proportion macroconidia and ascospores of *F. graminearum*/*G. zeae*. Approximately 35 days after inoculation, 20 wheat spikes in each of 11 severity categories were tagged. At maturity, spikes in each category were harvested and prepared for DON and PCR analyses. *F. graminearum* genomic DNA was extracted from each sample and a SYBR green-based real time polymerase chain reaction (RT-PCR) assay used to quantify fungal biomass. DON content was quantified by GC-MS at the USWBSI-sponsored laboratory at the University of Minnesota. Results from an analysis of covariance showed that DON content (ppm) increased with increasing FHB severity in all three cultivars. However, the rate of change in DON with change in severity (the regression slope) was greater for the susceptible cultivars than the resistant cultivar. The magnitude of the difference in DON content at a given level of severity among the cultivars was generally higher at high severity than at low severity. Contrastingly, estimated slopes for relationships between fungal biomass (log-transformed ng/mg) and DON (ppm) were similar for the three cultivars, suggesting similarity in DON accumulation with fungal colonization among the cultivars. However, the heights of the regression lines for the fungal biomass/DON relationships differed among the cultivars, indicating that for a similar level of fungal colonization, DON accumulation differed among the cultivars. Further research is in progress to evaluate these associations under different environmental conditions in an attempt to learn more about possible mechanisms involve in resistance to FHB and DON in wheat.

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This material is based upon work supported by the U.S. Department of Agriculture, under Agreement No. 59-0790-4-112. This is a cooperative project with the U.S. Wheat & Barley Scab Initiative. Any opinions, findings, conclusions, or recommendations expressed in this publication are those of the author(s) and do not necessarily reflect the view of the U.S. Department of Agriculture.

CONTRIBUTION OF LOCAL INOCULUM SOURCES TO REGIONAL ATMOSPHERIC POPULATIONS OF *GIBBERELLA ZEAE*.

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ABSTRACT

Decreases in tillage may have contributed to recent epidemics of FHB by increasing the amount of regional atmospheric inoculum available for infection. Where a large, regional source of atmospheric inoculum exists, crop rotation or tillage practices may not effectively reduce the risk of FHB in individual fields. An increased understanding of the contribution of local inoculum sources of *Gibberella zeae* (*Gz*) to regional atmospheric populations of the pathogen may aid in developing and/or excluding strategies for managing FHB. In 2007, we conducted 35 sampling flights with unmanned aerial vehicles (UAVs) 100 m above a large clonal inoculum source of *Gz* established at Virginia Tech's Kentland Farm. The UAVs were programmed to fly an orbital pattern such that one leg of the sampling path of UAV flew directly over the inoculum source during each of the passes. Our first flight was on 25 March at 11:30 am, and our last flight was on 24 May at 10:30 pm. We collected over 100 isolates of *Fusarium* spp. during these flights. All of the isolates were single-spored, grown in liquid culture, and suspended in 20% glycerol for cryogenic storage. We have tentatively identified a large portion of these isolates as *Gz*, and we will be conducting amplified fragment length polymorphisms (AFLPs) on these isolates in the coming months to unambiguously determine the percentage of the clonal isolate of *Gz* in our atmospheric collections. A series of runs with the atmospheric transport model HYSPLIT suggested that our inoculum source of *Gz* was transported at least a kilometer away from the ground surface within an hour. Our work continues to (1) elucidate the contribution of a local inoculum sources to atmospheric populations of the pathogen, and (2) develop and test a robust long-distance transport model for FHB forecasting/risk assessment. The ability to predict the regional transport of *Gz* from local inoculum sources may help refine risk models for FHB.

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ENVIRONMENTAL FACTORS INFLUENCING FHB SEVERITY AND DON IN BARLEY.

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ABSTRACT

We are investigating the relationship between environmental factors, crop stage, and barley genotype with Fusarium head blight (FHB) and DON accumulation in the grain. This project is associated with the established spring and winter wheat FHB-modeling efforts and aims to produce the information required to either validate one of the current FHB models for use in barley, or generate unique models.

Varieties of regionally adapted barley of both 2- and 6-row types were planted at multiple locations in the Northern Great Plains during the 2005-7 growing seasons. At least three varieties were common to each location. Plots were un-irrigated, a minimum of 1.5m x 4.6m in size, and replicated four times in a RCBD. Additional varieties were planted based upon availability and local producer preference. Crop stage was monitored regularly throughout the season and the date at which each plot was at Feekes 10.5 stage was noted. No additional inoculum was introduced into the plots. The incidence and severity of FHB was recorded on a minimum of 50 heads per plot at the soft-dough stage (approximately 21 days after heading). Environmental variables consisting of temperature, relative humidity, and precipitation were recorded by an on-site, or nearby, weather station.

Over the past three seasons, we have successfully collected data for 27 of the 38 locations planted. Unsuccessful locations were generally the result of extreme weather-related situations (e.g. floods) that resulted in crop destruction. The remaining locations provide a range in disease intensity, severity, and final deoxynivalenol concentration that we have used to identify weather variables, both simple and complex, that were associated with high FHB/DON situations in barley. For example, the average hourly temperature and relative humidity in the 10 days prior to full head emergence were both significantly correlated with final disease severity, but not DON content. In the available dataset, measurements of humidity after heading (e.g. vapor point depression) were the only factors associated with final DON concentration. From these results, we hypothesize that different environmental factors may be impacting this pathosystem in various ways and the development of a single model for both disease and DON prediction is unlikely.

DIFFERENTIAL SENSITIVITY TO TRIAZOLE-BASED FUNGICIDES AMONG ISOLATES OF *FUSARIUM GRAMINEARUM*.

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ABSTRACT

Samples of Fusarium head blight infected wheat spikes were collected from wheat fields across the state of Ohio. All isolates of *Fusarium graminearum* were identified based on colony and spore morphology. A subset of isolates from different Ohio counties was tested in vitro for sensitivity to Proline (41% prothioconazole) and Folicur (38.7% tebuconazole). Five isolates obtained from conventional wheat fields in Wood, Wayne, Shelby, Van Wert, and Delaware counties and a sixth from an organic wheat field in Wayne County were compared. The fungicides were dissolved in deionized water to achieve stock solutions with the proper concentration of active ingredient and added to PDA. Commercial grade prothioconazole and tebuconazole were evaluated at concentrations of 0.001, 0.01, 0.1 and 1.0 µg/ml and non-amended PDA was used as the control. A 5-mm-diameter plug from the edge of a fully colonized plate was transferred to the center of each plate for each concentration to be evaluated. Colony diameter was measured in three places once every 24 hours for seven consecutive days. The percent growth relative to growth on the control was calculated as the average colony diameter (at each concentration) minus 5 mm (diameter for the PDA plug) divided by the average colony diameter on the non-amended media, multiplied by 100. Sensitivity varied between Proline and Folicur and among isolates of *F. graminearum*. At concentrations of 0.1 and 1 µg/ml of active ingredient, isolates were generally more sensitive to Proline than Folicur, exhibiting slower and more restricted growth on Proline-amended media than on Folicur-amended media. The sensitivity profile of the isolates was similar for the two fungicides; the same isolates that exhibited the highest and lowest sensitivity to Folicur also exhibited the highest and lowest sensitivity to Proline. With the exception of isolate OHSHE6613, all isolates showed some level of growth on Folicur-amended media at all tested concentration. Conversely, with the exception of OHWAY1619, none of the isolates grew on Proline-amended media at 1.0 µg/ml. Research is in progress to conduct a more comprehensive evaluation of sensitivity of isolates from different wheat-growing regions to Folicur, Proline and other triazole-based fungicides.

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A METHOD FOR QUANTIFYING TRICHOTHECENES AND ERGOSTEROL IN SINGLE WHEAT FLORETS USING GAS CHROMATOGRAPHY WITH ELECTRON CAPTURE DETECTION.

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ABSTRACT

The relationship between fungal biomass and mycotoxin accumulation during Fusarium Head Blight infections is not completely understood. The purpose of this research is to develop a method to quantify deoxynivalenol and its acetylated derivatives as well as fungal biomass within single wheat florets. Ergosterol, a sterol unique to fungal cell walls, was used to estimate fungal biomass. Gas chromatography with electron capture detection (GC-ECD) and derivatization with hepta-fluorobutyric anhydride (HFBA) were chosen due to their sensitivity. Gas chromatography with mass spectroscopy was subsequently used to confirm these derivatization and detection methods. The extraction solvent used was acetonitrile-water (84:16). Wheat floret extracts were then cleaned through a charcoal alumina column. While previous work has shown GC-ECD to quantify HFBA-trichothecene derivatives, to our knowledge, no studies have used these protocols to also detect ergosterol. This method may be used to study FHB infection patterns within single wheat spikes and the extent of fungal colonization and toxin accumulation within single kernels with varying symptoms.

INFLUENCE OF INFECTION-TIMING ON FUSARIUM HEAD BLIGHT SEVERITY, WHEAT KERNEL DAMAGE AND DEOXYNIVALENOL ACCUMULATION DURING A 2007 FIELD STUDY.

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ABSTRACT

Previous field studies conducted in 2006 have shown that *Fusarium graminearum* infections during the grain-fill stages of wheat development may lead to kernels with low disease intensity yet significant levels (>2ppm) of deoxynivalenol (DON). Interestingly, infections during both flowering and grain-fill resulted in lower disease severity than when infections occurred during flowering alone. In the most susceptible cultivar a decrease in DON was also observed. The goal of the 2007 study was to gather additional data on infection-timing patterns. Three winter wheat cultivars were used in this field study: Hopewell (susceptible), Truman (moderately resistant) and Valor (moderately resistant). The experiment was a split-plot design with infection-timing treatment as the main effect and cultivar as the sub-plot. Four misting treatments were used to facilitate infection: ambient (no supplemental moisture), misting during flowering, misting during grain-fill and misting during flowering and grain-fill. Misting chambers and moveable greenhouses were used to supplement and prevent moisture respectively. All plots were spray inoculated with a mixture of four DON-producing *F. graminearum* isolates at anthesis and late milk stages. Misting treatments commenced immediately following inoculations and lasted four consecutive nights. Disease incidence and severity were measured in the field during dough stages. Following harvest, yield, kernel damage and DON accumulation were also assessed. Overall, disease intensity and DON levels were low, likely due to dry weather and low humidity during the growing season. In Hopewell, the amount of disease severity and percent kernel damage did not differ between ambient and misting during grain-fill treatments. However, the grain from the misting during grain-fill treatment contained significantly (*P* d•0.05) higher DON than that grown under ambient conditions. Also in Hopewell, disease severity and DON were significantly (*P* d•0.05) less under the misting during flowering and grain-fill treatment than in grain grown under the misting during flowering only treatment. This increased moisture – low disease and DON pattern warrants further study. Results from 2007 suggest late infections during grain-fill lead to grain with low disease intensity yet kernels with greater than 1ppm DON. Despite low disease pressure the data gathered in 2007 corroborates with overall infection-timing patterns observed in 2006.

CONTROL OF *FUSARIUM* INOCULUM PRODUCTION IN CORN RESIDUE BY MECHANICAL, BIOLOGICAL, AND CHEMICAL TREATMENTS.

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OBJECTIVES

I. Determine the effects of chopping of field corn residue on the saprophytic growth and sporulation of G*ibberella zeae* in the residue and the development of Fusarium head blight (FHB) in the following wheat crop.

II. Evaluate commercially available fungicides and biological agents as spring applications on the residue to disrupt the sporulation of *G. zeae* in the residue and reduce the development of FHB in wheat.

INTRODUCTION

Host resistance and fungicides individually can reduce head infection by *G. zeae* (=*Fusarium graminearum*) resulting in partial control reduction of FHB and deoxynivalenol (DON) formation. More complete control, however, will require the integration of these strategies with methods that reduce inoculum production in residue from previous crops. In this study, we examined mechanical, biological and chemical strategies to affect inoculum production. The mechanical strategy involved chopping of residue in the fall. By providing greater surface area for entry of saprophytic organisms and for contact with moisture and soil, chopping could hasten the decomposition of the residue and thus restrict growth of the pathogen. Some residue-colonizing microbes also might be antagonistic to the pathogen and, thus, might be effective in displacing the pathogen from the tissue or preventing its sporulation. The biological and chemical strategies involve application of biocontrol agents and fungicides, respectively, to the residue prior to flowering. Research with fungi, *Microsphaeropsis* sp. and *Trichoderma* sp., showed promise in using them to reduce pathogen growth and perithesia production in residue (Bujold and Paulitz, 2001; Fernandez, 1992; Gilbert and Fernando, 2004). Application of fungicides to residue has been investigated on a very limited basis. Tebuconazole impaired decomposition rate and eliminated *F. graminearum* in residues soaked in the fungicide (Yi et al. 2002), while captan applied to surface residue reduced numbers of fungi (including *Fusarium* spp.) and slowed residue decomposition (Beare et al., 1993). Thus, it appears that fungicides might directly inhibit the growth of the pathogen in residue but could also have a negative effect on colonization by competitive, decomposing microbes. In the limited studies on the treatment of residue with biological and chemical agents, fall applications were more effective in inhibiting pathogen growth than spring treatments. In this study, we examined whether spring treatments would exert sufficient impact on pathogen spore production in infested residue to reduce FHB and DON.

MATERIALS AND METHODS

Experiments were conducted in two University of Nebraska experiment station sites, ARDC near Mead and Haskell Agricultural Laboratory near Concord, to test the effects of fall mechanical treatments and spring biological and chemical treatments. Each experiment had a split-plot design with 'residue type' (chopped, unchopped, no residue) being the primary factor and 'spray treatment' (biocontrol products, fungicides, no treatment) the split factor. At each site, hard red winter wheat 'Overley' was block planted in fall 2006 into 4-acre fields having a previous soybean crop. Residue from BT corn in a neighboring field was chopped using a bush hog mower or left intact. The chopped and whole residue was left to decompose in place until early March, 2007. At that time, unchopped

corn debris was cut close to the ground with a sickle mower and residue of each type was collected and spread into 10' X 20' plots within the wheat field. Approximately 14 Kg (31 lb.) of residue was introduced per plot. There were three blocks containing one strip for each residue type. Each strip had six plots separated by 30'-wide buffer zones of wheat. Each plot within a strip was assigned one of six spray treatments which included three chemicals: Headline, Dithane DF, Prosaro; two biologicals: Serenade (*Bacillus subtilis* strain QST713) from AgraQuest, T-22 (*Trichoderma harzianum* strain KRL-AG2) produced by BioWorks; and a distilled water control. Spray treatments were applied to debris on the soil surface once at early stem extension stage (Feekes 6-8). Each material was applied at manufacturer's label rate for foliar applications in 20 gal water per acre using a CO_2-pressurized backpack sprayer with nozzles configured for spraying herbicides.

Residue samples were collected from plots containing chopped and unchopped residue plots at anthesis (Feekes 10.5) for determination of numbers of *G. zeae* ascospores. Samples were weighed and then washed in a standard amount of water with Tween. Ascospore concentration in each wash was determined by counting with a hemacytometer. Field data collected were FHB incidence and severity. DON content, yield of kernels, and percentage of *Fusarium* disease kernels (FDK) were measured after harvest. All data was subjected to ANOVA for split-plot design and Fisher's LSD was used for means separation.

RESULTS

There was a higher number of ascospores detected on chopped residue collected from Mead at anthesis than on whole residue (1.95×10^3 per g residue vs. 5.42×10^3 per g, respectively; $P = 0.012$). At Concord, there was no significant difference in ascospore numbers between residue types, with fewer than 2×10^3 per g being counted.

There was sufficient rain at both sites to cause moderate disease incidence, but disease severity was moderate at Mead and low at Concord. The effects of residue type and spring spray treatments on disease parameters differed between the two experiments. At Concord, there were significant spray treatment effects for FHB incidence and index, with Prosaro being the only treatment to have lower levels than the control (Table 1). Test weight was significantly higher in plots in which residue was sprayed with Headline as compared to the control. When data for each residue type was analyzed across spray treatments, the no-residue control had the lowest DON level and the highest yield measurements, but it also had the highest disease incidence. At Mead, there were significant residue by spray treatment interactions for disease incidence and index, but except for a decrease in disease index in chopped residue by Prosaro, none of the spray treatments reduced disease measurements in any residue types compared to the respective control (Table 2). Instead, Serenade and Headline increased disease incidence and index. Disease incidence, DON level and % FDK, averaged across spray treatments, were significantly higher in the whole residue and chopped residue plots than in the no-residue plots. In addition, presence of residue significantly decreased yield compared to no residue.

CONCLUSIONS

The results with Prosaro suggest there is some promise to the strategy of applying fungicides to residue in the spring to reduce FHB development in the wheat crop. The strategy by itself provided on low levels of disease control and DON reduction, so it would need to be integrated with host resistance and/or fungicide treatments applied to flowering heads. Given that Prosaro might be widely applied to flowering heads, it would be not be desirable to use the same product to treat residue as well because of increased the risks of selecting for resistant pathogen populations. Therefore, it is necessary to evaluate a larger selection of fungicides with different modes of action than Prosaro specifically for use as residue treatments. In this respect, biocontrol agents theoretically would be good candidates. While the results with the two biological products in this study were not encouraging, there are other commercial agents available for future testing.

The increased disease development and decreased yields observed in the presence of residue as com-

pared to no residue were agreement with other reports (Dill-Macky and Jones, 2000). While disease levels in chopped residue tended to be lower than in the whole residue, the differences largely were not significant. This could be related to chopping having only a small influence on sporulation, as suggested by the ascospore counts. Another explanation is that differences could have been masked by substantial aerial inoculum entering the experiment plots, as evidenced by the moderate levels of disease occurring in the no-residue plots. The wet weather experienced in eastern Nebraska during the experiments was atypical. In more typical drier years, the regional inoculum load might be lower and, thus, diminution of residue by chopping might exert a greater effect in reducing inoculum in a given field.

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DISCLAIMER

This is a cooperative project with the U.S. Wheat & Barley Scab Initiative. Any opinions, findings, conclusions, or recommendations expressed in this publication are those of the authors and do not necessarily reflect the view of the U.S. Department of Agriculture.

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Table 1. Results from 2007 residue management trial at Concord, Nebraska (Haskell Ag. Lab.)

Residue type	Spray treatment	Incid. (%)	Sev. (%)	Index (%)	DON (ppm)	FDK (%)	Yield (K)	200seed wt (g)
None	None	53	27	15	1.4	4.3	2.5	6.0
	Dithane DF	49	25	12	1.8	3.8	3.1	6.7
	Headline	46	22	10	1.8	2.7	3.0	6.5
	Prosaro	41	23	10	1.2	3.5	2.7	6.8
	Serenade	44	23	10	2.8	4.7	3.0	6.0
	T22	44	25	11	2.4	3.8	3.1	6.3
Whole	None	42	29	12	2.6	5.8	2.2	5.9
	Dithane DF	35	29	10	2.5	3.5	2.6	6.0
	Headline	41	24	10	2.8	4.7	2.5	6.4
	Prosaro	30	15	4	1.9	4.2	2.4	5.9
	Serenade	49	28	14	3.5	4.3	1.9	5.3
	T22	49	27	13	1.9	4.3	2.4	5.7
Chopped	None	47	28	14	2.4	4.5	2.2	6.0
	Dithane DF	33	26	9	1.8	4.3	2.2	6.0
	Headline	37	28	10	2.3	6.3	2.9	6.6
	Prosaro	33	26	8	1.4	3.3	3.1	6.3
	Serenade	43	30	13	2.3	5.2	2.5	6.0
	T22	36	21	8	2.2	5.7	1.6	5.3
P residue type X spray trt.		NS	NS	NS	NS	NS	NS	NS
None	Means across	46 a*	24	11	1.9 b	3.8	2.9 a	6.4 a
Whole	spray	41 ab	25	11	2.5 a	4.5	2.3 b	5.9 b
Chopped	treatments	38 b	26	10	2.1 ab	4.9	2.4 b	6.0 b
P residue type		0.013	NS	NS	0.050	NS	0.014	0.002
Mean	None	47 a	28	13 a	2.1 bc	4.9	2.3	6.0 bc
across	Dithane DF	39 ab	26	10 ab	2.0 bc	3.9	2.6	6.2 ab
residue	Headline	42 ab	25	10 ab	2.3 ab	4.6	2.8	6.5 a
types	Prosaro	35 b	21	7 b	1.5 c	3.7	2.7	6.3 ab
	Serenade	45 a	27	12 a	2.9 a	4.7	2.4	5.8 c
	T22	43 ab	24	11 ab	2.2 abc	4.6	2.4	5.8 c
P spray treatment		0.034	NS	0.050	0.018	NS	NS	0.001

* Letters denote means separation at P = 0.05.

Table 2. Results from 2007 residue management trial at Mead, Nebraska (ARDC).

Residue type	Spray treatment	Incid. (%)	Sev.(%)	Index (%)	DON (ppm)	FDK (%)	Yield (K)	200seed wt (g)
None	None	38 d*	29	11 e	5.3	11.0	2.7	6.3
	Dithane DF	50 bcd	53	26 abcde	5.4	9.0	3.1	6.8
	Headline	42 cd	39	17 cde	4.8	10.3	3.0	6.8
	Prosaro	42 cd	37	16 de	5.2	9.8	2.4	6.8
	Serenade	61 abc	62	38 abc	6.2	9.7	2.4	5.3
	T22	53 bcd	48	25 abcde	6.1	11.0	3.0	6.2
Whole	None	45 cd	35	16 cde	8.4	14.5	2.4	5.9
	Dithane DF	59 abc	57	34 abcd	8.0	13.8	1.8	6.4
	Headline	73 a	57	43 a	10.8	17.8	2.3	6.1
	Prosaro	52 bcd	44	25 abcde	10.0	14.0	1.6	5.9
	Serenade	67 ab	58	40 ab	9.7	11.5	1.9	5.8
	T22	53 bcd	43	23 abcde	12.0	14.3	2.4	5.8
Chopped	None	68 ab	59	40 ab	8.9	14.2	2.3	6.0
	Dithane DF	53 bcd	43	25 abcde	8.9	12.0	2.7	6.2
	Headline	49 bcd	40	20 bcde	7.7	14.1	2.5	6.2
	Prosaro	51 bcd	33	18 cde	7.0	12.5	2.6	6.4
	Serenade	58 abc	52	30 abcde	9.7	15.5	2.4	6.2
	T22	52 bcd	50	27 abcde	7.8	12.2	2.1	6.0
P residue type X spray trt.		0.024	NS	0.050	NS	NS	NS	NS
None	Means across	47 b	45	22	5.5 b	10.2 b	2.8 a	6.4
Whole	spray	58 a	49	30	9.8 a	14.3 a	2.1 c	6.0
Chopped	treatments	55 a	46	27	8.4 a	13.4 a	2.4 b	6.2
P residue type		0.008	NS	0.087	<0.001	0.006	<0.001	NS
Mean	None	50 b	42	23	7.5	13.2	2.5	6.0 ab
across	Dithane DF	54 ab	51	28	7.5	11.6	2.5	6.5 a
residue	Headline	55 ab	45	27	7.8	14.2	2.6	6.3 a
types	Prosaro	48 b	37	20	7.4	12.1	2.2	6.4 a
	Serenade	62 a	57	36	8.5	12.2	2.2	5.8 b
	T22	52 b	47	26	8.6	12.5	2.5	6.0 ab
P spray treatment		0.085	0.06	0.064	NS	NS	NS	0.039

* Letters denote means separation at P = 0.05.

EFFECTS OF SPRAY APPLICATION METHODS ON BIOCONTROL AGENT VIABILITY.

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OBJECTIVE

Determine the effects of commercial ground spray application systems on viability of representative biological control agents.

INTRODUCTION

All microorganisms used for biological control are sensitive to environmental extremes and thus, environmental conditions occurring during the application process could affect the viability of the agents and, consequently, affect disease control efficacy. Bacteria and yeast have been evaluated as biological agents in field trials for efficacy in controlling Fusarium head blight and reducing deoxynivalenol levels. Field tests conducted thus far utilized CO_2-pressurized backpack systems with care taken to avoid subjecting biological materials to temperature extremes. There is no information available as to how biological agents would respond to "real-world" conditions occurring during operation of commercial spray equipment. Under such conditions, accumulation of heat from sunlight or pump motors and sheer forces within the pumps, filters, nozzles and other systems mechanisms could possibly be injurious to biological agents.

MATERIALS AND METHODS

Three microorganisms were tested, each representing a different microorganism group: *Bacillus* sp. 1BA (Draper et al., 2001) representing spore-forming, gram-positive bacteria; *Lysobacter enzymogenes* C3 (Jochum and Yuen, 2006) representing gram-negative, non-spore-forming bacteria; and *Cryptococcus aureus* OH71-4 (Khan et al., 2004) representing epiphytic yeasts. Spontaneous mutants of the bacterial strains resistant to the drug rifampicin were used so that they could be detected on 10% tryptic soy agar (TSA) medium amended with the drug and a fungicide. The yeast OH71-4 was detected on 10% TSA amended with antibacterial drugs. Quantification of the agents in cell suspensions was done by dilution plating. Cultures of bacterial strains C3 and 1BA in chitin broth and nutrient broth, respectively, and yeast strain OH71-4 in a proprietary frozen concentrate (provided by D. Schisler, NCAUR) were used in two experiments. One hypothesis tested in the experiments was that heat accumulation in the biocontrol agent suspensions will reduce organism viability. Another hypothesis was that each stage of a spray application (i.e., agitation of cell suspension in tank, pumping of cell suspensions through the spray line, and discharge of cell suspension as droplets through nozzles) also will affect biocontrol agent viability.

Experiment 1 was conducted at North Dakota State University research facilities at Langdon using a customized spray system with a 10-gal. tank, shortened lines, and a single nozzle. These changes were made to accommodate small liquid volumes. In addition, ports were added to the line between the pump and the filter and between the filter and nozzle to allow collection of liquid samples. Otherwise all parts were standard as would be used on conventional spray systems, including a cast iron gear drive centrifugal pump (HYPRO Model 9006C-O) and XR8002 nozzle. The

bacterial cultures and yeast formulation were diluted with water in the tank to 4 gallons. The pump was operated at standard PTO speed (540 rpm) and manifold pressure was maintained at 40 psi. The contents of the tank were continuously agitated by recirculation (8 gal/min). At 10-minute intervals, temperature within the tank liquid was measured, and samples of liquid were collected from the tank and the two sampling ports. Liquid was then emitted from the nozzle and a sample of the spray collected.

Experiment 2 was conducted at the UNL Agronomy Research Farm using an unmodified commercial spray system having a piston pump (Ace model F-1), a 115-gal capacity tank, and the same nozzle type as in experiment 1. The biological materials were diluted to 50 gal with tap water. The suspensions were sprayed continuously out three nozzles under 40 psi pressure. As in experiment 1, temperature and tank liquid samples were collected at 10-minute intervals. Spray samples were collected from each of the nozzles as well.

RESULTS AND CONCLUSIONS

During experiment 1, temperatures in each of the biocontrol suspensions re-circulating in the tank rose rapidly due to transfer of heat from the pump (Fig. 1). Within 30-40 minutes, temperatures exceeded 50^{o}C, which is injurious or lethal to most microorganisms. The heat accumulation was likely the primary factor leading to loss of viability of the biocontrol agents in the liquid (Fig. 2). In this respect, the three organisms exhibited widely different responses to similar rises in temperature. There was little difference in numbers of live cells between samples taken at various points in the spray system and samples collected from the tank (Fig. 3), indicating that passage of the cell suspensions through individual parts (pump, filter, and nozzle) had negligible effects on organism viability and did not account for the drops in population within the tank. When the biocontrol agents were sprayed through commercial equipment in experiment 2, temperatures in the liquid remained stable at favorable levels (Fig. 4). Because ambient temperatures and sunlight conditions were similar between experiments, the lower liquid temperatures recorded in experiment 2 can be attributed to the large liquid volume accumulating heat from the pump at a much slower rate. Consequently, there were no appreciable changes in biocontrol populations within the tank over time (Fig. 5). In addition, biocontrol agent population levels sprayed out of the nozzles were not significantly different from those measured in the tank (Fig. 6), thus confirming that the various mechanisms in a conventional spray system collectively have little effect on biocontrol agent viability. We conclude that when biocontrol agents eventually become available for application to cereals, they will be compatible in the most part with existing equipment used for ground applications of fungicides. Heat accumulation in the liquid may reduce biocontrol agent numbers, but this would most likely occur as tanks are close to being empty.

In experiment 1, the high recirculation rate relative to the small liquid volume resulted in foaming of the liquids in the tank. Foaming was particularly a problem with the culture of *Bacillus* 1BA, resulting in difficulties in maintaining stable manifold pressure. While it is unknown if foaming would affect the viability of the organisms in the liquid, foaming could hasten the degradation of antifungal proteins and antibiotics excreted into broth culture by the bacteria and, thus, could potentially reduce biocontrol effectiveness. In experiment 2, when the biocontrol materials were diluted into much larger water volumes, foaming was not a problem for the bacterium C3 and the yeast. Addition of the an antifoaming agent Biospumex 36K to the suspension of 1BA arrested foaming. Therefore, the use a nontoxic antifoaming agent is recommended as an adjuvant for those biocontrol materials for which foaming may be an issue.

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DISCLAIMER

Any opinions, findings, conclusions, or recommendations expressed in this publication are those of the authors and do not necessarily reflect the view of the U.S. Department of Agriculture.

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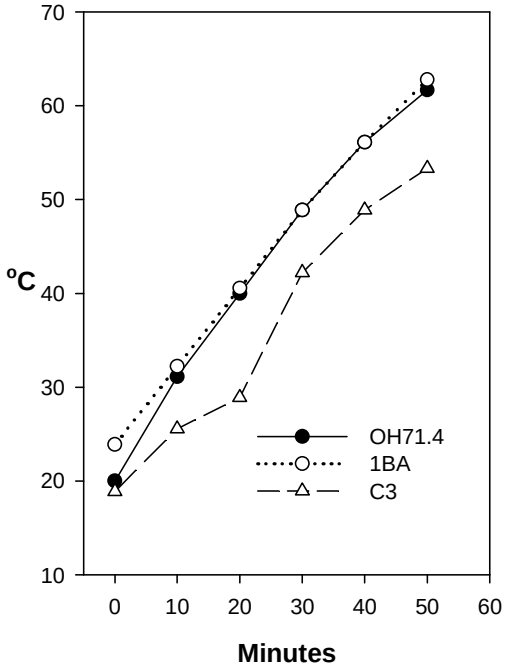

Fig. 1 Expt 1 - Temperatures in biocontrol agent suspensions in tank.

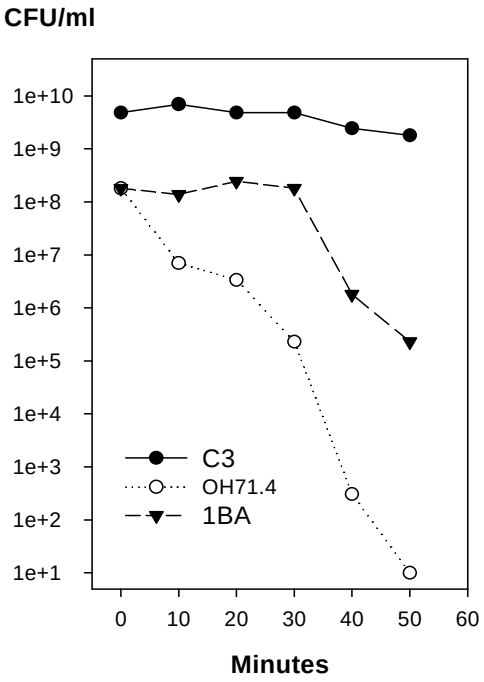

Fig. 2 Expt 1 - Biocontrol agent populations in suspensions in tank.

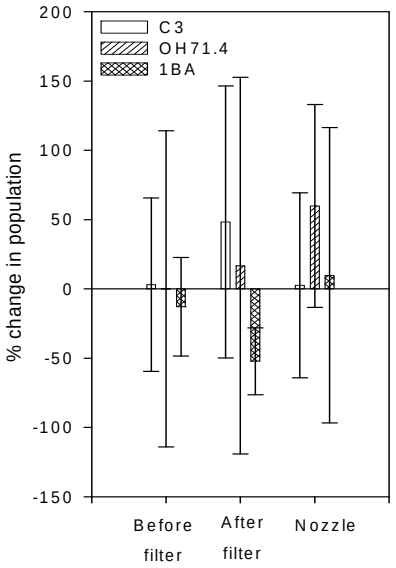

Fig. 3 Expt 1 – Changes in biocontrol agent populations at various points in spray system relative to tank populations. Standard deviations shown.

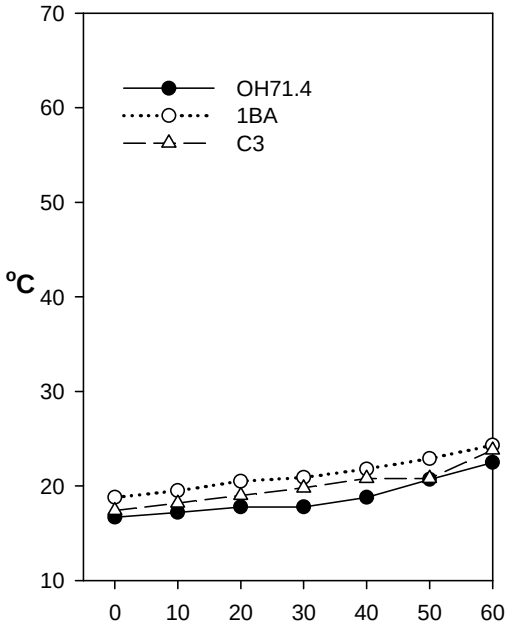

Fig. 4 Expt 2 - Temperatures in biocontrol agent suspensions in tank.

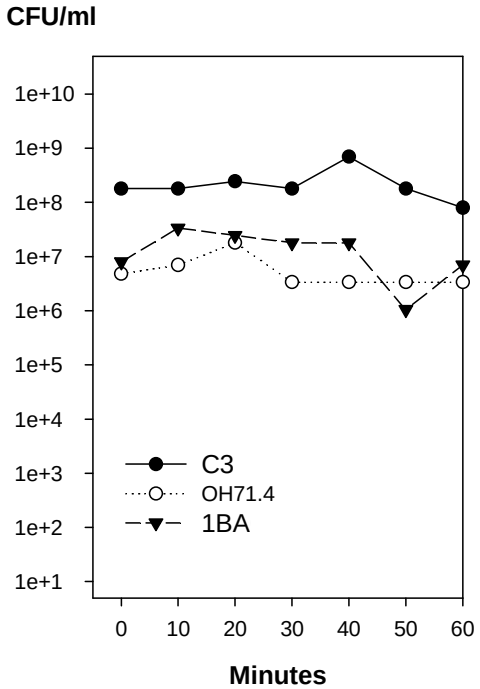

Fig. 5 Expt 2 - Biocontrol agent populations in suspensions in tank.

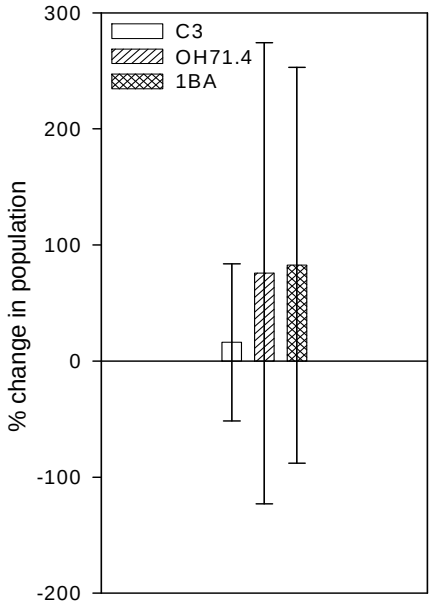

Fig. 6 Expt 2 – Changes in biocontrol agent populations at nozzles relative to tank populations. Standard deviations shown.

RESULTS FROM THE 2007 STANDARDIZED EVALUATION OF BIOLOGICAL AGENTS FOR THE CONTROL OF FUSARIUM HEAD BLIGHT ON WHEAT AND BARLEY.

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OBJECTIVE

To evaluate, using standardized methodology, a set of biological control agents applied alone and in combination with a fungicide for effectiveness in managing Fusarium head blight (FHB) and deoxynivalenol (DON) in wheat and barley across a range of environmental conditions.

INTRODUCTION

Some of the biological agents reported to have potential for controlling FHB are bacterial strains *Bacillus subtilis* TrigoCor 1448 (Stockwell et al., 2001) and *Bacillus* sp. 1BA (Draper et al., 2001), and *Lysobacter enzymogenes* strain C3 (Jochum et al., 2006). Each strain has shown efficacy in some field tests when evaluated separately (Stockwell et al., 2001; Jochum et al., 2006; Yuen and Jochum, 2004). In 2004 through 2006, they were directly compared for efficacy as part of the USWBSI-funded program for standardized evaluation of biological agents. Because combinations of biological control agents and fungicides were reported to be more effective in controlling FHB than the microorganisms or fungicides alone (DaLuz et al., 2003; Khan et al., 2004; Yuen and Jochum, 2004), standardized evaluations in 2005 and 2006, also compared these bacterial strains in combination with the fungicide tebuconazole. In the three years of testing, however, results were inconclusive as to the effectiveness of the treatments across a range of environmental conditions and crop genotypes (Yuen et al, 2004; Yuen et al., 2005; Yuen et al., 2006) due to low disease pressure in most or all test sites. Uniform fungicide trials in 2006 (Paul et al., 2006) showed increased yield and reduction of DON by a fungicide formulation Prosaro 421 SC that combines tebuconazole and prothioconazole. Therefore, trials in 2007 were designed to test the efficacy of the three biological agents, alone in combination with Prosaro, for the control of FHB and DON.

MATERIALS AND METHODS

Six trials were conducted across three states on barley and a range of wheat market classes (Table 1). In each trial, three bacterial biological agents (Table 2) were tested alone or in tank mix with the fungicide Prosaro 421 SC (6.5 fl oz/A). There also was a treatment of Prosaro alone and a non-treated control. A broth culture of each organism was provided by the originating laboratory and sent to the researcher in each location. The pre-application population of each agent in the inoculum was determined by the local researcher using dilution plating. All treatment liquids were amended with 0.125% Induce (v/v). One application was made per treatment at early flowering (Feekes 10.51) in 20 gal/acre using CO2-pressurized sprayers (approximately 40 psi) equipped with flat-fan nozzles oriented forward and backward. The size and number of replicate plots varied among trials. Some of the trials were inoculated with *Fusarium graminearum* spore suspensions and or inoculated corn grain, with mist irrigation systems utilized to stimulate infection. In all trials, FHB incidence (% heads infected per plot), severity (% spikelets infected per diseased head), and index (% plot severity) were determined from at least 40 heads per plot around 3

weeks after anthesis. The incidence of *Fusarium*-damaged kernels (%FDK), as well as yield of seed and test weight, were determined after harvest. Samples from each plot were sent to the North Dakota State University Veterinary Diagnostic Laboratory, Fargo, ND for analysis of DON content. Analysis of variance was performed on results from each trial separately, with Duncan's multiple range test used for means separation. Data from Missouri and Nebraska trials were analyzed together using ProcMixed (SAS), with trials being treated as fixed variables and the LSD method used to separate LS means. Results from South Dakota were excluded because of low disease levels.

RESULTS AND DISCUSSION

Weather conditions in South Dakota were dry, preventing significant disease or DON production. Wet weather in Missouri and Nebraska resulted in higher FHB development. Low temperatures in Missouri occurring during heading, however, caused damage to wheat heads and kernels, particularly in cv. Elkhart, making assessments of FHB severity and FDK difficult.

The most effective biological agent applied alone was *L. enzymogenes* C3, reducing disease severity in two trials and disease index averaged across four trials (Tables 3A and 3B). No combination of a biocontrol agent with Prosaro 421 SC exhibited greater efficacy than the fungicide alone. None of the biocontrol agents alone increased yields over the control (data not shown). Prosaro 421 SC applied alone or in combination with a biological agent was effective in reducing FHB measures and DON levels in multiple trials (Tables 3A and 3B). The fungicide alone and in combinations with some biocontrol agents increase plot yields in the two Nebraska trials and in the Missouri with 'Roane' (data not shown).

The collective results from this year's multistate trials indicated *L. enzymogenes* C3 to the be the most effective biocontrol agent across a range of environments, but treatments with the bacterium are not as effective or as consistent as treatment with Prosaro 421 SC. No benefit was revealed in this study from combining biocontrol agents with Prosaro 421 SC, contrary to past studies with biocontrol agent-tebuconazole combinations. The difference in results is most likely related to the greater effectiveness of Prosaro 421 SC over tebuconazole. Therefore, it may be desirable to explore combinations of biocontrol agents with less efficacious fungicides as a means to broaden the selection of treatments that can be used to protect florets from *Fusarium* infection.

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DISCLAIMER

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Table 1. 2007 uniform biological control trial locations, crop cultivars, and researchers

State (location)	Crop market class and cultivar	Researcher and Institution
MO	Soft red winter wheat 'Roane'	L. Sweets, University of Missouri
MO	Soft red winter wheat 'Elkhart'	L. Sweets, University of Missouri
NE -1 (Mead)	Hard red winter wheat '2137'	C. Jochum & G. Yuen, University of Nebraska
NE - 2 (Lincoln)	Hard red winter wheat '2137'	C. Jochum & G. Yuen, University of Nebraska
SD	Hard red spring wheat 'Briggs'	K. Ruden & B. Bleakley, South Dakota St. Univ.
SD	Six-rowed barley 'Robust'	K. Ruden & B. Bleakley, South Dakota St. Univ.

Table 2. Biological control agents tested in 2007 uniform trials.

Organism	Supplier
Bacillus sp.1BA	Bruce Bleakley, South Dakota State University
Bacillus subtilis TrigoCor 1448	Gary Bergstrom, Cornell University
Lysobacter enzymogenes C3	Gary Yuen, University of Nebraska

Table 3A. 2007 results across six uniform biocontrol trials denoted by state and crop

Treatment		MO 'Roane'	MO 'Elkhart'	NE-1 2137	NE-2 2137	SD Briggs	SD Barley	LS mean
INCIDENCE (% heads infected)								
Control		12 a*	15 c	95 a	68 a	2	57	59 a
Prosaro		7 cd	22 abc	58 b	47 c	1	42	40 b
1BA		9 bc	18 bc	87 a	70 a	3	62	58 a
1BA + Prosaro		6 d	24 abc	65 b	52 bc	2	65	44 b
TrigoCor 1448		10 ab	22 abc	92 a	69 a	1	90	60 a
TrigoCor 1448 + Prosaro		7 cd	30 a	62 b	47 c	1	50	43 b
C3		12 a	26 ab	84 a	62 ab	1	79	56 a
C3 + Prosaro		7 cd	16 c	70 b	58 abc	1	84	46 b
	P	<0.001	0.045	<0.001	<0.001	0.758	0.174	<0.0001
SEVERITY (% spikelets infected)								
Control		16 a	32	38.0 a	29 b	45	5	31 a
Prosaro		6 c	32	15.6 e	28 b	17	5	21 cd
1BA		12 ab	23	33.0 ab	28 b	13	5	27 bc
1BA + Prosaro		7 bc	36	20.0 de	28 b	12	5	23 bcd
TrigoCor 1448		11 ab	26	35.8 a	28 b	38	7	28 ab
TrigoCor 1448 + Prosaro		9 bc	34	22.1 cde	36 a	22	5	26 abc
C3		10 bc	28	28.1 bc	28 b	27	6	25 abcd
C3 + Prosaro		9 bc	19	24.7 cd	26 b	5	10	20 d
	P	0.017	0.175	<0.001	0.039	0.755	.0545	<0.0001

*Means separation (P=0.05) shown only when treatment effect was significant.

Table 3B. 2007 results across six uniform biocontrol trials denoted by state and crop.

Treatment		MO 'Roane'	MO 'Elkhart'	NE - 1 2137	NE - 2 2137	SD Wheat	SD Barley	LS Mean
INDEX (plot severity)								
Control		1.9 a	4	36 a	20 a	1.1	3 b	20 a
Prosaro		0.4 c	8	9 e	13 c	0.7	2 b	9 d
1BA		1.1 b	4	29 ab	20 a	0.6	3 b	18 ab
1BA + Prosaro		0.4 c	9	14 de	14 bc	0.3	3 b	11 d
TrigoCor 1448		1.2 b	7	33 a	19 ab	0.8	6 ab	19 ab
TrigoCor 1448 + Prosaro		0.6 bc	10	15 de	17 abc	0.3	2 b	12 cd
C3		1.1 b	8	24 bc	17 abc	0.5	5 ab	15 bc
C3 + Prosaro		0.7 bc	4	18 cd	15 bc	0.2	8 a	11 d
	P	0.001	0.133	<0.001	0.039	0.894	0.029	<0.0001
FDK (%)								
Control		10	16	18	15	1.2	ND#	10
Prosaro		7	20	10	9	0.5	ND	7
1BA		11	22	16	15	0.8	ND	11
1BA + Prosaro		9	18	13	12	0.5	ND	9
TrigoCor 1448		12	19	12	12	1.0	ND	10
TrigoCor 1448 + Prosaro		11	23	11	11	0.8	ND	10
C3		12	19	15	13	0.8	ND	10
C3 + Prosaro		5	17	14	14	0.8	ND	9
	P	0.164	0.232	0.150	0.130	0.517	---	0.121
DON (ppm)								
Control		<0.5	0.8 c	5.4 a	3.6 a	<0.5	<0.5	3.7 a†
Prosaro		<0.5	1.4 ab	2.6 c	2.3 b	<0.5	<0.5	2.2 b
1BA		<0.5	1.1 bc	4.9 a	3.4 a	<0.5	<0.5	3.5 a
1BA + Prosaro		<0.5	1.2 abc	2.9 c	2.7 ab	<0.5	<0.5	2.3 b
TrigoCor 1448		<0.5	1.0 bc	4.6 a	3.3 a	<0.5	<0.5	3.4 a
TrigoCor 1448 + Prosaro		<0.5	1.6 a	2.6 c	2.1 b	<0.5	<0.5	2.2 b
C3		<0.5	1.3 ab	4.2 ab	3.4 a	<0.5	<0.5	3.3 a
C3 + Prosaro		<0.5	1.0 bc	2.9 bc	2.3 b	<0.5	<0.5	2.2 b
	P	---	0.027	<0.001	0.003	---	---	<0.0001

*Means separation (P=0.05) shown only when treatment effect was significant.
#ND=no data.
†Data from MO 'Elkhart' and NE trials were used to calculate LS means for DON.