Efficacy and Stability of Integrating Fungicide and Cultivar Resistance to Manage Fusarium Head Blight and Deoxynivalenol in Wheat

K. T. Willyerd, C. Li, and **L. V. Madden,** Department of Plant Pathology, The Ohio State University, Ohio Agricultural Research and Development Center, Wooster 44691; **C. A. Bradley,** Department of Crop Sciences, University of Illinois, Urbana 61801; **G. C. Bergstrom,** Department of Plant Pathology and Plant-Microbe Biology, Cornell University, Ithaca, NY 14853; **L. E. Sweets,** Division of Plant Sciences, University of Missouri, Columbia 65211; **M. McMullen,** Department of Plant Pathology, and **J. K. Ransom,** Department of Plant Sciences, North Dakota State University, Fargo 58108; **A. Grybauskas,** Department of Plant Science and Landscape Management, University of Maryland, College Park 20742; **L. Osborne,** Pioneer Hi-Bred International, Brookings SD 57006; **S. N. Wegulo,** Department of Plant Pathology, University of Nebraska-Lincoln, Lincoln 68583; **D. E. Hershman,** Department of Plant Pathology, The University of Kentucky, Princeton, 42445; **K. Wise,** Department of Botany and Plant Pathology, Purdue University, West Lafayette, IN 47907; **W. W. Bockus,** Department of Plant Pathology, Kansas State University, Manhattan 66506; **D. Groth,** Louisiana State University Agricultural Center Rice Research Station, Rayne 70578; **R. Dill-Macky,** Department of Plant Pathology, University of Minnesota, St. Paul 55108; **E. Milus,** Department of Plant Pathology, University of Arkansas, Fayetteville 72701; **P. D. Esker,** Department of Plant Pathology, University of Wisconsin-Madison, Madison 53706; **K. D. Waxman,** Department of Plant Pathology and Plant-Microbe Biology, Cornell University; **E. A. Adee** and **S. E. Ebelhar,** Department of Crop Sciences, University of Illinois; **B. G. Young,** Department of Plant, Soil, and Agricultural Systems, Southern Illinois University, Carbondale 62901; and **P. A. Paul,** Department of Plant Pathology, The Ohio State University, Ohio Agricultural Research and Development Center

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Abstract

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Integration of host resistance and prothioconazole + tebuconazole fungicide application at anthesis to manage Fusarium head blight (FHB) and deoxynivalenol (DON) in wheat was evaluated using data from over 40 trials in 12 U.S. states. Means of FHB index (index) and DON from up to six resistance class–fungicide management combinations per trial (susceptible treated [S_TR] and untreated [S_UT]; moderately susceptible treated [MS_TR] and untreated [MS_UT]; moderately resistant treated [MR_TR] and untreated [MR_UT]) were used in multivariate meta-analyses, and mean log response ratios across trials were estimated and transformed to estimate mean percent control ($\overline{C}$) due to the management combinations relative to S_UT. All combinations led to a significant reduction in index and DON ($P <$ 0.001). MR_TR was the most effective combination, with a $\overline{C}$ of 76% for index and 71% for DON, followed by MS_TR (71 and 58%, respectively), MR_UT (54 and 51%, respectively), S_TR (53 and 39%, respectively), and MS_UT (43 and 30%, respectively). Calculations based on the principle of treatment independence showed that the combination of fungicide application and resistance was additive in terms of percent control for index and DON. Management combinations were ranked based on percent control relative to S_UT within each trial, and nonparametric analyses were performed to determine management combination stability across environments (trials) using the Kendall coefficient of concordance (W). There was a significant concordance of management combinations for both index and DON ($P < 0.001$), indicating a nonrandom ranking across environments and relatively low variability in the within-environment ranking of management combinations. MR_TR had the highest mean rank (best control relative to S_UT) and was one of the most stable management combinations across environments, with low rank stability variance (0.99 for index and 0.67 for DON). MS_UT had the lowest mean rank (poorest control) but was also one of the most stable management combinations. Based on Piepho's nonparametric rank-based variance homogeneity U test, there was an interaction of management combination and environment for index ($P = 0.011$) but not for DON ($P = 0.147$), indicating that the rank ordering for index depended somewhat on environment. In conclusion, although the magnitude of percent control will likely vary among environments, integrating a single tebuconazole + prothioconazole application at anthesis with cultivar resistance will be a more effective and stable management practice for both index and DON than either approach used alone.

Fusarium head blight (FHB) of small grain crops is primarily caused by the fungal pathogen *Fusarium graminearum* Schwabe in the United States (4,26). This pathogen produces mycotoxins, including deoxynivalenol (DON), which contaminate kernels during infection, persist in storage, and are potentially harmful to humans and animals if consumed (32,33). FHB symptoms are generally positively correlated with DON accumulation (30). Both disease development and toxin accumulation are strongly influenced by environmental conditions such as rainfall, relative humidity, and temperature before, during, and after anthesis of the host crop (3,12). Together, FHB and DON reduce grain yield, quality, safety, and marketability, leading to economic losses in every sector of the small grain industry (21).

Several options for minimizing losses due to FHB and DON are available, including cultural practices, host resistance, and fungicides. *F. graminearum* overwinters on crop residues from small grain crops, corn (*Zea mays*), and other grassy weeds, as well as soybean stubble (6,15,26). Rotating these crops with a non-host or burying crop residue through tillage limits in-field sources of primary inoculum, thereby reducing *F. graminearum* infection and DON contamination (1). However, *F. graminearum* spores have been found to travel great distances by air, leading to FHB de-

Corresponding author: P. A. Paul, E-mail: paul.661@osu.edu

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velopment in fields that have been rotated with non-host crops or tilled to bury host residues (20,37). Cultivars with moderate resistance to FHB are commercially available in most hexaploid wheat market classes but none of these are completely resistant to FHB. Host resistance in the *F. graminearum*–wheat system is a very complex trait, with several different types reported (resistance to initial infection, spread, and DON accumulation; 2,38). Although these types have been shown to be interrelated (39), resistance to FHB does not always parallel resistance to DON accumulation (16). Even the most resistant wheat cultivars may sustain economically significant levels of DON contamination (>2 ppm), with or without visual symptoms of FHB (29). Moreover, cultivar reaction to FHB and DON is strongly influenced by environmental conditions at the time of anthesis and early grain fill (24), leading to situations in which cultivars with similar resistance classifications show considerably different disease and toxin levels within and across environments (44). Fungicides with demethylation inhibitor chemistry have been shown to be the most effective chemical control strategy for reducing FHB and DON, when applied at early anthesis (8,28). However, even the most effective fungicides provide, on average, only 47 to 56% and 39 to 50% control of FHB index and DON, respectively, with efficacy being influenced by environment, background disease and DON levels, and wheat class (28).

Integrated management of FHB is often recommended as the most effective strategy for minimizing losses due to FHB and DON (22). In general, combining an appropriately timed fungicide application with a moderately resistant cultivar usually results in the greatest reductions in disease intensity and toxin accumulation (46). However, the reported efficacy of this approach in terms of percent reduction in FHB and DON has varied among individual integrated management trials (9,45,46). In addition, results from some trials have shown that, under certain conditions, cultivar resistance or fungicide treatment alone may provide levels of FHB and DON reduction comparable with that of the resistance–fungicide management combination (44,46). Such variability is likely due to differences in environmental conditions (including weather and inoculum density) among trials, which impact disease intensity, toxin accumulation, fungicide efficacy, and cultivar reaction.

Year-to-year or location-to-location variations among studies conducted using the same cultivar–fungicide combinations provide circumstantial evidence that fungicide–environment and genotype–environment interactions may affect the magnitude of disease and DON reductions or the relative effect of different combinations of cultivar resistance and fungicide treatment (1,22,43–46). Understanding the efficacy and stability of integrating cultivar resistance and fungicide treatment across environments is vital for making general best-management recommendations to growers. Uniform integrated management trials have been conducted in several U.S. small grain-growing regions from 2007 to 2010. Synthesizing the data from these trials will enable us to understand the value of combining management strategies in terms of overall magnitude and stability of FHB and DON reduction. Meta-analysis has been demonstrated as a useful approach for providing such a synthesis of plant disease responses to management strategies across trials (25,28,31). The consistency of treatment or genotype effects across trials also can be assessed using various nonparametric approaches for stability analysis (19,34). These methods directly consider the rank order of the effects among trials to determine the stability of the relationships.

The objectives of this study were to assess the efficacy and stability of host resistance–fungicide management combinations against FHB and DON in several hexaploid wheat classes. Using data from field experiments conducted from 2007 to 2010, multivariate random-effects meta-analyses were conducted to evaluate the efficacy of integrating cultivar resistance and fungicide application on FHB index and DON. In addition, a nonparametric analysis of concordance of ranking was performed to determine the stability of this integrated strategy across different locations, years, and wheat classes.

Materials and Methods

Uniform integrated management trials. Field trials, coordinated by researchers collaborating as part of the United States Wheat and Barley Scab Initiative (USWBSI), were conducted from 2007 to 2010 in spring and winter wheat regions of the United States. Data were collected from a total of 12 states, with multiple trials in some states, representing four wheat market classes (Table 1). The experimental design was a randomized complete block with a split-plot arrangement of fungicide treatment and cultivar in three to six replicate blocks. Most trials used fungicide as the whole plot and cultivar as the subplot; however, some locations used the opposite arrangement. A mixed-model analysis (17) took into account the appropriate whole and subplot structure of individual trials. Additionally, at some locations, similar trials were established in fields with and without host crop residue, representing different crop rotation sequences as a third factor. In some cases, when residue was included in the trial, it was the highest level in the experimental layout (e.g., whole plot), with the other two factors then comprising the subplot and sub-subplot factors.

All trials were conducted with at least two fungicide treatments: an untreated check and the fungicide tebuconazole + prothioconazole applied at anthesis (Feekes 10.5.1) (13). The fungicide was available as either a premixed product under the trade name Prosaro 421 SC (19% tebuconazole + 19% prothioconazole; Bayer CropScience, Research Triangle Park, NC) or a tank mix of Folicur 3.6 F (38.7% tebuconazole; Bayer CropScience) and Proline 480 SC (41% prothioconazole; Bayer CropScience). Prosaro was applied at 475 ml/ha, whereas the Proline + Folicur tank mix was applied as each product at 219 ml/ha at spray volumes between 94 and 187 liters/ha. A non-ionic surfactant was added to each treatment at a rate of 0.125% (vol/vol) 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 trial, between 2 and 20 locally adapted wheat cultivars were planted and coded as susceptible, moderately susceptible, or moderately resistant. Some trials had multiple cultivars in each category; however, there was at least one moderately resistant and one susceptible cultivar in nearly all trials (Table 1). Trials with soft white winter wheat were the exception, having only susceptible and moderately susceptible cultivars. Resistance categories were used instead of the individual cultivars in the analyses outlined below. In nine trials, artificial inoculation of all plots was included as part of the protocol. In eight of these trials, plots were spray inoculated with a macroconidial or ascospore suspension (5,11) during anthesis of each cultivar, approximately 24 h after the fungicide treatment was applied; in the remaining inoculated trial, *F. graminearum*-colonized corn kernels were spread in the plots followed by mist irrigation to enhance inoculum production and infection. All other trials were conducted under natural infection. FHB index (index = mean percentage of diseased area per spike, also known as plot severity; 40) was assessed approximately 3 weeks after anthesis on 20 to 100 spikes/plot. Following harvest, grain samples were ground in appropriate laboratory mills until they resembled whole wheat flour, an adequate particle size for DON extraction and analysis. DON analysis was performed using gas chromatography with mass spectrometry or electron capture detection, based on the method described by Tacke and Casper (41), at one of the USWBSI DON-testing laboratories at North Dakota State University, University of Minnesota, or Virginia Polytechnic Institute and State University.

Quantitative synthesis of resistance–fungicide effects on FHB and DON. The focus of this analysis was the interaction between fungicide treatment and cultivar resistance. The main purpose of the integrated management trials was to determine the combined effects of fungicide and cultivar resistance on FHB and DON. For those trials with cropping sequence as the whole-plot factor, each cropping sequence was treated as a separate trial. This is justified by the fact that the other factors were randomized within each cropping sequence, and each sequence can be considered a type of environment. There were 53 trials, including 3 hard red spring, 8 hard red winter, 38 soft red winter, and 4 soft white winter wheat trials (Table 1).

As the first stage in the analysis, each resistance class–fungicide combination was given a different numerical code, representing six unique management combinations: 1 = susceptible, untreated (S_UT [check]); 2 = moderately susceptible, untreated (MS_UT);

Table 1. Experiments from uniform integrated management trials across spring and winter wheat-growing regions of the United States performed from 2007 to 2010 to evaluate the effects of cultivar resistance and fungicide treatment on Fusarium head blight (FHB) index and deoxynivalenol (DON) in wheat

						Number of cultivars[a]			Susceptible, untreated check[b]	
Code[c]	**Wheat class[d]**	**Location[e]**	**Year**	**Previous crop[f]**	**Inoculated[g]**	**S**	**MR**	**MS**	**Index >2%**	**DON >1 ppm**
1	HRSW	Brookings, SD	2009	Host	No	1	1	2	Yes	Yes
2	HRSW	Fargo, ND	2010	Host	Yes	2	1	1	Yes	Yes
3	HRSW	Lisbon, ND	2007	Non-host	No	1	3	15	Yes	No
4	HRWW	Brookings, SD	2009	Host	No	1	1	1	Yes	Yes
5	HRWW	Brookings, SD	2010	Host	No	1	1	1	No	Yes
6	HRWW	Forman, ND	2010	Host	No	12	4	2	Yes	Yes
7	HRWW	Lisbon, ND	2007	Host	No	2	–	18	Yes	Yes
8	HRWW	Manhattan, KS	2007	Host	Yes	1	1	1	No	Yes
9	HRWW	Meade, NE	2007	Non-host	Yes	1	1	1	Yes	No
10	HRWW	Meade, NE	2008	Host	No	1	1	1	Yes	Yes
11	HRWW	Prosper, ND	2010	Host	No	12	4	2	Yes	No
12	SRWW	Aurora, NY	2010	Non-host	Yes	1	1	–	Yes	Yes
13	SRWW	Aurora, NY	2010	Non-host	Yes	1	1	–	No	Yes
14	SRWW	Aurora, NY	2009	Host	No	1	1	–	Yes	Yes
15	SRWW	Aurora, NY	2009	Non-host	No	1	1	–	Yes	Yes
16	SRWW	Beltsville, MD	2009	Host	No	1	2	3	Yes	Yes
17	SRWW	Beltsville, MD	2009	Non-host	No	1	2	3	Yes	Yes
18	SRWW	Columbia, MO	2007	Host	No	1	1	3	Yes	Yes
19	SRWW	Columbia, MO	2007	Non-host	No	1	1	3	Yes	Yes
20	SRWW	Columbia, MO	2008	Host	No	1	1	3	Yes	Yes
21	SRWW	Columbia, MO	2008	Non-host	No	1	1	3	Yes	Yes
22	SRWW	Columbia, MO	2009	Host	No	2	1	2	Yes	Yes
23	SRWW	Columbia, MO	2009	Non-host	No	2	1	2	Yes	Yes
24	SRWW	Columbia, MO	2010	Non-host	No	2	1	2	Yes	Yes
25	SRWW	Carbondale, IL	2009	Host	No	2	2	2	Yes	Yes
26	SRWW	Carbondale, IL	2009	Non-host	No	2	2	2	Yes	Yes
27	SRWW	Carbondale, IL	2010	…	No	2	2	1	Yes	Yes
28	SRWW	Crowley, LA	2008	…	No	2	2	2	Yes	No
29	SRWW	Dixon Springs, IL	2009	Host	No	1	2	5	Yes	No
30	SRWW	Dixon Springs, IL	2009	Non-host	No	1	2	5	Yes	No
31	SRWW	Dixon Springs, IL	2010	Host	No	3	3	2	Yes	Yes
32	SRWW	Monmouth, IL	2009	Host	No	1	2	6	No	Yes
33	SRWW	Monmouth, IL	2009	Non-host	No	1	2	6	No	Yes
34	SRWW	Princeton, KY	2009	Host	No	1	1	1	Yes	Yes
35	SRWW	Princeton, KY	2010	Host	No	1	2	–	Yes	No
36	SRWW	Urbana, IL	2009	Host	No	1	2	3	Yes	Yes
37	SRWW	Urbana, IL	2009	Host	No	1	2	3	Yes	Yes
38	SRWW	Urbana, IL	2009	Non-host	No	1	2	3	Yes	Yes
39	SRWW	Urbana, IL	2010	Host	No	2	3	1	Yes	Yes
40	SRWW	Urbana, IL	2010	Host	No	2	3	1	Yes	Yes
41	SRWW	Urbana, IL	2010	Non-host	No	2	3	1	Yes	Yes
42	SRWW	West Lafayette, IN	2010	Host	No	2	2	2	Yes	Yes
43	SRWW	Wooster, OH	2008	Host	Yes	2	1	3	Yes	Yes
44	SRWW	Wooster, OH	2009	Host	Yes	2	1	3	Yes	Yes
45	SRWW	Queenstown, MD	2009	Host	No	1	2	3	Yes	Yes
46	SRWW	Queenstown, MD	2009	Non-host	No	1	2	3	Yes	Yes
47	SWWW	Aurora, NY	2009	Host	No	1	–	1	Yes	Yes
48	SWWW	Aurora, NY	2009	Non-host	No	1	–	1	No	Yes
49	SWWW	Aurora, NY	2010	Non-host	Yes	1	–	1	Yes	Yes
50	SWWW	Aurora, NY	2010	Non-host	Yes	1	–	1	Yes	Yes
51	SRWW	Monmouth, IL	2010	Host	No	1	2	3	No	Yes
52	SRWW	Monmouth, IL	2010	Non-host	No	1	2	3	No	Yes
53	SRWW	Columbia, MO	2010	Host	No	2	1	2	Yes	Yes

[a] Number of cultivars in each of three resistance or susceptibility categories; susceptible (S), moderately susceptible (MS), and moderately resistant (MR). Resistance classifications were specific for each region and market class and were based on visual assessments of FHB intensity in FHB nurseries or cultivar performance trials over multiple years.

[b] Only experiments with >2% Fusarium head blight index or >1 ppm DON in the susceptible-untreated check treatment were considered.

[c] Code assigned to each study/experiment.

[d] Wheat market class: HRSW = hard red spring wheat, SRWW = soft red winter wheat, HRWW = hard red winter wheat, SWWW = soft white winter wheat.

[e] City and state in which the experiment was conducted.

[f] Experiment established in a field or plot previously planted with a host or non-host crop for the causal agent of FHB, *Fusarium graminearum*. Host crops included corn and wheat and non-host crops were either soybean or canola. Two experiments were established in fallowed fields; … = previous crop was unknown.

[g] Plots were either artificially inoculated (Yes) or naturally infected (No). Inoculation consisted of either the application of *F. graminearum* spore suspensions to wheat spike at anthesis, approximately 24 h before fungicide treatments were applied or *F. graminearum*-infected corn kernels broadcast in the plots prior to anthesis.

3 = moderately resistant, untreated (MR_UT); 4 = susceptible, fungicide treated (S_TR); 5 = moderately susceptible, fungicide treated (MS_TR); and 6 = moderately resistant, fungicide treated (MR_TR). Separate linear mixed models were fitted to the data from each trial, with management combination as the fixed effect, and block and the combination of the whole-plot factor and block as random effects. Standard errors for the interaction (i.e., management combination) means were estimated based on the fitted mixed model and experimental layout. The square of the standard error is known as the sampling variance in second-stage analyses.

Response ratios were calculated for each trial, as $R = \bar{X}_{Mgmt}/\bar{X}_{Check}$, where $\bar{X}_{Check}$ is the mean index or DON for S_UT, and $\bar{X}_{Mgmt}$ is the mean index or DON for one of the other five resistance–fungicide management combinations (MS_UT, MR_UT, S_TR, MS_TR, and MR_TR). The log of the response ratio (L) was then calculated as the effect size for each management combination as:

$$L = \ln(R) = \ln\left(\bar{X}_{Mgmt}/\bar{X}_{Check}\right) = \ln\left(\bar{X}_{Mgmt}\right) - \ln\left(\bar{X}_{Check}\right) \tag{1}$$

L was used for analysis because of its statistical properties (7,18). Up to five different L values were calculated for each study (labeled $L^{(MS\_UT)}, \ldots, L^{(MR\_TR)}$). The sampling variance for L was estimated using the method in Madden and Paul (18).

Stage two of the analysis was based on Paul et al. (27,28). Multivariate random-effects meta-analytical models were fitted to the index and DON data, with log of the response as the dependent variable (7) used to estimate the overall expected index and DON for different cultivar resistance–fungicide treatment management combinations. A vector consisting of the log of the means for each management combination was used for each study. The log mean for management combination i (i = S_UT, MS_UT, MR_UT, S_TR, MS_TR, or MR_TR) in study j ($j = 1, \ldots, K$) was defined as $Y_{ij} = \ln\left(\bar{X}_{ij}\right)$, where X is either index or DON. The within-study or sampling variance of the log means was estimated as $s_{ij}^2 = \upsilon/\bar{X}_{ij}^2$, where υ is the square of the standard error of the interaction mean from the individual trial (based on the X data). Note that υ from an individual study takes into account the experimental layout, number of replicates (usually three to six), and number of cultivars in the designated category.

Only trials with >2% index or >1 ppm DON in the S_UT check (Table 1) were included in the data matrix. Extremely low index or DON in the check (the reference treatment) leads to uncertainty in estimating the log response ratio (7), the effect size used to evaluate the effect of management combinations on index and DON (see below). In addition, one cannot determine the effect of a management practice on index and DON if these responses in the reference treatment are too low for there to be any effect to measure. The final data matrix consisted of 45 trials for index and 46 for DON.

Multivariate random-effects models (for index and DON separately) were fitted to the data using maximum likelihood with the MIXED procedure of SAS (17), as described by van Houwelingen et al. (42) and Paul et al. (28). Reasons for this meta-analytical model are given in Paul et al. (28). The estimated effect sizes of interest, the expected (mean) log response ratios, together with their standard errors and confidence intervals (CIs), were determined using *contrast* and *lsmeans* statements in PROC MIXED. For a given management combination, the mean log ratio is estimated as:

$$\bar{L} = \bar{Y}_{Mgmt} - \bar{Y}_{Check} \tag{2}$$

where $\bar{Y}_{Mgmt}$ is the estimated least-squares mean log response for each management combination and $\bar{Y}_{Check}$ is the least-squares mean log response for the check (nominally, S_UT). Equation 2 produces a log ratio because the log of the ratio is equal to the difference in logs (see equation 1). A standard normal test statistic (Z) was used to determine whether the log response ratios between the S_UT and other management combinations were statistically different from zero. Estimated mean percent control ($\bar{C}$) for index and DON provided by each management combination, relative to the S_UT, were calculated from the $\bar{L}$ values as $\bar{C} = \left[1 - \exp\left(\bar{L}\right)\right] \times 100$. Confidence intervals for percent control were calculated in a similar fashion from the upper and lower limits of the 95% CI around $\bar{L}$.

To specifically address questions about whether responses to fungicide depended on cultivar susceptibility, additional contrasts were calculated with either MS_UT or MR_UT as the check (see equation 2) in order to estimate the log mean response ratio and percent control for MS_TR and MR_TR. Confidence intervals and significance tests were performed as done for S_UT as the check.

Stability of resistance–fungicide interaction effects on index and DON. As discussed by Hedges (7) and Madden and Paul (18), a direct parametric (normal-distribution-based) meta-analysis of C_{ij} would not be appropriate for this stability analysis; however, this variable can be analyzed directly using nonparametric methods. Therefore, nonparametric methods were used to determine if the rank order of the effects of management combination on index and DON depended on the trial. For the purpose of this nonparametric stability analysis, each individual trial was considered a separate environment in the broad sense. From the list of 45 studies for index and 46 for DON used in the meta-analyses above, studies with one or more missing management combinations were omitted (a requirement for this nonparametric analysis), leaving a total of 37 studies ($N = 37$ trials or environments) for both index and DON. The percent control of index (C_{IND}) and DON (C_{DON}) relative to the untreated susceptible check (S_UT) with the ith management combination (Mgmt) in the jth environment was calculated as:

$$C_{Mgmt,j} = C_{ij} = \frac{\bar{X}_{Check,j} - \bar{X}_{Mgmt,j}}{\bar{X}_{Check,j}} \times 100 \tag{3}$$

Using the protocol outlined in Madden et al. (19), the arithmetic mean percent control for the ith management combination across all environments ($\bar{C}_{i\bullet}$), overall mean percent control across all tested management combinations ($\bar{C}_{\bullet\bullet}$), rank of C_{ij} for the management combination within each environment (R_{ij}), mean rank for ith management combination across all environments ($\bar{R}_{i\bullet}$), and the overall mean rank ($\bar{R}_{\bullet\bullet}$) were estimated for index (Table 2) and DON (Table 3). Variance of the ranks across all environments (the rank stability variance; $S_i^{(2)}$) was also calculated for each management combination as described by Madden et al. (19). $S_i^{(2)}$ equal to or close to zero will occur when there is no interaction of management and environment for the ranks or, equivalently, when the ranking is stable across environments.

Kendall's coefficient of concordance (W) (10) was calculated to evaluate the agreement of rankings across trials. For W, a value of 1 indicates that ranks are identical within each environment (maximum concordance) and, hence, that environment does not affect the rank order (i.e., that there is no rank interaction and no residual variation in the ranks). When $W = 1$, the sum of the $S_i^{(2)}$ values is 0. A small W (close to 0) means that there is no concordance in rankings (10); that is, the rankings vary with environment. Using W, along with the total number of environments ($N = 37$) and management combinations ($K = 5$), separate Friedman test statistics (T) were calculated for index and DON to formally test the null hypothesis of random ranking of management combinations across trials (lack of concordance of ranking or, equivalently, lack of a [consistent] treatment effect). With $N > 10$, T has a χ^2 distribution under the null hypothesis, with $K - 1$ degrees of freedom.

Management combination–environment interaction. Even when W is close to 1 and the test for concordance is significant, indicating that the rank order of management combinations does not depend on environment (suggesting a strong main effect of treatment combination), there may still be interaction on the original scale (percent control; C). Such an interaction may be manifested as differences among trials in the magnitude of the percent control due to the management combination on index and DON. Moreover, when W is significant but substantially less than 1, there is opportunity for a rank interaction of management combination and environment in addition to a main effect of management combination.

A nonparametric procedure developed by Piepho (34,35) was used to formally test for the interaction in terms of percent control. The protocol, as explained by Madden et al. (19), involves first determining contrasts of the response variable (percent control) for each management combination between pairs of environments. If C_{ij} and $C_{ij'}$ represent percent control for management combination i in environments j and j', respectively, then one can define $D_{il} = C_{ij'} - C_{ij}$ as the difference (contrast) between these environments. Thus, the data sets are reduced from $N = 37$ (rows in Tables 2 and 3) to $n = 1 + (37 - 3)/2 = 18$ "environments" ($l = 1, \ldots, 18$).

The new D_{il} contrast values are then ranked across management combinations within each row of the $n = 18$ new "environments" of contrasts (R_{il}), and the mean ranks ($\overline{R}_{i\bullet}$) and rank variances (U_i) (equation 11 in Madden et al. [19]) were calculated for each management combination. Piepho's U stability measure (34) was calculated based on U_i to test the stability of FHB management combination across environment. The null hypothesis tested was that the rank orders within each row of contrasts were independent, with each ordering of the management combinations being equally likely. With a large number of contrasts (such as here), U has a χ^2 distribution under the null hypothesis, with $K - 1$ degrees of freedom. A large U is indicative of a lack of homogeneity of rank variances, meaning that one or more of the rank variances (U_i) are large, reflecting an interaction. All nonparametric calculations were performed with a macro written by Madden et al. (19).

Results

Index and DON levels. Mean index and DON varied considerably among trials and management combinations (Fig. 1). Averaged across the 53 environments, S_UT had the highest mean level of index (14.8%) and DON (6.15 ppm), followed by MS_UT (10.5% and 4.9 ppm), S_TR (8.1% and 4.1 ppm), MR_UT (7.3% and 3.0 ppm), and MS_TR (6.4% and 3.0 ppm). The lowest mean levels of index and DON were found in the MR_TR (5.0% and 2.0 ppm) management combination. All means reported in this paragraph are arithmetic averages, and trials are not weighted as in meta-analysis. From the selected trials with >1 ppm mean DON in the S_UT check, DON contamination of grain in the MS_TR and MR_TR management combinations exceeded 2 ppm, an economically criti-

Table 2. Percent control of Fusarium head blight index (C_{ij}) with different cultivar resistance–fungicide management combinations (i) relative to the susceptible, untreated check in 37 studies (j), ranking of percent control (R_{ij}) for each management combination within each study, mean rank ($\overline{R}_{i\bullet}$) across studies, rank stability variance ($S_i^{(2)}$), and rank variance of contrasts (U_i) for each management combination

	Management combination (i)[a]									
	Percent index control (C_{ij})[b]					Rank (R_{ij})				
Study (j)[c]	MS_UT	MR_UT	S_TR	MS_TR	MR_TR	MS_UT	MR_UT	S_TR	MS_TR	MR_TR
1	30.35	44.57	47.11	67.62	64.35	1	2	3	5	4
2	68.17	81.41	78.31	91.55	97.89	1	3	2	4	5
3	81.42	95.09	85.14	94.38	97.84	1	4	2	3	5
4	3.90	76.23	57.56	65.96	72.16	1	5	2	3	4
6	85.47	58.10	76.17	97.50	84.39	4	1	2	5	3
9	–9.63	4.24	–25.98	–8.30	30.26	2	4	1	3	5
10	–16.30	32.95	5.62	–12.29	29.17	1	5	3	2	4
11	83.39	92.79	76.76	99.42	99.77	2	3	1	4	5
16	84.59	94.39	64.68	96.36	98.39	2	3	1	4	5
17	87.55	72.70	56.32	96.93	95.69	3	2	1	5	4
18	68.49	98.77	–61.14	47.30	97.73	3	5	1	2	4
19	87.85	96.99	35.90	80.20	95.97	3	5	1	2	4
20	69.70	92.63	16.40	82.69	89.54	2	5	1	3	4
21	84.93	93.22	58.06	90.95	96.39	2	4	1	3	5
22	34.00	47.79	40.75	60.44	66.11	1	3	2	4	5
23	23.85	39.60	12.06	50.32	34.83	2	4	1	5	3
24	22.64	35.68	7.86	46.17	36.66	2	3	1	5	4
25	–2.87	0.30	28.15	24.16	21.92	1	2	5	4	3
26	–6.73	16.10	30.23	30.93	30.21	1	2	4	5	3
27	1.99	33.96	63.39	38.98	73.30	1	2	4	3	5
28	29.41	59.40	49.67	33.33	76.47	1	4	3	2	5
29	67.29	25.29	80.23	87.43	53.72	3	1	4	5	2
30	61.58	–1.11	80.32	89.73	73.02	2	1	4	5	3
31	85.50	37.86	87.90	96.15	88.21	2	1	3	5	4
34	–10.76	65.82	89.05	56.59	92.26	1	3	4	2	5
36	25.23	43.99	92.64	75.63	93.32	1	2	4	3	5
37	62.77	67.58	81.35	84.21	79.85	1	2	4	5	3
38	22.92	24.61	58.83	79.81	95.43	1	2	3	4	5
39	80.80	1.54	85.17	96.06	29.70	3	1	4	5	2
40	79.88	55.64	61.03	83.53	64.30	4	1	2	5	3
41	63.90	5.01	79.57	78.89	56.29	3	1	5	4	2
42	2.73	61.27	81.46	58.89	92.05	1	3	4	2	5
43	60.27	97.47	60.53	80.42	96.77	1	5	2	3	4
44	22.93	81.32	66.67	56.23	83.53	1	4	3	2	5
45	80.95	96.16	63.23	93.56	98.43	2	4	1	3	5
46	91.54	95.13	73.90	97.88	98.62	2	3	1	4	5
53	33.75	42.85	35.97	58.45	65.59	1	3	2	4	5
$\overline{R}_{i\bullet}$	…	…	…	…	…	1.78	2.92	2.49	3.70	4.11
$S_i^{(2)}$	…	…	…	…	…	0.84	1.85	1.76	1.27	0.99
U_i	…	…	…	…	…	1.78	3.38	1.67	1.83	1.33

[a] Resistance–fungicide integrated management combinations: MS_UT = moderately susceptible, untreated; MR_UT = moderately resistant, untreated; S_TR = susceptible, fungicide treated; MS_TR = moderately susceptible, fungicide treated; and MR_TR = moderately resistant, fungicide treated.

[b] Percent control (C) of Fusarium head blight index (mean proportion of diseased spikelets per spike) estimated for each management combination relative to the susceptible, untreated using equation 3.

[c] Each study was considered a unique environment representing a different combination of wheat class, cropping system, location, and year.

cal threshold, in 36.4 and 22.0% of the observations, respectively, compared with 74.1% of the observations from S_UT.

Meta-analysis of effect of cultivar resistance–fungicide management combination on FHB and DON. The efficacy of resistance–fungicide combinations against FHB and DON was quantitatively evaluated based on the magnitude and significance of mean log-transformed response ratios ($\bar{L}$) for each combination relative to the S_UT combination. $\bar{L}$ was statistically different from zero for both index and DON, for all tested management combinations ($P < 0.001$; Table 4). For each combination, weighted mean percent control ($\bar{C}$) of index and DON and their 95% CIs were calculated from the $\bar{L}$ values and their CIs (Table 4). A large negative $\bar{L}$ value indicates a high positive percent control. Of all the tested management combinations, MR_TR had the lowest $\bar{L}$ for index, which corresponded to the highest mean percent control (75.7%; Table 4). This was followed by the MS_TR, MR_UT, S_TR, and MS_UT combinations, with mean percent control values of 70.9, 54.4, 52.9, and 42.8%, respectively. The MR_TR combination also resulted in the greatest reduction in DON, relative to the S_UT, with the smallest $\bar{L}$ and, correspondingly, the highest $\bar{C}$. $\bar{C}$ was 71.0% for MR_TR, followed by 58.4% for MS_TR, 50.7% for MR_UT, 38.9% for S_TR, and 30.1% for MS_UT.

The relative magnitude of index reduction due to fungicide treatment varied with the reference treatment (S_UT, MS_UT, or MR_UT). The $\bar{L}$ values were significantly different from zero for all comparisons between fungicide treatments and the untreated checks within the same resistance class; that is, S_TR versus S_UT, MS_TR versus MS_UT, and MR_TR versus MR_UT (Table 4). For index, the largest negative $\bar{L}$ value and, correspondingly, the highest $\bar{C}$ was observed when susceptible cultivars were treated with a fungicide rather than when the moderately susceptible or moderately resistant cultivars were treated. In particular, the mean percent control of index with fungicide relative to the untreated check for susceptible cultivars (S_TR versus S_UT) was 52.9%, compared with 49.2% for the moderately susceptible cultivars (MS_TR versus MS_UT) and 46.7% for the moderately resistant cultivars (MR_TR versus MR_UT). For DON, the $\bar{L}$ values for comparisons between treated and untreated plots within each resistance class were also significantly different from zero; how-

Table 3. Percent control of deoxynivalenol (DON) content of grain (C_{ij}) with different cultivar resistance–fungicide treatment management combinations (i) relative to the susceptible, untreated check in 37 studies (j), ranking of percent control (R_{ij}) for each management combination within each study, mean rank ($\bar{R}_{i\bullet}$) across studies, rank stability variance ($S_i^{(2)}$), and rank variance of contrasts (U_i) for each management combination

	Management combination (i)[a]									
	Percent DON control (C_{ij})[b]					Rank (R_{ij})				
Study (j)[c]	MS_UT	MR_UT	S_TR	MS_TR	MR_TR	MS_UT	MR_UT	S_TR	MS_TR	MR_TR
1	67.05	49.87	47.55	82.82	77.78	3	2	1	5	4
2	40.11	83.05	72.32	86.44	90.96	1	3	2	4	5
4	37.39	43.04	51.74	64.78	67.39	1	2	3	4	5
5	23.97	30.14	26.03	49.32	50.00	1	3	2	4	5
6	13.82	33.33	25.11	32.79	58.81	1	4	2	3	5
8	–12.72	–26.47	1.22	–1.22	17.59	2	1	4	3	5
10	–49.92	–34.28	16.47	–25.79	–37.60	1	3	5	4	2
16	51.83	72.57	20.79	74.15	80.10	2	3	1	4	5
17	64.24	71.22	28.68	72.36	83.99	2	3	1	4	5
18	–11.93	–14.21	–22.11	–47.89	–22.11	5	4	2	1	3
19	59.62	76.28	–8.97	56.62	77.56	3	4	1	2	5
20	55.83	76.31	33.57	69.40	80.23	2	4	1	3	5
21	62.25	79.44	37.63	78.40	87.11	2	4	1	3	5
22	58.21	70.54	62.10	83.13	88.97	1	3	2	4	5
23	72.45	75.96	10.85	71.12	59.27	4	5	1	3	2
24	62.32	68.25	11.92	61.46	82.58	3	4	1	2	5
25	25.54	64.57	32.73	70.14	80.79	1	3	2	4	5
26	42.12	62.51	49.55	72.57	89.11	1	3	2	4	5
27	33.81	58.39	41.68	48.45	78.90	1	4	2	3	5
31	3.23	42.07	24.82	34.97	69.70	1	4	2	3	5
32	29.05	60.09	4.59	42.42	85.09	2	4	1	3	5
33	6.71	63.18	17.76	55.96	82.35	1	4	2	3	5
34	25.32	68.35	61.52	71.77	81.39	1	3	2	4	5
36	17.60	54.74	40.69	45.14	74.69	1	4	2	3	5
37	1.36	21.63	18.37	–14.73	59.83	2	4	3	1	5
38	–9.36	38.84	59.70	25.32	73.59	1	3	4	2	5
39	60.30	36.75	42.17	69.26	64.63	3	1	2	5	4
40	52.06	40.92	31.40	67.19	61.39	3	2	1	5	4
41	48.84	41.17	32.01	71.26	65.21	3	2	1	5	4
42	17.01	65.40	69.05	67.12	88.93	1	2	4	3	5
43	43.84	51.13	66.60	83.91	84.83	1	2	3	4	5
44	53.65	79.68	83.70	73.29	83.88	1	3	4	2	5
45	78.83	93.26	30.00	87.08	95.87	2	4	1	3	5
46	74.29	87.93	46.52	85.79	90.89	2	4	1	3	5
51	12.30	24.13	48.26	52.05	66.25	1	2	3	4	5
52	13.90	–8.64	40.34	59.21	62.03	2	1	3	4	5
53	53.02	77.13	–3.18	54.65	81.42	2	4	1	3	5
$\bar{R}_{i\bullet}$	…	…	…	…	…	1.81	3.11	2.07	3.35	4.66
$S_i^{(2)}$	…	…	…	…	…	0.99	1.04	1.22	1.01	0.67
U_i	…	…	…	…	…	2.00	2.17	2.83	1.72	1.28

[a] Resistance–fungicide integrated management combinations: MS_UT = moderately susceptible, untreated; MR_UT = moderately resistant, untreated; S_TR = susceptible, fungicide treated; MS_TR = moderately susceptible, fungicide treated; and MR_TR = moderately resistant, fungicide treated.

[b] Percent control (C) of deoxynivalenol contamination of grain estimated for each management combination relative to the susceptible, untreated using equation 3.

[c] Each study was considered a unique environment representing a different combination of wheat class, cropping system, location, and year.

ever, the mean magnitude of the fungicide effect ($\bar{L}$ and $\bar{C}$ values) was very similar among the three resistance classes, with $\bar{C}$ of 38.9, 40.6, and 41.2% for the susceptible, moderately susceptible, and resistant cultivars, respectively (Table 4).

The width of the 95% CIs around $\bar{C}$ ranged from 15 to 27% for index and 13 to 23% for DON. In general, for comparisons between treated and untreated within a given resistance class (S_TR versus S_UT, MS_TR versus MS_UT, and MR_TR versus MR_UT), the width of the 95% CI was slightly narrower (15 to 18% for index and 14 to 17% for DON) than for comparisons across resistance classes (15 to 27% for index and 13 to 23% for DON).

Stability of cultivar resistance–fungicide treatment effect on FHB and DON. Percent control of index and DON relative to the S_UT for each management combination and environment (C_{ij}) are found in Tables 2 and 3, respectively. Across the 37 environments, C_{ij} for index ranged from –16.3 to 91.5% for MS_UT, –1.11 to 98.8% for MR_UT, –61.1 to 92.6% for S_TR, –12.3 to 99.4% for MS_TR, and 21.9 to 99.8% for MR_TR (Table 2). For DON, the corresponding ranges were from –49.9 to 78.8, –34.3 to 93.3, –22.1 to 83.7, –47.9 to 87.1, and –37.6 to 95.9% for MS_UT, MR_UT, S_TR, MS_TR, and MR_TR, respectively (Table 3).

Like C_{ij}, the ranking of a given management combination per environment (R_{ij}) also varied among environments. However, MR_TR mostly received rankings of 4 and 5 (best control), while MS_UT mostly received rankings of 1 and 2 (poorest control) across environments (Tables 2 and 3). For index, the MR_TR and MS_TR combinations received the highest mean ranks across the 37 environments ($\bar{R}_{MR\_TR\bullet} = 4.11, \bar{R}_{MS\_TR\bullet} = 3.70$), followed by MR_UT (2.92), S_TR (2.49), and MS_UT (1.78) (Table 2). For percent control of DON, the same general trend was observed, with MR_TR having the highest mean ranking across 37 environments ($\bar{R}_{MR\_TR\bullet} = 4.66$) (Table 3). This was followed by the MS_TR (3.35), MR_UT (3.11), S_TR (2.07), and MS_UT (1.81) management combinations.

The variance of rankings across environments ($S_i^{(2)}$) for each management combination was calculated as a measure of rank stability. For index, the greatest rank stability (lowest variance) was found for the MS_UT combination ($S_{MS\_UT}^{(2)} = 0.84$), followed by MR_TR (0.99), MS_TR (1.27), S_TR (1.76), and MR_UT (1.85). Thus, S_TR and MR_UT had the greatest inconsistencies in rankings. For DON, the lowest rank variance was found for MR_TR ($S_{MR\_TR}^{(2)} = 0.67$) and the highest for S_TR (1.22), with intermediate and similar variance values for the other management combinations (1.01 for MS_TR, 1.04 for MS_UT, and 0.99 for MR_UT). The highest rank variances for DON were considerably lower than the higher rank variances for index, indicating fewer inconsistencies in the management combination rankings for DON.

For index and DON, there was a highly significant ($P < 0.001$) concordance based on the T statistic for ranks (Table 5). This indicated that the management combinations clearly affected the rankings of percent control of index or DON compared with the check, and that there was an overall concordance in the rankings across trials. However, with Kendall's coefficient of concordance (W) considerably less than 1 (Table 5), especially for index, the rankings were clearly not identical across the trials.

Management combination–environment interaction. The values of W less than 1 (Table 5), and large $S_i^{(2)}$ values (Tables 2 and 3) can reflect random variation or could be (in part) a reflection of an interaction of management combination with environment on the original percent-control scale (19), which may or not be manifested in the ranks. The Piepho (34) test can help clarify these possibilities. The contrast rank variance (U_i) was smallest for MR_TR for both index and DON and largest for MR_UT for index and S_TR for DON (Tables 2 and 3). The value of $U_{MR\_UT} = 3.38$ for index is indicative of large differences among the trials for the ranking of this management combination. The U_i range across management combinations (difference between the highest and lowest) was wider for index (2.05) than DON (1.56). Piepho's U measure of stability was statistically significant for index (P = 0.011) but not for DON (P = 0.147) (Table 5), signifying an interaction for index but with less evidence of such an interaction for DON. This reflected the fact that at least one of the contrast rank variances (U_i) was large for index (Table 2; 3.38 for MR_UT) but all U_i values were of fairly similar magnitude for DON (Table 3).

The environment–management combination interaction for index can be addressed by reconsidering the rankings in Table 2. Although MS_UT generally had the lowest within-trial rank, there was a collection of environments (trial numbers 9, 11, 16, 17, 18, 19, 20, 21, 23, 24, 45, and 46, mostly from Maryland and Missouri) where S_TR had the lowest rank. Many of these trials also had large ranks for MR_UT (which, overall, had an intermediate rank). There was also a collection of environments (trial numbers 6, 29, 30, 31, 39, 40, and 41, mostly from Illinois) where MR_UT had the lowest within-trial ranks, with some of these trials (numbers 6, 29, 39, 40, and 41) also having relatively large ranks for MS_UT.

For DON, similar trends were observed for the interaction between environment and management combination, with S_TR

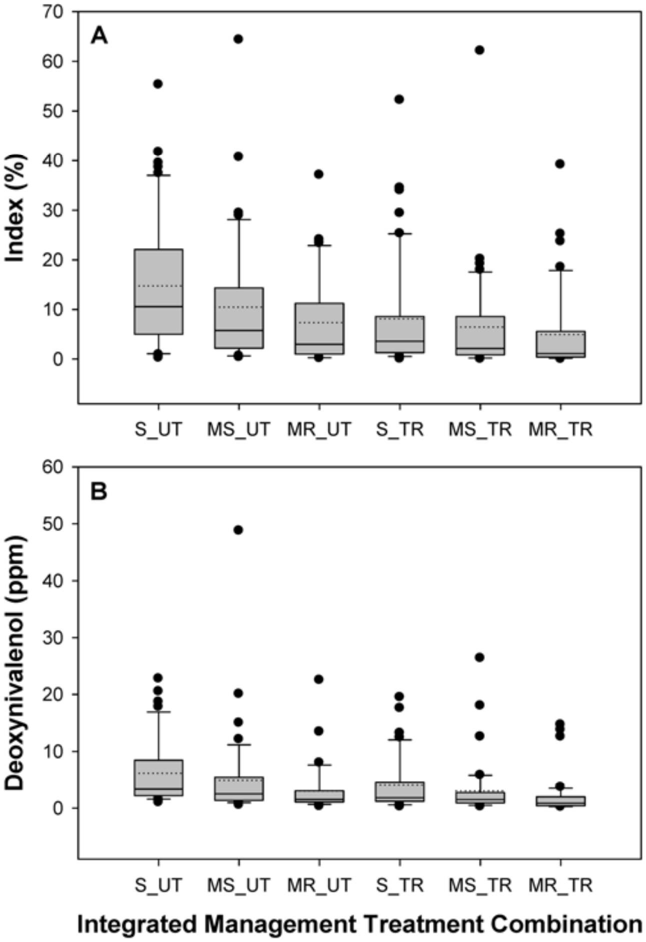

Fig. 1. Box plots of **A,** mean Fusarium head blight (FHB) index (mean percentage of disease spikelets per spike) and **B,** deoxynivalenol accumulation in harvested grain for six cultivar resistance–fungicide integrated management combinations—susceptible, untreated (S_UT); moderately susceptible, untreated (MS_UT) moderately resistant, untreated (MR_UT); susceptible, treated (S_TR), moderately susceptible, treated (MS_TR); and moderately resistant, treated (MR_TR)—from inoculated and uninoculated winter and spring wheat experiments conducted from 2007 to 2010 in 53 unique environments. Fungicide treatment consisted of a single application of a pre- or tank-mix of tebuconazole + prothioconazole at anthesis. Solid and dotted lines within the box represent median and mean, respectively, and top and bottom lines of the boxes represent the 75th and 25th percentiles of the data. Whiskers extending above and below boxes represent the 10th and 90th percentiles and circles indicate outliers.

rather than MS_UT receiving the lowest within-trial rank in more than a third of the environments (trial numbers 1, 16, 17, 19, 20, 21, 23, 24, 32, 40, 41, 45, 46, and 53). On the other hand, MR_TR, the management combination with the highest overall mean rank, received low-to-intermediate rank in 3 of the 37 trials (numbers 10, 18, and 23), and MS_TR, the combination with the second highest overall mean rank, received the lowest or second lowest rank in six trials (numbers 18, 19, 24, 37, 38, and 44).

Discussion

Meta-analysis was employed, in part, to determine the efficacy of combining cultivars with varying levels of resistance to FHB (based on visual symptoms) with a single application of fungicide at anthesis to manage FHB and DON. Based on situations with >2% index and >1 ppm DON in the checks (the conditions when management is needed), all combinations of host resistance and foliar fungicide significantly reduced FHB and DON relative to S_UT. However, the magnitude of the response varied among the management combinations, with MR_TR providing the highest percent control (>70%) for both index and DON relative to S_UT, followed by MS_TR, MR_UT, S_TR, and MS_UT. This ordering of management-combination means from the meta-analysis was supported by the results from the nonparametric concordance analysis, based on the *W* and *T* statistics. However, the nonparametric stability analysis confirmed that, especially for index, despite the overall ranking, there were groups of trials where different rankings occurred. Of all the management combinations tested, the relative efficacy of MR_UT for control of index was the most likely to vary with trial.

Relative to S_UT, in terms of percent control, MR_TR outperformed moderate resistance alone (MR_UT) by approximately 20%, on average, for both index and DON, and fungicide alone (S_TR) by approximately 23% for index and 32% for DON. Comparisons between treated and untreated plots within susceptible, moderately susceptible, and moderately resistant classes (S_TR versus S_UT, MS_TR versus MS_UT, and MR_TR versus MR_UT, respectively) showed that mean percent control for index tended to be higher when susceptible cultivars were treated than when moderately susceptible or moderately resistant cultivars were treated. Because the level of index and DON in the reference treatment in these comparisons (the denominator in equation 1) was lower for the more resistant cultivars, a fixed absolute reduction in index would translate into a larger percent control for the more resistant cultivars. Because this was not observed, the actual reduction in index (not relative change) was smaller for the more resistant cultivars than the susceptible ones. Interestingly, the percent control of DON with a tebuconazole + prothioconazole application in the current investigation was similar for the three resistance classes. Thus, the actual reduction in DON in ppm was proportional to the DON level in the reference treatment (which varied with resistance class).

The observed overall superior efficacy (based on percent control) of combining moderate resistance with tebuconazole +

Table 5. Statistics for testing for concordance of ranks and rank homoscedasticity of percent control of Fusarium head blight index and deoxynivalenol (DON) concentration of harvested grain as a result of five cultivar resistance–fungicide management combinations in *N* wheat-growing environments

Statistic[a]	Index	DON
N	37	37
W	0.35	0.52
T	51.37	76.79
df for *T*	4	4
P for *T*	<0.001	<0.001
U	13.17	6.79
df for *U*	4	4
P for *U*	0.011	0.147

[a] *N* = number studies or environments; *W* = Kendall's coefficient of concordance (10), where a value of 1 indicates that ranks are identical within each environment (maximum concordance) and 0 means that there is no concordance in rankings; *T* = Friedman test statistics used to test for lack concordance of ranks within environments; *U* = Piepho's *U* test statistic used to test for management combination–environment interaction, where a large *U* value indicates a lack of homogeneity of rank variances, meaning that one or more of the rank variances are large, reflecting an interaction; df = degrees of freedom; and *P* = level of significance.

Table 4. Log of the response ratio, percent control, and corresponding statistics for the effect of integrated management combinations on Fusarium head blight (FHB) index and deoxynivalenol (DON) in winter and spring wheat

		Effect size[a]						Mean percent control[b]			
Combinations[c]	*K*[d]	$\bar{L}$	$SE(\bar{L})$	CI_L	CI_U	*Z*	*P*	$\bar{C}$	CI_L	CI_U	$\hat{\sigma}^2$ [e]
Index											
MS_UT vs. S_UT	38	–0.56	0.122	–0.80	–0.32	–4.64	<0.001	42.80	27.54	54.84	0.533
MR_UT vs. S_UT	44	–0.79	0.146	–1.05	–0.52	–5.77	<0.001	54.40	40.44	65.09	0.630
S_TR vs. S_UT	45	–0.75	0.084	–0.91	–0.60	–9.34	<0.001	52.94	44.86	59.83	0.188
MS_TR vs. S_UT	38	–1.24	0.160	–1.53	–0.94	–8.29	<0.001	70.92	61.05	78.29	0.748
MR_TR vs. S_UT	44	–1.42	0.154	–1.72	–1.11	–9.00	<0.001	75.72	66.94	82.16	0.807
MS_TR vs. MS_UT	38	–0.68	0.086	–0.85	–0.51	–7.84	<0.001	49.16	39.79	57.07	0.175
MR_TR vs. MR_UT	44	–0.63	0.087	–0.80	–0.46	–7.20	<0.001	46.74	36.77	55.14	0.117
DON											
MS_UT vs. S_UT	38	–0.36	0.084	–0.52	–0.19	–4.24	<0.001	30.06	17.46	40.73	0.278
MR–UT vs. S_UT	45	–0.71	0.101	–0.91	–0.51	–6.98	<0.001	50.68	39.83	59.57	0.379
S_TR vs. S_UT	46	–0.49	0.058	–0.61	–0.38	–8.42	<0.001	38.85	31.42	45.47	0.107
MS_TR vs. S_UT	38	–0.88	0.092	–1.06	–0.70	–9.53	<0.001	58.42	50.19	65.29	0.308
MR_TR vs. S_UT	45	–1.24	0.116	–1.47	–1.01	–10.66	<0.001	71.01	63.59	76.91	0.444
MS_TR vs. MS_UT	38	–0.52	0.062	–0.64	–0.40	–8.38	<0.001	40.55	32.85	47.37	0.104
MR_TR vs. MR_UT	45	–0.53	0.075	–0.68	–0.39	–7.13	<0.001	41.21	31.95	49.21	0.086

[a] $\bar{L}$ = mean log of the response ratio between each management combination and the S-untreated check; SE($\bar{L}$) = standard error of $\bar{L}$; *Z* = standard normal test statistic; *P* = significance level; CI_U and CI_L = upper and lower limits of the 95% confidence interval around $\bar{L}$.

[b] $\bar{C}$ = mean percent control as estimated from $\bar{L}$ as $\bar{C} = [1 - \exp(\bar{L})] \times 100$; CI_U and CI_L = upper and lower limits of the 95% confidence interval around $\bar{C}$.

[c] FHB index (proportion of diseased spikelets per spike) and deoxynivalenol (DON) from harvested grain (ppm). Cultivar resistance–fungicide management combinations compared with the susceptible-untreated check, where S = susceptible, MS = moderately susceptible, or MR = moderately resistant cultivars were treated (TR) with fungicide (tebuconazole + prothioconazole) or left untreated (UT).

[d] Total number of unique environments or studies (where index >2% or DON >1 ppm in S-untreated check) used in each analysis (based on availability of index and DON data for each combination).

[e] Estimated between-study variance.

prothioconazole application at anthesis (MR_TR) over resistance alone (MR_UT) and fungicide treatment alone (S_TR), and the superior efficacy of moderate resistance alone (MR_UT) over fungicide alone (S_TR) against DON, were consistent with results from some other studies conducted across fewer environments and wheat classes (1,22,43). This is not surprising because, by definition, the moderately resistant cultivars would be expected to have lower levels of index and DON than the untreated susceptible check, and treating these cultivars with a fungicide would be expected to reduce disease and toxin levels even further relative to S_UT. Moreover, relative to fungicide treatment, moderate resistance contributes to disease and toxin reduction through a variety of mechanisms, including reduction in infection (type I resistance), colonization of the spike (type II resistance), and DON accumulation and/or detoxification (2,14,38,47). Fungicides are only effective for a relatively short period, whereas resistance mechanisms act for a longer time during the growing season, potentially contributing to greater efficacy against DON than fungicide application alone.

Questions have been raised as to whether the combined effect of fungicide treatment and cultivar resistance is additive or synergistic. With percent control (a variable on a multiplicative scale) as the response, there is no unambiguous meaning of additivity or synergy for combined effects; however, one cannot simply add the percentages when making comparisons. Perfect additivity can be defined as $\overline{C}_{MR\_TR}$ being equal to

$$1-\left[\left(1-\overline{C}_{S\_TR}\right)\times\left(1-\overline{C}_{MR\_UT}\right)\right]$$

where each percent control value is written as a proportion. From the meta-analysis, the overall mean percent control of index was 52.9% for S_TR and 54.4% for MR_UT. Therefore, 1 – [(1 – 0.529) × (1 – 0.544)] = 0.785, which is very similar and just slightly higher than 0.757 for $\overline{C}_{MR\_TR}$ from the meta-analysis (Table 4). For DON, with $\overline{C}_{S\_TR}$ = 38.9% and $\overline{C}_{MR\_UT}$ = 50.7%, the corresponding $\overline{C}_{MR\_TR}$ value is 0.699 (= 1 – [(1 – 0.389) × (1 – 0.507)]), almost equal to 0.710 from the meta-analysis. These results suggest that, based on percent control, the effect of integrating a single tebuconazole + prothioconazole application at anthesis with cultivar resistance is additive for both index and DON.

Results from this investigation on the expected percent control of index and DON due to tebuconazole + prothioconazole application to susceptible cultivars are very comparable with those from a meta-analysis by Paul et al. (28) on data from uniform fungicide trials, in which mainly susceptible cultivars were used. Here S_TR resulted in 52.9% control of index and 38.8% control of DON, compared with 51.8 and 41.7% in Paul et al. (28) for index and DON, respectively. However, our findings for expected values are somewhat contrary to those reported by Wegulo et al. (43). Percent tebuconazole + prothioconazole efficacy (equivalent to percent control as used here) in reducing index on susceptible cultivars was considerably lower in the study by Wegulo et al. (43) (22.5%) than the mean values in this investigation and in Paul et al. (28). In addition, Wegulo et al. (43) reported that percent control of index and DON with tebuconazole + prothioconazole was greater on moderately resistant than on susceptible cultivars. Interestingly, however, the magnitude of the fungicide effect (mean percent control) on index on moderately resistant cultivars (45.6%) was very similar to the value observed in the current investigation (46.7%). We also noted that the percent control values found across all trials (Table 2) for S_TR in our investigation included values even smaller than those reported by Wegulo et al. (43); thus, there could be several possible explanations for their specific results. The most likely explanation is the very high index values (55 to 98%) in the check plots in Wegulo et al. (43), whereas the mean index values in the current investigation ranged from 2 to 65%. Paul et al. (27) previously showed an effect of baseline index and DON on percent control but the available data did not allow them to consider baseline index values above 65%.

For all combinations of fungicide and cultivar resistance tested in this study, the magnitude of mean index reduction relative to the untreated susceptible was greater than the magnitude of DON reduction. However, the difference in efficacy between index and DON for moderate resistance–fungicide treatment (MR_TR: 75.7 to 71.0% = 4.7%) or resistance alone (MR_UT: 54.4 to 50.7% = 3.7%) was much smaller than for fungicide treatment alone (S_TR: 52.9 to 38.9% = 14.0%). Paul et al. (27,28) also observed greater percent control of index than of DON with fungicide application alone (on susceptible cultivars) and speculated that the inferior efficacy against DON was probably due to DON contamination from late-season primary infections, fungal colonization of spikes late in the season, and possibly even secondary infections well after anthesis, when the single fungicide application would no longer provide protection. DON contamination after the period of greatest fungicide effect may cause the fungicide to appear less effective than it initially is. However, when the fungicide is combined with moderate resistance, it is possible that primary infection around anthesis and spread within the spike are reduced by type I and type II resistance and the fungicide activity, and infection and colonization after anthesis are reduced by type I and type II resistance. Hence, in agreement with our results, it seems reasonable to hypothesize that this combined effect of fungicide and resistance likely reduces DON contamination between anthesis and harvest, attenuating the disparity between efficacy against index and efficacy against DON.

In addition to being the most effective management combination, MR_TR also was one of the most stable in terms of rank order across the 37 environments evaluated in this investigation, as indicated by the relatively small rank stability variance ($S_i^{(2)}$). Although the ranking of MR_TR based on percent control varied somewhat among environments, this management combination generally received the highest within-study rankings (ranks of 4 or 5 in 73% of the trials for index and 92% of the trials for DON), leading to the highest mean rank. The worst management combination (i.e., the one that resulted in the lowest mean percent control, MS_UT) also was generally stable across the 37 environments, with the lowest mean rank and one of the lowest rank stability variances. However, there were a few trials where MS_UT had one of the larger percents control for index (trial numbers 6 and 40). For both index and DON, the least stable management combinations, based on the large $S_i^{(2)}$ values, were MR_UT and S_TR. For instance, although S_TR had the second lowest mean rank for percent control of index, all five possible rankings were found more than once across the trials, and there was a collection of trials where S_TR was ranked fourth.

Environmental effects on resistance response and fungicide efficacy across trials could partially explain the relatively high rank stability variances for S_TR and MR_UT, especially for index. The fact that the lowest rank variances (highest stabilities) were for the extreme combinations in terms of efficacy (generally the best and worst management combinations, MR_TR and MS_UT, respectively) suggests that the combined effect of moderate resistance and fungicide treatment (MR_TR) was probably too strong for the environment (broadly speaking) to cause the rank order to change substantially from one trial to another. Similarly, at the other end of the efficacy spectrum, with MS_UT, the combination of a lower level of resistance (relatively speaking) and lack of fungicide protection was too “strong” for environment to have a major effect on the rank order in these trials with mostly high inoculum levels. The effects of intermediate management combinations (resistance without fungicide and fungicide without resistance), on the other hand, were apparently not strong enough to override (i.e., “resist”) environmental effects on the results, leading to greater variation in the within-trial ranks.

With the magnitude of the $S_i^{(2)}$ values, the W concordance coefficient was substantially below 1, especially for index. Even though the Friedman test was highly significant, showing that there was not an overall random ordering of management combinations, there was ample “room” for interactions of trials and management combinations in addition to random variation. This served as the impetus for using Piepho's *U* statistic (34) to test equality of stability variances based on contrasts between trials for each of the management combinations. The Piepho approach removes the main effect of management combination to more directly assess the interaction. Here, we found significant nonhomogeneity of variances of the ranked contrasts for index but not DON, indicating an interaction for percent control of the former but not the latter response. This interaction was evident by the fact that there were distinct groups of trials from specific locations for which the rankings for some management combinations were contrary to the overall mean rankings. For instance, there was a collection of trials, mostly from Illinois, for which MR_UT, the management combination with the third highest mean rank, had the lowest rank (instead of MS_UT, the combination with the lowest mean rank). Interaction for percent control of index may be attributed to several factors, including differential reactions among cultivars within a given resistance class or similar reactions among cultivars in different resistance classes as influenced by wheat type, genetics, and maturity; variability in fungicide effects, local pathogen population, and weather conditions; and complex interactions involving these factors. Wheat type (27,28), weather conditions (36), cultivar reaction, and pathogen aggressiveness (23) have all been reported to influence fungicide efficacy against FHB and DON.

To our knowledge, this is the first comprehensive quantitative analysis of efficacy and stability of FHB and DON integrated management strategies across multiple environments. Based on the meta- and stability analyses, we conclude that (i) combining moderate resistance with fungicide treatment is more effective at reducing FHB and DON than either resistance or fungicide treatment alone; (ii) relative to fungicide alone (for a susceptible cultivar), which is generally more effective against index than DON, integrating resistance and fungicide or using resistance alone provide comparable levels of index and DON reduction; (iii) the integrated approach is more stable across environments in terms of rank order than the individual approaches (MR_TR had the lowest rank variances); (iv) there was more evidence for an interaction of environment and management combination for index than for DON; (v) in terms of percent control (a response on a multiplicative scale), there was an additive effect of fungicide (S_TR) and resistance (MR_UT) on both index and DON; and (vi) percent control of index with fungicide tended to be lower when applied to moderately resistant cultivars (the untreated moderately resistant cultivar used as the reference) than to susceptible cultivars. These results, based on data from studies representing different wheat market classes, weather conditions, baseline levels of disease and toxin, and cropping practices, suggest that, regardless of local study-specific factors, MR_TR was the most effective and stable management practice for FHB and DON. Some of the intermediate management combinations, however, could vary in their rank order for different trials, especially for index. For reasons discussed above and based on results from uniform fungicide efficacy studies (27,28), even though MR_TR will generally give the best control relative to other management combinations tested, we anticipate that the magnitude of the actual percent control (not necessarily the rank) will vary with wheat class, baseline levels of disease and DON, and cropping sequence. Further investigation is needed to formally evaluate the effects of some of these study-specific factors on percent control. The available data did not allow for such an analysis here. For instance, nearly 72% of trials included in this study used soft red winter wheat cultivars, 15% used hard red winter wheat, 8% used soft white winter wheat, and only 6% used hard red spring wheat. More studies from underrepresented wheat classes are required to adequately evaluate the effects of wheat class of percent control due to MR_TR. Future analyses should include crop rotation or cropping sequence as either a moderator variable in a meta-analysis or a third integrated management strategy in a multifactorial, multilocation analysis. Evidence from uniform integrated management trials suggest that such a three-tiered approach, combining a moderately resistant cultivar and fungicide application at anthesis with rotation following a non-host crop, results in greater reductions in index and DON than an approach based on one or two management strategies (1,44).

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