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Effect of fungicide combinations for fusarium head blight control on disease incidence, grain yield and quality of winter wheat, spring wheat and barley

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Abstract: This study investigates the effects of timing of fungicide applications alone or in combinations on Fusarium Head Blight (FHB), seed deoxynivalenol (DON) concentrations, dominant leaf diseases, grain yield, and thousand kernel weight (TKW) in winter wheat, spring wheat, and barley in the Atlantic region of Canada. The experiments were conducted for three years (2010-2012) with fungicide treatment as the main factor. Selected commercially available fungicide treatments were applied at two timings: 1) ZGS 39: check; propiconazole/trifloxystrobin (125 g ha^{-1}); propiconazole (125 g ha^{-1}); and pyraclostrobin (100 g ha^{-1}) and 2) ZGS 60: check; prothioconazole (200 g ha^{-1}); prothioconazole/tebuconazole (200 g ha^{-1}); and metaconazole (90 g ha^{-1}). Results show that a single fungicide application was not sufficient to achieve a high yield with good seed quality. Reduction of visual FHB infection due to fungicide applications did not guarantee a reduction in seed DON concentrations. Fungicide application pyraclostrobin at ZGS 39 + prothioconazole/tebuconazole at ZGS 60 was the best treatment consistently providing the highest crop yield and seed quality, including lowered DON.

Keywords: Fungicide, Fusarium, Deoxynivalenol, Wheat, Barley

Abbreviations: FHB, Fusarium head blight; DON, Deoxynivalenol; ZGS, Zadoks growth stage;

TKW, Thousand kernel weight; FDK, Fusarium damaged kernels.

INTRODUCTION

Fusarium head blight (FHB) is caused primarily by *Fusarium graminearum* Schwabe [teleomorph *Gibberella zeae* (Schweinitz) Petch] and *Fusarium culmorum* [(W.G.Smith) Sacc.] (Chelkowski et al. 2000). These pathogens infect spikes and reduce grain yield by decreasing grain number per ear, thousand kernel weight (TKW), and grain weight per ear (Chelkowski et al. 2000; Martin 2004). Besides yield losses, *Fusarium* spp. infection also decreases grain quality by causing mycotoxin contamination of grain (Charmley et al. 1994; Jones and Mirocha 1999; Salas et al. 1999). The principal mycotoxin of concern in Canada is deoxynivalenol (DON) which threatens human and animal health (Joffe 1978; D'Mello et al. 1999; Góral et al. 2002). In the Atlantic region of Canada, *F. graminearum* is the primary pathogen causing FHB (Martin 2004). It can be found on all common cereals, such as wheat, barley and oats (Martin 2004). Its visual symptoms are the most obvious in spring wheat but the absence of visual symptoms does not assure low mycotoxin content (Martin 2004).

For successful FHB control, fungicide application timing, rate and selection are all considered to be important in wheat and barley (Mesterházy 2003). Fernandez et al. (2012) in Saskatchewan reported that a single tebuconazole application at ZGS 60 resulted in the most consistent reduction in FHB and improved test weight and kernel weight when compared to a single application at ZGS 31-36 (stem elongation) or at ZGS 37 (flag leaf emergence). Double fungicide applications at ZGS 31 or 37 and at ZGS 60 did not show any better efficacy on disease control than the single application at ZGS 60 (Fernandez et al. 2012). Fernandez et al. (2012) also noted that fungicide application at ZGS 31-36 or ZGS 37 might cause kernel discolouration which is considered as a grain downgrade. Yoshida et al. (2012) reported that thiophanate-methyl application at ZGS 77 (late milk) reduced fusarium damaged kernels (FDK

%) as well as mycotoxin concentration. However, it did not reduce FHB severity significantly (Yoshida et al. 2012). A more crucial timing for FHB control is ZGS 65 (flowering half complete) (Siranidou and Buchenauer, 2001; Magan et al. 2002; Yoshida et al. 2012).

There is a paucity of information on the efficacy of single foliar fungicide application and combined fungicide applications at different growth stages to control leaf diseases and FHB in winter wheat, spring wheat and spring barley in the Atlantic region of Canada. In the absence of such information, growers have had to depend on anecdotal evidence and not always unbiased advice. The objectives of this study were to investigate the effect of timing of fungicide applications alone or in combinations on diseases and grain yield in winter wheat, spring wheat and spring barley.

MATERIALS AND METHODS

Plant Material

Field research was conducted during 2010 – 2012 with cereal cultivars popular with local growers: winter wheat variety ‘Emmit’ planted at Canning, NS; spring wheat variety ‘AC Helena’ at Truro, NS and 6-row spring barley variety ‘AC Westech’ at Truro, NS. Cultivars chosen were recommended varieties for the Maritime region. The idea was to compare a winter wheat (Emmit) to a spring wheat cultivar (AC Helena) as they tend to have different disease pressure levels especially FHB due to the temperatures and moisture conditions at the time of anthesis. Emmit is a soft red winter wheat moderately susceptible to FHB. AC Helena is a high yielding milling wheat but it is slightly more susceptible to FHB than lower yielding cultivars adapted to Eastern Canada (Nass et al, 2000). AC Westech, a six-row barley variety (Choo et al, 1999), was chosen as six-row varieties have a tendency to be more susceptible to FHB than

2-row barley due to the shape of the head which acts to maintain more moisture around the florets.

Fungicide Treatment

Fungicide treatments included 1) early fungicide treatments (applied at ZGS 39 – flag leaf ligule); 2) late fungicide treatments (applied at ZGS 60 – beginning of anthesis) and double fungicide treatments (applied at both ZGS 39 and ZGS 60). Fungicides applied at ZGS 39 were: 1) propiconazole (Tilt® 250 EC; 125 g a.i. ha⁻¹; cost \$26 ha⁻¹); 2) propiconazole/trifloxystrobin (Stratego® 250 EC; 125 g a.i. ha⁻¹; cost \$26 ha⁻¹) and 3) pyraclostrobin (Headline® 250 EC; 100 g a.i. ha⁻¹; cost \$49 ha⁻¹) and at ZGS 60 were: 1) prothioconazole (Proline® 480 SC; 200 g a.i. ha⁻¹; cost \$100 ha⁻¹); 2) metaconazole (Caramba® 90; 90 g a.i. ha⁻¹; cost \$49 ha⁻¹) and 3) prothioconazole/tebuconazole (Prosaro® 250 EC; 200 g a.i. ha⁻¹; cost \$49 ha⁻¹). Sixteen treatments were used including no fungicide treatment check, single fungicide treatments at either of the two timings and double fungicide treatments (see Table 1 for full treatment list).

Experimental Design and Data Collection

The experiment was conducted in small plots (2 m * 5 m with 15 cm row spacing) with 16 treatment combinations arranged in a randomized complete block design with 4 blocks.

Treatments were applied at the highest label recommended rate with a bicycle sprayer equipped with Double Turbo Tee Jet nozzles at a water volume of 330 L ha⁻¹ (Anonymous, 2009; Anonymous, 2013; Anonymous, 2014; Anonymous, 2015a; Anonymous, 2015b; Anonymous, 2015c). Ratings of septoria leaf blight (SLB), powdery mildew, net/spot blotch and scald based on a 0-9 disease severity scale were recorded three times at approximately ZGS 60, ZGS 71

(Kernel watery) and ZGS 85 (Soft dough) in each cropping season. Disease severity at ZGS 60 was rated before the late fungicide applications to evaluate the efficacy of the early applied fungicides. Fusarium head blight (FHB) symptoms were assessed on the winter and spring wheat during ZGS 80-85 by rating the whole plot on a 1-10 scale for incidence and a 1-10 scale for severity and calculating an FHB index by multiplying incidence by severity for a value between 1 and 100. Mature grain was harvested with a Hege 125C small plot combine. Harvested area was 1.25 m * 5 m. Seed was collected, dried, cleaned and weighed. Test weights and thousand kernel weights (TKW) were determined for each plot. Deoxynivalenol (DON) analysis was conducted on sub samples of 150 g from bulked samples of replications (one and two) and (three and four) of each treatment. The level of DON was measured with a competitive direct ELISA test (Neogen Veratox for DON 5/5 kit) by A & L Canada Laboratories Inc. in London, Ontario. This test detects DON levels of 0.5 ppm and above. In treatments where the DON concentration was reported as 0, A & L Canada Laboratories Inc. reported there was no detectable DON and was reported as BDL (Below Detectable Limit).

Statistical Analysis

Year effect was combined with block effect as a new block effect which is considered as a random effect. Fungicide treatment effect was considered as a fixed effect. Different species were analyzed separately. Disease severity of powdery mildew, septoria, blotch and scald were not analyzed when the highest severity scored less than or equal to 3 in a 0 (no infection) to 9 (severe) scale. Analysis of variance (ANOVA) was performed using Proc Mixed in SAS software (version 9.3, SAS Institute Inc., Cary, NC). Fisher's LSD multiple means comparison test was used to compare treatment means when ANOVA indicated a significant effect by the fungicide treatments. Contrast comparisons were applied to compare the treatment means of the untreated

check, early, late and double fungicide application programs.

RESULTS

‘Emmit’ Winter Wheat

Experimental treatments had significant effects on yield, TKW, FHB severity, powdery mildew severity at ZGS 71 and septoria severity at all disease evaluation timings (Table 2). DON concentrations of the samples were all 0 which indicates below detectable limits of 0.5 ppm. ‘Emmit’ yielded significantly higher when receiving applications of treatment (trt) 6 than with trt1, trt2, trt3, trt4, trt5, trt9 and trt10, while the untreated check yielded the lowest (Figure 1). The TKW of ‘Emmit’ was significantly higher with trt10 applications compared to trt1, trt2, trt3, trt4, trt5, trt7, trt8, trt9 and trt13. Untreated “Emmit” produced the lightest seed. FHB index was found to be significantly higher with no fungicide applications compared to all other treatments. Powdery mildew severity evaluated at ZGS 60 and ZGS 85 showed no significant effects by fungicide treatments. At ZGS 71, except with trt2, trt3, trt4, trt5 and trt13, ‘Emmit’ with no fungicide treatments had significantly higher powdery mildew severity than the other treatments. Septoria severity evaluated at all three timings was significantly affected by fungicide treatments. Trt9 applied at ZGS 39 significantly reduced septoria infection evaluated at ZGS 60 compared with the check and trt13. Untreated “Emmit” showed significantly higher septoria infection at both ZGS 71 and ZGS 85 than the other treatments except trt2, trt3, trt4 and trt5 at ZGS 71 and trt4 and trt13 at ZGS 85 (Figure 1).

When compared to the check, early fungicide treatments significantly decreased FHB severity, powdery mildew severity at ZGS 71 and septoria severity at both ZGS 71 and ZGS 85 (Table 3). Late fungicide treatments significantly increased the yield, decreased FHB severity and decreased septoria severity at ZGS 85. Double fungicide treatments significantly increased

the yield and TKW. They also had significantly better than check efficacy on infections of FHB, powdery mildew at ZGS 71 and septoria at ZGS 71 and ZGS 85 (Table 3).

Between early, late and double fungicide treatments, double fungicide treatments performed significantly better than late fungicide treatments on all of the variables measured except for powdery mildew severity at ZGS 85 (Table 3). Compared to early fungicide treatments, double fungicide treatments had significantly greater yield and TKW, less FHB infection and less septoria infection at ZGS 85. Early fungicide treatments yielded significantly less than the late fungicide treatments. However, 'Emmit' treated with early applied fungicides showed significantly lower septoria infection at ZGS 71 than those treated with late applied fungicides (Table 3).

'AC Helena' Spring Wheat

Experimental treatments significantly affected yield, TKW, FHB severity, and septoria severity at all evaluation timings and DON concentrations (Table 2). Trt15 yielded the highest with no significant difference from trt12 and trt16 (Figure 2). The untreated check yielded the lowest and significantly lower than all the other treatments. TKW was the highest when 'AC Helena' was treated with trt16, but was not significantly different from trt7, trt11 and trt15. TKW was significantly less and FHB severity was significantly higher in the untreated treatment than all the other treatments. DON levels in 'AC Helena' were found to be significantly higher when treated with trt13 than the other treatments except trt5, trt7 and trt15. No treatment significantly decreased the DON levels compared to the check. Septoria severity was significantly reduced by trt13 at ZGS 60 compared to the check. At ZGS 71, trt6, trt7, trt8, trt10, trt11, trt12, trt14, trt15 and trt16 showed significantly better efficacy on septoria control than the check. At ZGS 81, except for trt2, trt5 and trt9, all of the other fungicide treatments had significantly lower septoria

severity than the check (Figure 2).

Compared to the check, early, late and double fungicide treatments significantly increased the yield and the TKW and significantly decreased FHB infection (Table 3). Although all fungicide treatments applied at different timings significantly decreased FHB infection, they did not show any advantage in DON reduction than the check. In contrast, early fungicide treatments produced significantly more DON than the check. For septoria, double fungicide treatments significantly reduced septoria severity at both ZGS 71 and ZGS 85, while late fungicide treatments showed their efficacy at ZGS 85 (Table 3).

When comparing the effects of the early, late and double fungicide treatments, double fungicide treatments had significantly greater yield, greater TKW, less FHB infection, lower septoria severity than both the early and late fungicide treatments (Table 3). It also significantly reduced DON level than the early fungicide treatments. Early fungicide treatments showed significantly higher FHB infection and DON concentrations than the late fungicide treatments. However, septoria infection was significantly inhibited by the early fungicide treatments, more than the late fungicide treatments at ZGS 71 (Table 3).

‘AC Westech’ 6-row Spring Barley

Experimental treatments had significant effects on yield, TKW, DON concentrations and blotch severity at ZGS 85 (Table 2). ‘AC Westech’ treated with trt14 and trt16 yielded the highest (Figure 3). However, their yields did not differ significantly from trt13. Untreated ‘AC Westech’ yielded significantly lower than all the other treatments. Trt14 and trt16 had the highest TKW but was not significantly higher than trt8, trt10 and trt12. The untreated check had a significantly lower TKW than all the other treatments. The check had significantly more blotch infection than the early and double fungicide treatments. ‘AC Westech’ had significantly higher DON

concentrations when treated with trt9 and trt13 than all the other treatments. Trt2 and trt11 are the only two treatments that showed significantly lower DON concentrations than the check (Figure 3).

Compared to the check, all the fungicide application timings resulted in significantly greater yield and TKW (Table 3). Early and double fungicide treatments significantly reduced blotch severity. Late fungicide treatments significantly decreased DON concentrations but early fungicide treatments significantly increased DON concentrations (Table 3).

Double fungicide treatments had significantly higher yield and TKW than both the early and late fungicide treatments (Table 3). They also showed significantly lower DON concentrations than the early fungicide treatments and lower blotch severity than the late fungicide treatments. Early fungicide treatments had significantly higher yield, higher DON concentrations and lower blotch severity than the late fungicide treatments (Table 3).

DISCUSSION

In the test years for this study, the severity level of powdery mildew was in the range of no infection to light severity in 'AC Helena' spring wheat and 'AC Westech' spring barley. The winter wheat variety, 'Emmit', is the only variety that showed moderate powdery mildew infection during the period 2010 to 2012. Both septoria and FHB were observed to be more severe and more frequently occurred compared to powdery mildew. Leaf and/or net blotch was observed every year on barley Scald infection, however, did not occur in 2011 and was light in 2012. It showed moderate to high infection in 2010. However, no fungicide showed good efficacy on its control. Table 3 shows that double fungicide treatments provided more crop protection against the assessed diseases through the whole cropping season than the single applied fungicides. Single applied fungicides, although exhibiting their efficacy to the diseases

on some growth stages, could not maintain the protection for the whole cropping season.

DON concentrations were very low in winter wheat 'Emmit'. The DON concentration was much higher in spring wheat 'AC Helena' ranging from 2.1 ppm to 6.3 ppm (exceeding the 2 ppm limitation for human consumption). The lower DON concentrations were not found to be correlated with the lower FHB scores in AC Helena; e.g., early fungicide treatments, trt5 and trt13, had significantly less apparent FHB infection than the check. However, their DON levels were significantly higher than that of the check. A similar result was also reported in Martin and Johnston's (1982) research conducted in the Atlantic Region. They found the reduced FHB severity did not result in a reduction of the DON concentration. In the barley variety AC Westech, although there was no visual sign of FHB infection, DON was detected. Trt9 and trt13 applied at ZGS 39 resulted in significantly higher DON levels (1.3 and 1.7 ppm, respectively) over the other treatments. These early applied fungicides at ZGS 39, although they were not labeled for FHB control (Anonymous, 2009; Anonymous, 2015b; Anonymous 2015c), showed some efficacy in FHB control. However, their application increased the DON concentration (Table 3). In Hutcheon and Jordon's (1992) research, they demonstrated that fungicides applied at ZGS 39 were less effective in reducing DON concentrations compared to the fungicides applied at later growth stages between ZGS 59 to ZGS 70. Besides the timing effect of fungicide application, different fungicides were reported to have varying influences on different FHB pathogens (Siranidou and Buchenauer, 2001; Magan et al. 2002). Simpson et al. (2001) found that the application of the triazole-based fungicide (fungicide class 3), tebuconazole, effectively reduced the colonization and DON concentrations caused by the toxigenic *Fusarium* spp., such as *F. culmorum* and *F. avenaceum*. The application of strobilurin-based fungicide (fungicide class 11), azoxystrobin, had little effect on the *Fusarium* spp., but significantly inhibited the growth of the

non-toxicogenic pathogen *Microdochium nivale* which can develop undistinguishable symptoms from the *Fusarium* spp. without producing mycotoxins. The inhibition of *M. nivale* resulted in a greater colonization by the toxicogenic *Fusarium* spp. and thus the increased DON concentrations in grains (Simpson et al. 2001). Among the triazole-based fungicides - tebuconazole, metconazole, prothioconazole, propiconazole and prothioconazole+tebuconazole, Paul et al. (2008) found that propiconazole was the least effective while prothioconazole+tebuconazole was the most effective on both FHB infection and DON level. The similar fungicide-dependent effects were also observed in our experiment. The highest DON concentrations were found in AC Helena and AC Westech when trt13, which contains only a strobilurin-based active ingredient in its formula, was applied. The trt5 containing propiconazole and the trt9 containing propiconazole+trifloxystrobin reduced FHB infection. However, their application had no or, in some cases, negative effects on DON concentration compared to the check. Therefore, the fungicide application timing associated with the types of chemicals used in the formula were considered to be the two reasons for the lack of efficacy observed in the early applied fungicides on FHB and DON level.

Fungicides applied at ZGS 39, ZGS 60 or both growth stages, in general, significantly increased the yield when compared to the check in all tested varieties. The yield increase was observed to be more dramatic in spring cereal varieties than in the winter variety. When treated with double fungicide treatments, the average yield was increased by 1.2 t ha^{-1} (approximately 36% increase relative to check) in spring wheat “AC Helena” and 1.4 t ha^{-1} (approximately 52% increase relative to check) in spring barley. Average yield increase of the winter wheat variety – ‘Emmit’ was relatively low at 0.6 t ha^{-1} (approximately 10% increase over the check) compared to the yield increases of the spring cereal varieties. In all cases when there were significant

differences among fungicide application timings on the yield and TKW, double fungicide treatments always resulted in the significantly greater yield and TKW than either of the single fungicide timings.

To provide the best crop protection against the range of the cereal diseases while ensuring the best seed quality, double fungicide applications are recommended. Single applications at either ZGS 39 or ZGS 60 are not sufficient to provide the season-long plant protection against the disease. Among the double fungicide treatments, trt16 was the only treatment having consistently higher yield and TKW while having the lower ratings on the diseases and DON levels across the varieties and years. The cost of this treatment combination in the test years was approximately \$100 ha⁻¹ so would have to be considered by the farmer in relation to expected yield increases and commodity costs. With that proviso in mind, trt16 (pyraclostrobin early and prothioconazole/tebuconazole late) is the treatment we recommend to use for cereals in Atlantic region of Canada.

CONCLUSION

Among the assessed crop diseases, FHB and septoria in wheat varieties were more frequently observed than powdery mildew; FHB and blotch in barley occurred more frequently than scald in this experiment. One fungicide application at either ZGS 39 or ZGS 60 was not enough to achieve a high yield with good seed qualities. In fact, the use of a fungicide at ZGS 39 may increase the potential for DON development later. To further improve the yield and quality through the control of leaf and head diseases, pyraclostrobin applied at ZGS 39 and prothioconazole/tebuconazole applied at ZGS 60 was found to be the best fungicide combination for winter wheat, spring wheat and barley varieties tested.

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Table 1. Fungicide Treatments

Treatment Number	Fungicide applied at ZGS 39	Fungicide applied at ZGS 60
1	check	check
2	check	prothioconazole
3	check	metaconazole
4	check	prothioconazole/tebuconazole
5	propiconazole	check
6	propiconazole	prothioconazole
7	propiconazole	metaconazole
8	propiconazole	prothioconazole/tebuconazole
9	propiconazole/trifloxystrobin	check
10	propiconazole/trifloxystrobin	prothioconazole
11	propiconazole/trifloxystrobin	metaconazole
12	propiconazole/trifloxystrobin	prothioconazole/tebuconazole
13	pyraclostrobin	check
14	pyraclostrobin	prothioconazole
15	pyraclostrobin	metaconazole
16	pyraclostrobin	prothioconazole/tebuconazole

Table 2. Probability values (level of significance) of fungicide treatments' effect on yield, thousand kernel weight (TKW), fusarium head blight (FHB), powdery mildew severity at ZGS 60 (M60), ZGS 71 (M71) and ZGS 85 (M85), septoria severity at ZGS 60 (S60), ZGS 71 (S71) and ZGS 85 (S85), deoxynivalenol (DON), blotch severity at ZGS 85 (B85) and scald severity at ZGS 85 (Sc85) during 2010 to 2012 on various cereal varieties

Variety	Year	Measured Variables											
		Yield	TKW	FHB	M60	M71	M85	S60	S71	S85	DON	B85	Sc85
Emmit	2010-2012	<0.001	0.002	<0.001	0.213	<0.001	0.543 ^a	0.046	0.003	<0.001	NA	NA	NA
AC Helena	2010-2012	<0.001	<0.001	<0.001 ^b	NA	NA	NA	0.045 ^a	<0.001 ^c	<0.001 ^d	0.001	NA	NA
AC Westech	2010-2012	<0.001	<0.001	NA	NA	NA	NA	NA	NA	NA	<0.001	<0.001	0.949 ^a

a: Only 2010's data were analyzed due to the low ratings from the other years

b: Only 2010 and 2011's data were analyzed due to the low ratings from year 2012

c: Only 2011's data were analyzed due to the low ratings from the other years

d: Only 2011 and 2012's data were analyzed due to the low ratings from year 2010

Table 3. Mean difference ($\bar{D}$) and corresponding probability values (P) of contrast comparisons between the effects of the check (CK; no fungicides), early (E; ZGS 39), late (L; ZGS 60) and double (DB; ZGS 39 and ZGS 60) fungicide treatments on yield, thousand kernel weight (TKW), fusarium head blight (FHB), powdery mildew severity at ZGS 71 (M71) and ZGS 85 (M85), septoria severity at ZGS 71 (S71) and ZGS 85 (S85), deoxynivalenol (DON), blotch severity at ZGS 85 (B85) and scald severity at ZGS 85 (Sc85)

Response	Contrast	Emmit		AC Helena		AC Westech	
		$\bar{D}$	P	$\bar{D}$	P	$\bar{D}$	P
Yield (t ha ⁻¹)	E vs. CK	0.2	0.052	0.7	<0.001	1.1	<0.001
	L vs. CK	0.4	0.004	0.8	<0.001	0.7	<0.001
	DB vs. CK	0.6	<0.001	1.2	<0.001	1.4	<0.001
	E vs. L	- 0.2	0.027	- 0.1	0.058	0.4	<0.001
	E vs. DB	- 0.4	<0.001	- 0.5	<0.001	- 0.3	<0.001
	L vs. DB	- 0.2	0.030	- 0.4	<0.001	- 0.7	<0.001
TKW (g)	E vs. CK	0.4	0.507	4.1	<0.001	5.0	<0.001
	L vs. CK	0.7	0.286	4.7	<0.001	4.1	<0.001
	DB vs. CK	1.8	0.002	6.9	<0.001	7.2	<0.001
	E vs. L	- 0.2	0.573	- 0.6	0.186	0.9	0.077
	E vs. DB	- 1.4	<0.001	- 2.8	<0.001	- 2.2	<0.001
	L vs. DB	- 1.2	0.001	- 2.2	<0.001	- 3.1	<0.001
FHB (%)	E vs. CK	- 11.7	<0.001	- 20.8	<0.001	NA	NA
	L vs. CK	- 13.2	<0.001	- 32.6	<0.001	NA	NA
	DB vs. CK	- 15.6	<0.001	- 38.0	<0.001	NA	NA
	E vs. L	1.5	0.314	11.9	<0.001	NA	NA
	E vs. DB	3.9	0.001	17.2	<0.001	NA	NA
	L vs. DB	2.4	0.046	5.4	0.002	NA	NA
M71 (0-9)	E vs. CK	- 0.9	0.004	NA	NA	NA	NA
	L vs. CK	- 0.6	0.074	NA	NA	NA	NA
	DB vs. CK	- 1.2	<0.001	NA	NA	NA	NA
	E vs. L	0.4	0.117	NA	NA	NA	NA
	E vs. DB	0.3	0.183	NA	NA	NA	NA
	L vs. DB	0.6	0.001	NA	NA	NA	NA

M85 (0-9)	E vs. CK	- 0.4	0.544	NA	NA	NA	NA
	L vs. CK	- 0.5	0.443	NA	NA	NA	NA
	DB vs. CK	- 1.1	0.062	NA	NA	NA	NA
	E vs. L	0.1	0.849	NA	NA	NA	NA
	E vs. DB	0.7	0.083	NA	NA	NA	NA
	L vs. DB	0.6	0.100	NA	NA	NA	NA
S71 (0-9)	E vs. CK	- 1.0	0.001	- 0.8	0.101	NA	NA
	L vs. CK	- 0.5	0.095	0.2	0.712	NA	NA
	DB vs. CK	- 1.2	<0.001	- 1.9	<0.001	NA	NA
	E vs. L	- 0.5	0.026	- 0.9	0.006	NA	NA
	E vs. DB	0.2	0.263	1.2	<0.001	NA	NA
	L vs. DB	0.7	<0.001	2.1	<0.001	NA	NA
S85 (0-9)	E vs. CK	- 1.3	0.001	- 1.6	0.092	NA	NA
	L vs. CK	- 1.2	0.002	- 2.5	0.004	NA	NA
	DB vs. CK	- 1.9	<0.001	- 3.7	<0.001	NA	NA
	E vs. L	- 0.1	0.754	0.9	0.087	NA	NA
	E vs. DB	0.7	0.003	2.1	<0.001	NA	NA
	L vs. DB	0.8	<0.001	1.2	0.002	NA	NA
DON (ppm)	E vs. CK	NA	NA	1.8	0.009	0.6	0.003
	L vs. CK	NA	NA	- 0.3	0.689	- 0.5	0.015
	DB vs. CK	NA	NA	0.4	0.554	- 0.3	0.082
	E vs. L	NA	NA	2.1	<0.001	1.1	<0.001
	E vs. DB	NA	NA	1.4	<0.001	0.9	<0.001
	L vs. DB	NA	NA	- 0.7	0.106	- 0.2	0.134
B85 (0-9)	E vs. CK	NA	NA	NA	NA	- 1.5	<0.001
	L vs. CK	NA	NA	NA	NA	- 0.5	0.107
	DB vs. CK	NA	NA	NA	NA	- 1.6	<0.001
	E vs. L	NA	NA	NA	NA	- 1.0	<0.001
	E vs. DB	NA	NA	NA	NA	0.2	0.380
	L vs. DB	NA	NA	NA	NA	1.1	<0.001
Sc85	E vs. CK	NA	NA	NA	NA	- 0.4	0.622
	L vs. CK	NA	NA	NA	NA	0	1
	DB vs. CK	NA	NA	NA	NA	- 0.3	0.692

(0-9)	E vs. L	NA	NA	NA	NA	- 0.4	0.486
	E vs. DB	NA	NA	NA	NA	- 0.1	0.820
	L vs. DB	NA	NA	NA	NA	0.3	0.531

“ $\overline{D}$ ” = mean difference of the effects between two fungicide treatments in the contrast. The difference is calculated as the mean of the former fungicide treatment relative to the mean of the later fungicide treatment in the contrast. For instance, in the contrast of “E vs. CK” on yield, the difference ($\overline{D}$) = mean yield of “E” – mean yield of “CK”

For Review Only

Figure 1. Effects of treatments on various responding variables for winter wheat variety – ‘Emmit’. Means with the same letter are not significantly different from each other at a 0.05 significance level

Figure 2. Effects of treatments on various responding variables for spring wheat variety – ‘AC Helena’. Means with the same letter are not significantly different from each other at a 0.05 significance level

Figure 3. Effects of treatments on various responding variables for spring barley variety – ‘AC Westech’. Means with the same letter are not significantly different from each other at a 0.05 significance level