

EVALUATING THE EFFECTS OF FUNGICIDE ON SILAGE CORN QUALITY AND HYGIENE

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Introduction

In Wisconsin, dairy cows eat on average 100 pounds of feed a day, about half of which typically is corn silage. Corn silage is an extremely important part of a dairy cow's diet and provides much of the needed calorie and nutrition input. When making corn silage, the whole plant, including the stalk and ear is chopped and put into bunkers. In these large, cement bunkers the chopped corn is packed down tightly and covered in order to begin fermentation as quickly as possible. Ensiling through fermentation is the best way to preserve the quality and nutrition of feed and prevent degradation. Plant diseases that originate in the field, such as stalk rots and foliar blights, can reduce overall yield and result in non-optimal moisture content of silage corn. This can influence proper bunker packing and ensiling resulting in low quality feeds and secondary problems.

Gibberella ear rot and stalk rot are caused by *Fusarium graminearum* (White, 2016). Beyond reducing the quality of silage, *F. graminearum* produces the mycotoxin deoxynivalenol, also known as DON or vomitoxin. Mycotoxins are toxic metabolites produced by some fungal pathogens during their secondary metabolisms that negatively impact the consumer. DON consumption is associated with low weights, feed refusal, and vomiting in livestock. There are estimated ranges of allowable levels of DON contamination within livestock feed, and milk production in dairy cows has been shown to be negatively impacted with as little as 1 ppm of DON (Goeser, 2015). Currently, DON levels are typically associated with ear rots in grain corn, but there is no understanding of the extent of DON accumulation throughout the rest of the corn plant (White, 2016). Because corn silage uses the entire plant, it is important to understand the impact of fungicide treatments on *F. graminearum* and DON accumulation throughout the entire corn plant. Thus, the objectives of our study were as follows:

1. Understand the impact of fungicide treatments on deoxynivalenol production by *Fusarium graminearum* in silage corn.
2. Understand the location of deoxynivalenol accumulation within the ear and stalk portions of the corn plant.

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Materials and Methods

Fungicide Trial

The fungicide trial occurred in 2018 and 2019 at the Arlington Agricultural Research Station (AARS) in Arlington, Wisconsin. Two brown midrib (BMR) silage corn hybrid varieties, P0956AMX and F2F627 were planted at a rate of 35,000 seeds per acre in 15' x 20' six-row plots. Fungicide treatments were applied between V6 and R2 growth stages (Table 1). Foliar disease and ear rot ratings were taken throughout the growing season for multiple diseases including Gibberella ear rot, gray leaf spot, northern corn leaf blight, tar spot, and southern rust. Lodging scores and a push test were also completed.

A small-plot silage chopper harvested the center two rows of each plot and dry-matter yield was determined. Samples were taken for forage quality and DON analyses at Rock River Laboratory, Inc. in Watertown, WI. Factors such as Dry Matter yield, moisture content, crude protein %, aNDF%, starch %, ash %, TTNDFD %, and more were measured for each silage sample. TTNDFD % is a quantitative predictor of fiber digestion (Combs, 2015).

Table 1. Fungicide treatments, timings, and application rates for 2018 and 2019 silage corn fungicide trials in Arlington, WI.

Application Time	Treatment	Year	
		2018	2019
	Non-Treated Check	x	x
V6	Miravis Neo 13.7 FL OZ/A;NIS 0.25%	x	x
V6 R1	Miravis Neo 13.7 FL OZ/A V6;NIS 0.25 % V/V V6 Miravis Neo 13.7 FL OZ/A R1		x
V14	Miravis Neo 13.7 FL OZ/A V12-V14	x	x
R1	Proline 5.7 FL OZ/A	x	x
	Headline AMP 14.4 FL OZ/A	x	x
	Delaro 8 FL OZ/A	x	x
	Miravis Neo 13.7 FL OZ/A	x	x
	Miravis Ace 13.7 FL OZ/A	x	x
	Topguard 10 FL OZ/A	x	x
	Lucento 5 FL OZ/A	x	x
R2	Miravis Neo 13.7 FL OZ/A	x	x
	Proline 5.7 FL OZ/A	x	
	Headline AMP 14.4 FL OZ/A	x	
	Delaro 8 FL OZ/A	x	

Partitioned Sample Experiment

Samples were hand harvested from rows 2 and 5 of select treatments of the silage fungicide trial at AARS. The selected treatments included the non-treated check, Headline AMP applied at 14.4 FL OZ/A at R1 (silking), and Proline at 5.7 FL OZ/A at R1. Five plants were randomly selected from each plot and stalk and ear (including husk and cobb) portions were

separated, chopped, and then dried down. Deoxynivalenol concentrations in parts per million (ppm) were determined using the RIDASCREEN® DON R5906 kit from R-Biopharm, Inc. Absolute RT-PCR was used to determine the quantity of *Fusarium graminearum* DNA (pg/ng) within the sample of plant material.

Results and Discussion

In the whole plot silage samples, we found no significant differences in yield or digestibility (TTNDFD) for any fungicide treatment for either hybrid in either year. These results along with disease severity scores and DON contamination can be found in Tables 2-5 for each hybrid in both years. Due to environmental conditions, there was much higher disease pressure in 2018 than 2019. In 2018, ear rot severity was very high and DON concentrations reached up to 30 ppm (Table 3), which is 30 times the recommended amount for dairy feed. In 2019, the maximum DON concentration was 2.2 ppm (Table 4) and averaged below 1 ppm for both hybrids. For F2F627 in 2018, 4 treatments were found to significantly decrease DON contamination and 1 separate treatment was found to significantly decrease ear rot severity levels. No significant decreases of DON or ear rot severity were observed in P0956AMX in 2018. In 2019, 3 different treatments significantly decreased DON concentrations in P0956AMX, while there were no significant reductions in ear rot severity in either hybrid or DON in F2F627. The ear rot severity scores and DON concentrations were not correlated for either hybrid in each year of the trial. This lack of correlation suggests another source of DON contamination, in addition to the ear rot phase, is responsible for the DON levels observed in finished feed.

In the partitioned samples, both DON and *F. graminearum* DNA was detected in all ear and stalk samples. This shows the ability for DON to accumulate throughout the plant. The majority of the accumulation of DON varied between the ear or stalk parts for each hybrid by year. We found higher DON levels in the 2018 F2F627 ear samples compared to the stalk samples for all treatments tested (*data not shown*). In the 2018 P0956AMX samples, DON accumulated in the stalk to a higher level than the ear. In 2019, again likely due to environmental conditions, there were lower levels of DON overall with the highest being found in the non-treated stalk samples for both hybrids. *F. graminearum* fungal accumulation in 2018 followed a similar trend to DON accumulation in F2F627. However, in P0956AMX, the location of *F. graminearum* fungal accumulation did not correspond with DON accumulation with more fungal DNA being detected in the ear parts and more DON being detected in the stalks.

This work shows that there are differential responses between BMR hybrids to fungicide treatments. Environmental conditions can contribute greatly to disease development and DON accumulation and impact where DON is found in silage corn plants. The work also demonstrates that DON can be found in all parts of the corn plant. While more work is needed to understand how DON accumulates in both the ear and stalk, breeding efforts should focus on slowing the growth of *F. graminearum* and accumulation of DON in both stalks and ears.

Table 2. Northern corn leaf blight severity, tar spot severity, ear rot severity, dry matter yield, TTNDFD, and deoxynivalenol (DON) for P0956AMX BMR corn hybrid treated with fungicide or not treated with fungicide in Wisconsin, 2018.

Treatment and rate/A (growth stage at application)	Northern Corn Leaf Blight Severity(%) ^{z, u}	Tar Spot Severity (%) ^{y, u}	Ear Rot Severity (%) ^x	Yield (tons dry matter/a)	TTNDFD (%) ^w	DON (ppm) ^v
Non-Treated Check	25.0 a	3.8 a	2.1	12.9	34.9	9.4
Miravis Neo 2.5SE 13.7 fl oz (V12)	16.3 bc	2.1 b-d	2.9	13.6	36.2	7.7
Miravis Neo 2.5SE 13.7 fl oz (V6) ^t	17.5 c-e	1.8 b-d	1.4	12.6	37.3	8.4
Topguard 1.04SC 10 fl oz (R1)	6.1 e	1.4 b-d	4.9	11.9	38.5	12.9
Proline 480SC 5.7 fl oz (R1)	14.3 b-d	1.2 b-d	3.1	11.8	36.8	8.5
Miravis Neo 2.5SE 13.7 fl oz (R2)	8.1 de	1.2 b-d	1.6	12.1	36.7	9.8
Miravis Ace 5.2SC 13.7 fl oz (R1)	11.3 c-e	1.0 cd	3.3	12.2	37.8	9.8
Proline 480SC 5.7 fl oz (R2)	11.3 c-e	1.0 cd	1.4	12.8	36.3	10.0
Delaro 325SC 8 fl oz (R1)	11.8 c-e	1.0 cd	2.1	11.9	36.7	10.5
Lucento 4.17SC 5 fl oz (R1)	8.0 de	0.8 cd	1.5	11.8	37.1	8.5
Headline AMP 1.68SC 14.4 fl oz (R1)	14.3 b-d	0.8 cd	1.4	13.0	35.9	11.9
Delaro 325SC 8 fl oz (R2)	10.5 c-e	0.6 d	2.1	11.7	38.5	8.2
Headline AMP 1.68SC 14.4 fl oz (R2)	13.0 c-e	0.6 d	1.0	12.0	37.1	11.9
Miravis Neo 2.5SE 13.7 fl oz (R1)	9.8 b-e	0.6 d	1.0	12.5	36.4	17.9
Fisher's LSD ($\alpha=0.05$)	7.4	1.1	ns ^s	ns ^s	ns ^s	ns ^s

^zNorthern corn leaf blight severity was visually assessed as the average % ear leaf symptomatic per plot with the aid of a standard area diagram; means for each plot were used in the analysis

^yTar spot severity was visually assessed as the average % ear leaf symptomatic per plot with the aid of a standard area diagram; means for each plot were used in the analysis

^xEar rot severity assessed visually on 5 ears per plot

^wTotal-Tract Neutral Detergent Fiber Digestibility

^vDeoxynivalenol (DON) content were analyzed for each plot; means for each plot were used in the analysis

^uMeans followed by the same letter are not significantly different based on Fisher's Least Significant Difference (LSD; $\alpha=0.05$)

^tTreatments including the non-ionic surfactant Induce 90SL at 0.25 %v/v

^sns = not significant ($\alpha=0.05$)

Table 3. Northern corn leaf blight severity, tar spot severity, ear rot severity, dry matter yield, TTNDFD, and deoxynivalenol (DON) for F2F627 BMR corn hybrid treated with fungicide or not treated with fungicide in Wisconsin in 2018.

Treatment and rate/A (growth stage at application)	Northern Corn					
	Leaf Blight Severity (%) ^{z,u}	Tar Spot Severity (%) ^{y,u}	Ear Rot Severity (%) ^x	Yield (tons dry matter/a)	TTNDFD (%) ^w	DON (ppm) ^v
Miravis Neo 2.5SE 13.7 fl oz (V12)	27.5 c-e	11.3 a	4.6	11.6	36.2	18.6
Non-Treated Check	62.5 a	10.5 ab	8.8	11.0	38.7	21.2
Proline 480SC 5.7 fl oz (R2)	27.5 c-e	8.6 a-c	6.5	10.4	39.4	10.7
Proline 480SC 5.7 fl oz (R1)	31.3 c-f	7.4 a-d	10.4	11.0	38.5	13.2
Miravis Neo 2.5SE 13.7 fl oz (R1)	21.3 de	6.9 a-d	11.1	11.1	39.5	17.2
Miravis Ace 5.2SC 13.7 fl oz (R1)	42.5 bc	6.3 b-f	7.7	11.7	39.7	15.7
Lucento 4.17SC 5 fl oz (R1)	18.8 ef	5.8 b-e	4.5	12.2	37.5	18.0
Topguard 1.04SC 10 fl oz (R1)	23.8 de	5.6 c-e	4.8	10.7	38.1	15.1
Miravis Neo 2.5SE 13.7 fl oz (R2)	15.0 e	5.5 c-e	7.9	10.7	39.9	30.3
Miravis Neo 2.5SE 13.7 fl oz (V6) ^l	50.0 ab	4.9 c-e	10.0	11.0	37.0	12.0
Delaro 325SC 8 fl oz (R1)	22.5 de	4.3 c-e	12.9	11.2	37.8	17.7
Headline AMP 1.68SC 14.4 fl oz (R1)	36.3 b-d	2.8 d-f	14.2	10.6	38.7	18.7
Delaro 325SC 8 fl oz (R2)	28.8 c-e	2.0 ef	9.7	10.5	37.1	12.7
Headline AMP 1.68SC 14.4 fl oz (R2)	17.5 ef	1.4 e	18.4	11.5	40.9	14.9
Fisher's LSD ($\alpha=0.05$)	15.9	4.8	ns ^s	ns ^s	ns ^s	ns ^s

^zNorthern corn leaf blight severity was visually assessed as the average % ear leaf symptomatic per plot with the aid of a standard area diagram; means for each plot were used in the analysis

^yTar spot severity was visually assessed as the average % ear leaf symptomatic per plot with the aid of a standard area diagram; means for each plot were used in the analysis

^xEar rot severity assessed visually on 5 ears per plot

^wTotal-Tract Neutral Detergent Fiber Digestibility

^vDeoxynivalenol (DON) content were analyzed for each plot; means for each plot were used in the analysis

^uMeans followed by the same letter are not significantly different based on Fisher's Least Significant Difference (LSD; $\alpha=0.05$)

^lTreatments including the non-ionic surfactant Induce 90SL at 0.25 %v/v

^sns = not significant ($\alpha=0.05$)

Table 4. Southern rust severity, tar spot severity, ear rot severity, dry matter yield, TTNDFD, and deoxynivalenol (DON) for P0956AMX BMR corn hybrid treated with fungicide or not treated with fungicide in Wisconsin, 2019.

Treatment and rate/A (growth stage at application)	Southern Rust Severity (%) ^{z, u}	Tar Spot Severity (%) ^{y, u}	Ear Rot Severity (%) ^x	Yield (tons dry matter/a)	TTNDFD (%) ^w	DON (ppm) ^{v, u}
Non-Treated Check	2.8 a	0.8 b	1.5	10.0	43.6	1.6 a
Miravis Neo 2.5SE 13.7 fl oz (V6) ^l	1.4 bc	2.2 a	10.3	9.1	46.1	1.3 ab
Miravis Neo 2.5SE 13.7 fl oz (V6) ^l						
Miravis Neo 2.5SE 13.7 fl oz (R1)	2.1 ab	0.9 b	1.2	10.3	45.3	2.2 a
Miravis Neo 2.5SE 13.7 fl oz (V14)	2.2 ab	0.9 b	3.3	9.0	47.0	0.2 d
Proline 480SC 5.7 fl oz (R1)	1.9 ab	0.6 b	0.3	9.7	47.1	0.5 a-d
Topguard 1.04SC 10 fl oz (R1)	2.9 a	0.8 b	1.1	10.2	46.3	1.3 ab
Delaro 325SC 8.0 fl oz (R1)	1.2 bc	0.7 b	1.5	10.2	44.0	0.3 b-d
Headline AMP 1.68SC 14.4 fl oz (R1)	2.4 ab	0.7 b	0.7	10.5	46.1	0.2 cd
Lucento 4.17SC 5.0 fl oz (R1)	0.8 c	0.6 b	0.6	9.4	46.4	1.0 a-c
Miravis Ace 5.2SC 13.7 fl oz (R1)	1.8 ab	1.0 b	1.3	9.5	45.1	0.6 a-d
Miravis Neo 2.5SE 13.7 fl oz (R1)	2.1 ab	0.8 b	2.7	10.3	46.1	0.6 a-d
Miravis Neo 2.5SE 13.7 fl oz (R2)	2.0 ab	0.6 b	1.1	9.8	45.8	1.6 a
<i>P-value</i>	<0.05	<0.05	ns ^s	ns ^s	ns ^s	<0.05

^zSouthern rust severity was visually assessed as the average % ear leaf symptomatic per plot with the aid of a standard area diagram; means for each plot were used in the analysis.

^yTar spot severity was visually assessed as the average % ear leaf symptomatic per plot with the aid of a standard area diagram; means for each plot were used in the analysis.

^xEar rot severity assessed visually on 5 ears per plot.

^wTotal-Tract Neutral Detergent Fiber Digestibility

^vDeoxynivalenol (DON) content were analyzed for each plot; means for each plot were used in the analysis.

^uMeans followed by the same letter are not significantly different based on Fisher's Least Significant Difference (LSD; $\alpha=0.05$).

^lTreatments including the non-ionic surfactant Induce 90SL at 0.25 %v/v

^sns = not significant ($\alpha=0.05$)

Table 5. Southern rust severity, tar spot severity, ear rot severity, dry matter yield, TTNDFD, and deoxynivalenol (DON) for F2F627 BMR corn hybrid treated with fungicide or not treated with fungicide in Wisconsin, 2019.

Treatment and rate/A (growth stage at application)	Southern Rust Severity (%) ^{z, u}	Tar Spot Severity (%) ^y	Ear Rot Severity (%) ^x	Yield (tons dry matter/a)	TTNDFD (%) ^w	DON (ppm) ^v
Non-Treated Check	9.25 ab	0.91	0.05	9.09	47.32	0.12
Miravis Neo 2.5SE 13.7 fl oz (V6) ^t	7.60 a-c	0.82	0.05	8.24	46.37	0.20
Miravis Neo 2.5SE 13.7 fl oz (V6) ^t						
Miravis Neo 2.5SE 13.7 fl oz (R1)	4.61 b-d	0.83	0.05	8.80	47.77	0.08
Miravis Neo 2.5SE 13.7 fl oz (V14)	3.08 d	0.85	0.00	8.79	47.88	0.02
Miravis Neo 2.5SE 13.7 fl oz (R1)	2.94 d	0.99	0.25	9.24	45.72	0.08
Miravis Ace 5.2SC 13.7 fl oz (R1)	6.58 a-c	0.79	0.00	9.05	48.22	0.07
Lucento 4.17SC 5.0 fl oz (R1)	0.84 e	0.87	0.50	8.75	48.37	0.22
Topguard 1.04SC 10 fl oz (R1)	9.72 a	0.65	0.05	8.57	47.21	0.09
Proline 480SC 5.7 fl oz (R1)	3.97 cd	1.08	0.25	8.67	48.96	0.05
Headline AMP 1.68SC 14.4 fl oz (R1)	5.10 a-d	0.79	0.30	9.70	47.02	0.02
Delaro 325SC 8.0 fl oz (R1)	3.91 cd	0.95	0.30	7.91	49.46	0.01
Miravis Neo 2.5SE 13.7 fl oz (R2)	5.24 a-d	0.67	0.50	8.58	48.94	0.08
<i>P-value</i>	<0.0001	ns ^s	ns ^s	ns ^s	ns ^s	ns ^s

^zSouthern rust severity was visually assessed as the average % ear leaf symptomatic per plot with the aid of a standard area diagram; means for each plot were used in the analysis.

^yTar spot severity was visually assessed as the average % ear leaf symptomatic per plot with the aid of a standard area diagram; means for each plot were used in the analysis.

^xEar rot severity assessed visually on 5 ears per plot.

^wTotal-Tract Neutral Detergent Fiber Digestibility

^vDeoxynivalenol (DON) content were analyzed for each plot; means for each plot were used in the analysis.

^uMeans followed by the same letter are not significantly different based on Fisher's Least Significant Difference (LSD; $\alpha=0.05$).

^tTreatments including the non-ionic surfactant Induce 90SL at 0.25 %v/v

^sns = not significant ($\alpha=0.05$)

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