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Corn Yield Response to Pyraclostrobin with Foliar Fertilizers

John Shetley[1], Kelly A. Nelson[2], William G. Stevens[3], David Dunn[3], Bruce Burdick[4], Peter P. Motavalli[1], James T. English[5], & Christopher J. Dudenhoeffer[2]

[1] Department of Soil, Environmental and Atmospheric Sciences, University of Missouri, Columbia, MO, USA

[2] Division of Plant Sciences, University of Missouri, Novelty, MO, USA

[3] Division of Plant Sciences, University of Missouri, Portageville, MO, USA

[4] Hundley-Whaley Center, University of Missouri, Albany, MO, USA

[5] Division of Plant Sciences, University of Missouri, Columbia, MO, USA

Correspondence: Kelly A. Nelson, University of Missouri, Lee Greenley Jr. Memorial Research Center, Novelty, MO 63460, USA. Tel: 1-660-739-4410. E-mail: nelsonke@missouri.edu

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Abstract

Strobilurin fungicides, including pyraclostrobin, protect many crops from several fungal pathogens and create opportunities to increase plant health and yields. However, corn (*Zea mays* L.) and many other plants' physiological responses to pyraclostrobin include increases in processes that require nutrients. By applying foliar fertilizers, growers can adjust their nutrient-management strategy based on the plant's reaction to pyraclostrobin. Identifying plants' increased nutrient demands and meeting them with a foliar fertilizer at the time of fungicide application (tasselling) could increase yields. This study evaluated effects of foliar-applied pyraclostrobin at 0.11 kg ha^{-1} a.i. with or without 13 commonly available foliar fertilizers on yield, tissue macro- and micronutrient concentrations, severity of disease, and grain quality. Field research occurred at three University of Missouri research centers from 2008-2009. One foliar fertilizer, 0-0-30-0, caused up to 20% crop injury. Diseases affected plants in all six site-years, but overall severity was low (≤ 2%) and likely did not impact crop performance. Pyraclostrobin increased ear leaf B and Cu concentrations over all site-years seven days after treatment, and decreased N concentrations at one site-year. Grain yields increased 5% at two research sites with pyraclostrobin, and one location had increased grain moisture and grain oil at harvest. One foliar fertilizer, 30-0-0-0, increased grain yields by 10% at two sites compared to the non-treated control. However, foliar fertilizers showed no observable effects on grain quality characteristics, and none of the foliar fertilizers negatively impacted grain yield.

Keywords: corn, disease, foliar fertilizer, fungicide, pyraclostrobin, strobilurin

1. Introduction

High-yield corn production systems integrate fungicide applications to maximize plants' photosynthetic efficiency. A fungicide class first available in 1996 includes the strobilurin fungicides (Bartlett et al., 2002). Yield has increased up to 1.5 Mg ha^{-1} in corn hybrids susceptible to foliar pathogens and 0.38 Mg ha^{-1} in tolerant hybrids with a strobilurin fungicide such as pyraclostrobin (carbamic acid, [2,[[[1-(4-chlorophenyl)-1H-pyrazol-3-yl]oxy]methyl]phenyl]methoxy-, methyl ester) (Newman, 2009; Bartlett et al., 2002). In wheat, strobilurins increased grain yields over non-treated plants and from 1 to 11% over other classes of fungicides (Oerke et al., 2001).

Several factors account for corn yield increases with pyraclostrobin. The most direct reason is the reduction in numerous foliar diseases (Bartlett et al., 2002). Worldwide, losses in corn production from fungal and bacterial pathogens alone were estimated to be about 9% from 2001-2003 (Oerke, 2006). Fungal pathogens, including grey leaf spot (*Cercospora zea-maydis*), caused yield losses above 44% in Iowa, 69% in Virginia and complete crop failure on occasion (Ward & Nowell, 1998; Ward et al., 1999). Fungal pathogens colonize leaf tissue and often damage the leaves themselves. Leaf necrosis can reduce plants' photosynthetic area and efficiency. Fungal infections also reduce overall physiological efficiency by redirecting plant products and photoassimilates from producing and filling grain to fungal development, growth, metabolism, reproduction, plant defense reactions, and respiration of the infected tissue (Venancio et al., 2003). The goal for most fungicide applications is to suppress or eliminate foliar infections, especially in the upper canopy, in order to maintain photosynthetic efficiency (Ward & Nowell, 1998). This is critical because yield is a direct function of photosynthesis. The eight to nine leaves in the upper canopy supply 75-90% of the photosynthate that the ear requires during grain fill (Allison & Watson, 1966).

Strobilurins, including pyraclostrobin, are preventative fungicides that work by inhibiting spore germination (Butzen et al., 2005). The active compounds in strobilurins inhibit mitochondrial respiration by binding to the quinol or ubiquinone oxidation site in the cytochrome-bc_1 complex (Bartlett et al., 2002). Binding at this site halts electron transfer in the inner membrane of the mitochondria, which prevents adenosine triphosphate (ATP) from forming and so prevents further energy transfer in the fungal spore (Bartlett et al., 2002; Taiz & Zeiger, 2006).

Certain physiological alterations in plants may increase grain yield in the absence of disease (Bartlett et al., 2002). A decrease in the progression of senescence with increasing concentrations of pyraclostrobin in spinach leaves has been observed (Grossmann & Retzlaff, 1997). Maximum retardation of senescence showed 82% higher chlorophyll levels than the control. Although a delay in senescence could result in a longer duration of grain fill, it could also prolong dry-down and result in higher grain moisture. Glaab and Kaiser (1998) showed that exposure of spinach to kresoxim-methyl (methyl (E)-methoxyimino[2-(o-tolyloxymethyl)phenyl]acetate) increased nitrate reductase activity in the treated leaves even under lighted conditions. Köhle et al. (2002) showed an increase in nitrate assimilation and subsequent yield increase in wheat (*Triticum aestivum* L.) plants sprayed with pyraclostrobin when compared to a non-treated control. Further grain analysis showed similar C:N ratios, and relative protein contents of treated and untreated plants offered evidence that increased nitrate assimilation in plants contributed to increased growth (Köhle et al., 2002). If protein concentrations and C:N ratios are similar with increased nitrate assimilation, then the whole plant should have increased growth to compensate for the extra nitrate. Increased activity of certain nutrient-containing processes would logically result in greater demand for specific nutrients.

Several studies have established a link between plant nutrition and severity of disease. Plant nutrients such as Ca, K, Cl, Mn, B, and P are important for reducing disease severity in some instances (Brennan, 1992; Rupe et al., 2000; Sweeney et al., 2000; Sanogo & Yang, 2001; Thomason et al., 2001; Fixen et al., 2004). When combined with a soil fertility program that builds up and maintains optimal soil test nutrient levels, foliar-applied nutrients may directly and positively manage disease. For example, potassium chloride foliar sprays reduce symptoms of several fungal foliar pathogens on several cereal grain crops (Kettlewell et al., 2000). Foliar fertilizers may work in many circumstances leading to the availability of critical nutrients for optimum plant health and productivity.

Combining a foliar fertilizer with fungicide application may reduce application costs, improve disease suppression and nutrient response, and increase flexibility in managing crop response to environmental conditions during the growing season. Although producers cannot rely solely on foliar applications for fertilizer management (Curley, 1994; Johnson et al., 2001, Fernandez et al., 2006; Nelson et al., 2010), foliar fertilizers can complement soil amendments or provide an increase in nutrient supply during plants' peak demand times or when soil conditions restrict nutrient uptake (Fernandez & Eichert, 2009). Similarly, applying foliar fertilizers to match nutrient demands from physiological responses to pyraclostrobin may result in a synergistic yield increase. Pyraclostrobin, which has a range of fungicidal activity, is labeled for a variety of crops (Bartlett et al., 2002) and received a label for plant health benefits (BASF, 2010). Strobilurin fungicides continue to grow in popularity and usefulness. Combining pyraclostrobin and foliar fertilizers has great potential, not only for its possible synergistic benefit, but also the heightened flexibility it offers producers responding to conditions during the growing season. The objective of this research was to evaluate the effect of common foliar fertilizers in the presence and absence of pyraclostrobin on the severity of disease, ear leaf nutrient concentrations, grain quality, and yield.

2. Methods

2.1 Site Descriptions and Experimental Design

This study was conducted during 2008 and 2009 at three University of Missouri research centers near Novelty, Portageville, and Albany, Missouri. These locations offer differences in soil and climate, and they represent three of the state's major cropping areas. The diversity of temperature, rainfall, humidity, and soils offers different challenges and issues related to high-yield corn production.

The Greenley Memorial Research Center is located near Novelty in Northeastern Missouri (40°2′N, 92°14′W); Fisher Delta Research Center is near Portageville in Southeast Missouri (36°25′N, 89°42′W); and

Hundley-Whaley Center is in Northwest Missouri (40°15′N, 94°19′W) near Albany. All three locations were managed for high yielding corn, including two where irrigation was added. Irrigation not only provided adequate water to eliminate yield loss from drought stress, but also provided differing environmental conditions that could affect corn diseases. Novelty had supplemental sprinkler irrigation, while Portageville was flood-irrigated. Albany was not irrigated. Irrigation events were scheduled using the Woodruff chart (Henggeler, 2008). In addition to irrigation, fertility and pest management were targeted at producing high yields. Soil type, management practices, planting and harvest dates, and environmental conditions at the time of foliar fungicide and fertilizer application are reported in Table 1.

Table 1. Field information and management practices at each location in 2008 and 2009.

Field information and management practices	Novelty (Knox County)		Portageville (Pemiscot County)		Albany (Gentry County)	
	2008	2009	2008	2009	2008	2009
Soil series	Putnam silt loam†		Tiptonville sandy loam‡		Grundy silt loam§	
Previous crop	Corn	Corn	Soybean	Corn	Soybean	Soybean
Planting date	19 May	7 May	1 May	23 Apr.	21 May	22 May
Replications	5	4	4	4	3	5
Fertilizer rate (N-P_2O_5-K_2O kg ha^{-1})	260-80-110	260-90-130	180-0-0	200-0-0	180-70-90	250-70-90
Hybrid	DK63-42	DK63-42	P33N58	P33N58	DK62-43	DK62-43
Seeding rate (seeds ha^{-1})	86,500	86,500	86,500	86,500	69,200	72,900
Fungicide and foliar fertilizer application date	23 July	28 July	9-10 July	7 July	16 July	3 Aug.
Air temperature (°C)	26	21	24	26	32	31
Relative humidity (%)	50	90	80	72	70	65
Corn height (m)	2.45	1.83	3.05	2.13	3.05	2.64
Harvest date	10 Oct.	3 Nov.	22 Sep.	23 Oct.	21 Nov.	3 Nov.

Note. †fine, smectitic, mesic Vertic Albaqualfs; ‡fine-silty, mixed, thermic Typic Argiudolls; §fine, montmorillonitic, mesic Aquic Argiudolls.

Treatments consisted of a factorial arrangement of the 13 foliar fertilizers (Table 2) and fungicide (presence or absence of pyraclostrobin at 0.11 kg ai ha^{-1}) organized in a random complete block design with three to five replications depending on site and year (Table 1). Combinations of foliar fertilizer and fungicide also included a nonionic surfactant (Induce®, proprietary blend of alkyl aryl polyoxylkane ethers, free fatty acids, and dimothyl polysiloxane, Helena Chemical Company, Collierville, TN) at 0.25% vol./vol. All treatments simulated aerial application using a hand boom propelled by CO_2 (Bellspray, Inc. Opelousas, LA) at 28 L ha^{-1}. All applications were made when corn was at VT (tasseling) (Ritchie et al., 1993), which is congruent with typical preventative fungicide application (BASF, 2010). Foliar fertilizers and application rates were selected based on local availability and experience. Table 2 summarizes foliar fertilizer products and application rates. Plots were maintained weed-free as detailed in Table 3 by individual sites and years.

Table 2. Foliar fertilizer characteristics, manufacturers, and application rates

Fertilizer (%N-%P_2O_5-%K_2O-%S)	Trade name and manufacturer	Application amount
3-18-18-0	NA-CHURS/ALPINE Solutions, Marion, OH	18.7 L ha^{-1}
0-0-30-0	Double-OK, NA-CHURS/ALPINE Solutions, Marion, OH	18.7 L ha^{-1}
Potassium thiosulfate (0-0-25-17)	KTS, Tessenderlo Kerley Inc., Phoenix, AZ	9.4 L ha^{-1}
Potassium thiosulfate plus urea triazone (5-0-20-13)	Trisert K+, Tessenderlo Kerley Inc., Phoenix, AZ	14.0 L ha^{-1}
Potassium chloride (0-0-62-0)	PCS, Potash Corp. of Saskatchewan, Northbrook, IL	2.8 kg ha^{-1}
25-0-0-0 controlled release nitrogen as methylene urea and diurea	CoRoN, Helena Chemical Co., Collierville, TN	28.1 L ha^{-1}
24-0-1-0.6 slow release N with 0.25% B	Pacer N, Crop Production Services, Galesburg, IL	28.1 L ha^{-1}
22-0-2-1 with 0.25% B	Task Force Maize, Crop Production Services, Galesburg, IL	9.4 L ha^{-1}
30-0-0-0	Nitamin, Koch Agronomic Services, Wichita, KS	9.4 L ha^{-1}
Boron (10%)	NA-CHURS/ALPINE Solutions, Marion, OH	2.34 L ha^{-1}
Mn-chelate	NA-CHURS/ALPINE Solutions, Marion, OH	2.34 L ha^{-1}
Fe-Mo-Mn-B-Zn (0.3%-0.01%-3.2%-0.2%-2.1%) premix	MAX-IN, Winfield Solutions, LLC, St. Paul, MN	2.34 L ha^{-1}
6-0-0-0 with 10% Ca	Nutri-Cal, CSI Chemical Corp, Bondurant, IA	23.4 L ha^{-1}

Table 3. Crop protection chemical applications and rates for all six site-years

Site-Year	Herbicide	Rate (kg a.i. ha^{-1})	Timing	Date
Novelty 2008	Atrazine[†]	2.39	Preemergence	May 23
	S-metolachlor[‡]	1.89		
	Mesotrione[§]	0.210		
Novelty 2009	Atrazine	0.845	Preemergence	May 22
	Mesotrione	0.226		
	S-metolachlor	2.26		
	Glyphosate (N-(phosphonomethyl)glycine)	1.06	Postemergence	June 26
Portageville 2008	Glyphosate	1.49	Preemergence	May 1
	Atrazine	1.10		
	Glyphosate	1.49	Postemergence	June 6
	Atrazine	1.10		
Portageville 2009	Glyphosate	1.49	Preemergence	April 23
	Atrazine	1.10		
	Glyphosate	1.49	Postemergence	May 28
	Atrazine	1.10		
Albany 2008	Atrazine	3.40	Preemergence	May 21
	S-metolachlor	1.89		
	Glyphosate	0.409	Postemergence	June 16
Albany 2009	Atrazine	3.40	Preemergence	May 22
	S-metolachlor	1.89		
	Glyphosate	0.409	Postemergence	June 19

Note. [†](2-chloro-4-(ethylamino)-6-(isopropylamino)-s-triazine); [‡](2-Chloro-N-(2-ethyl-6-methylphenyl)-N-[(1S)-2-methoxy-1-methylethyl]acetamide); [§](2-[4-(methylsulfonyl)-2-nitrobenzoyl]-1,3-cyclohexanedione).

2.2 Measurements and Evaluations

Representative soil samples were taken from each experiment (Table 4) and evaluated to establish baseline levels for multiple essential plant macro- (2008 and 2009) and micro-nutrients (2009). Crop injury was rated on a scale of 0 (no visual injury) to 100% (complete crop death) 7 to 14 days after treatment (DAT). Severity of foliar disease was rated on a scale of 0 (no disease) to 100% (complete tissue colonization) 28 to 42 DAT.

Table 4. Soil characteristics for all six site-years

Year	Location	pH_s (0.01M $CaCl_2$)	Organic matter	CEC	Bray IP	Exchangeable (1M NH_4AO_c)									
						Ca	K	Mg	S	Zn	Fe	Mn	Cu	Cl	B
			g kg^{-1}	cmol$_c$ kg^{-1}	kg ha^{-1}	kg ha^{-1}									
2008	Novelty	6.0	20.0	14.9	39.2	4033	323	411	-†	-	-	-	-	-	-
	Portageville	6.2	13.0	9.7	38.1	3418	218	212	-	-	-	-	-	-	-
	Albany	5.8	26.0	18.8	69.4	5863	262	780	-	-	-	-	-	-	-
2009	Novelty	5.0	25.6	18.4	68	4239	531	517	6.1	0.6	127	23	1.2	4.1	0.56
	Portageville	5.6	12.8	11.0	101	3164	190	477	6.1	0.44	51	26	1.2	4.5	0.30
	Albany	5.7	29.5	19.1	66	5870	294	751	6.5	1.25	64	39	1.6	3.2	0.45

Note. †Data were not collected in 2008.

Corn ear leaf samples (10 plot^{-1}) were collected, dried at 60 to 70 °C in a forced air oven, ground in a Wiley Mill (Thomas Scientific, Swedesboro, NJ), and plant tissue nutrient concentrations determined for all treatments seven DAT. For elemental analysis, plant tissue was digested (Hach Digesdahl Digestion Apparatus, Hach Company, Loveland, CO) using H_2SO_4 and H_2O_2. Nitrogen concentrations were determined colorimetrically (Laverty, 1963; Keeney & Nelson, 1982) with a spectrophotometer (Genesys 10, Thermo Spectronic, Rochester, NY). Foliar P, K, Ca, Mg, Cu, Fe, Mn, Zn, and S concentrations were determined using inductively coupled plasma (ICP) spectroscopy (Mills & Jones, 1996). For Cl analysis, a 0.15 g sample was extracted with 30 mL distilled H_2O (Brown & Jackson, 1955). The suspension was agitated on an orbital shaker (Eberbach Corporation, Ann Arbor, MI), filtered (Ahlstrom 642 filter paper), and analyzed using a chloride ion-specific electrode compared to standard solutions. Boron was determined from dry ashed samples, which were dissolved in 1 *M* HCl, then diluted to 0.1 *M* HCl, and analyzed with ICP spectroscopy. Corn grain yields were determined from the two center rows with a small-plot combine (Kincaid Equipment, Haven, KS) and adjusted to 150 g kg^{-1} moisture prior to statistical analysis. Grain samples from all sites except Albany in 2008 were collected for protein, oil, and starch and analyzed using near infrared spectroscopy (Foss Infratec 1241, Eden Prairie, MN).

Unless otherwise specified, data were subjected to an analysis of variance and means separated using Fisher's Protected LSD at $P \leq 0.01$. Data were subjected to an *F Max* test for homogeneity (Kuehl, 1994) and combined over site-years when appropriate. Main effects were presented in the absence of significant interactions (SAS, 2010).

3. Results and Discussion

3.1 Precipitation and Irrigation

Precipitation amounts for the six site-year growing seasons, from 1 April to 31 November, varied widely. Most were over the ten-year average for the specific site, but one site was below average. Novelty experienced greater-than-average rainfall during both growing seasons. In 2008, rainfall totaled 1182 mm, 48% greater than the 10-year average of 796 mm, and in 2009 rainfall totaled 1087 mm, 36% greater than the average. Sprinklers irrigated the Novelty site both years. In 2008, rainfall and the 149 mm of irrigation water combined for 67% more water than average on the field (Figure 1). In 2009, rainfall plus 188 mm of irrigation water made the total 60% greater than average. In 2008, Portageville experienced 601 mm of rain, which was 20% below the 10-year average of 752 mm. In 2009, 971 mm of precipitation fell, resulting in a 29% increase over the average. Both years had flood irrigation amounts totaling approximately 610 mm (Figure 2). Total water was 61% and 110% greater than the 10-year average in 2008 and 2009, respectively. Albany had higher-than-average precipitation both years. In 2008, precipitation was 861 mm, 28% greater than the 10-year average of 670 mm. The following year, 863 mm of rain fell for a 28% increase over the average. No supplemental irrigation was given at Albany (Figure 3).

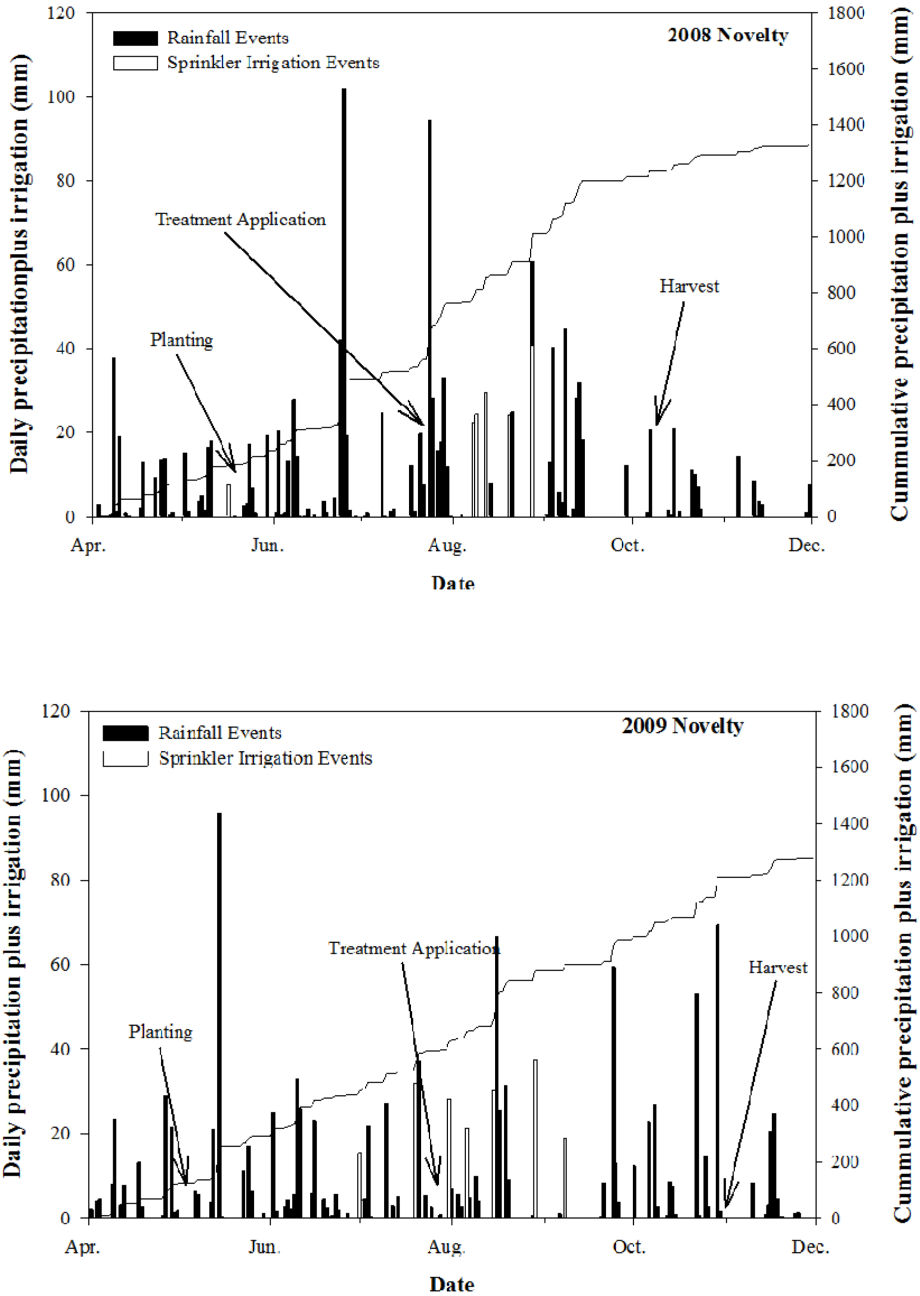


Figure 1. Precipitation and irrigation events, as well as planting, treatment application, and harvest dates at Novelty in 2008 and 2009

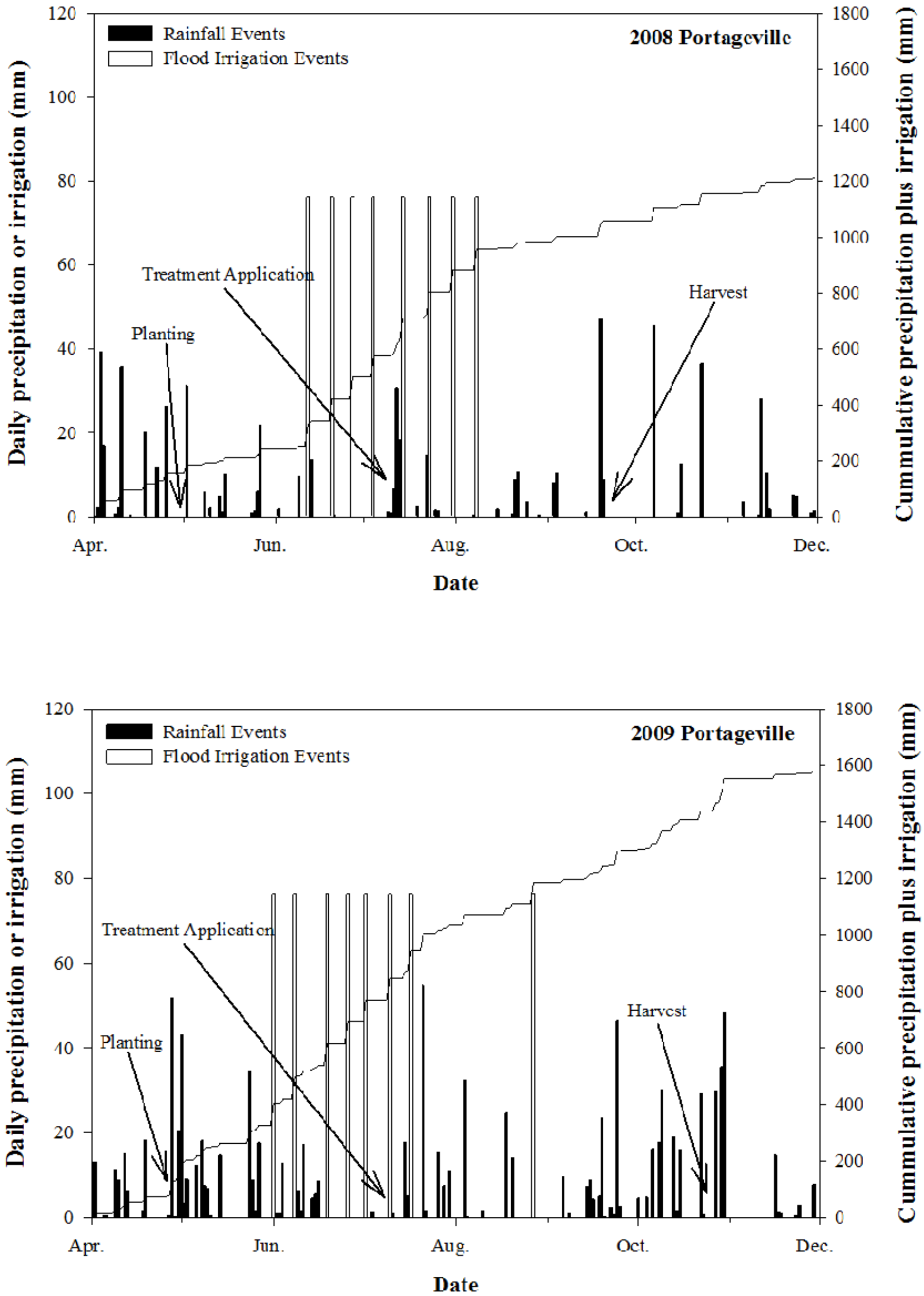
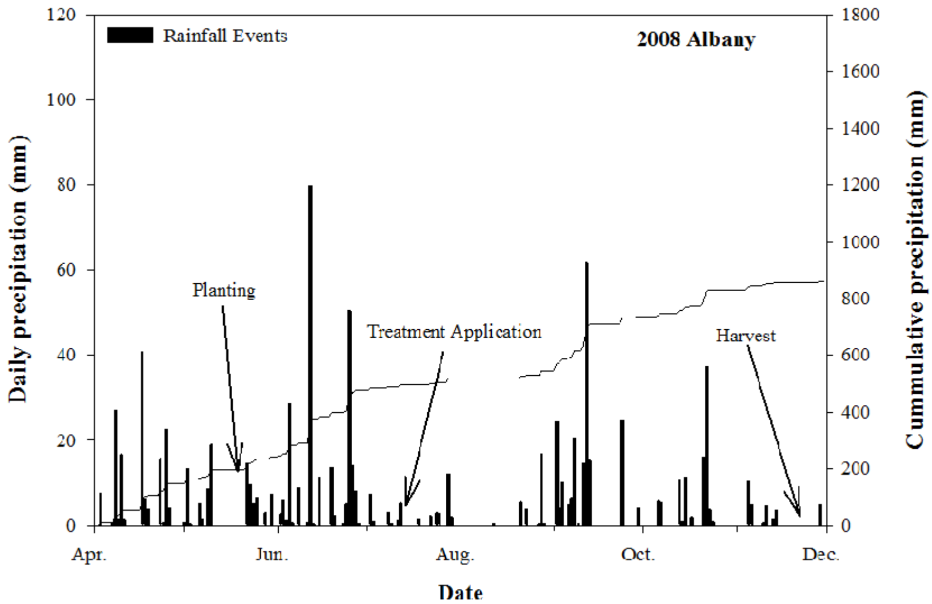


Figure 2. Precipitation and irrigation events, as well as planting, treatment application, and harvest dates at Portageville in 2008 and 2009



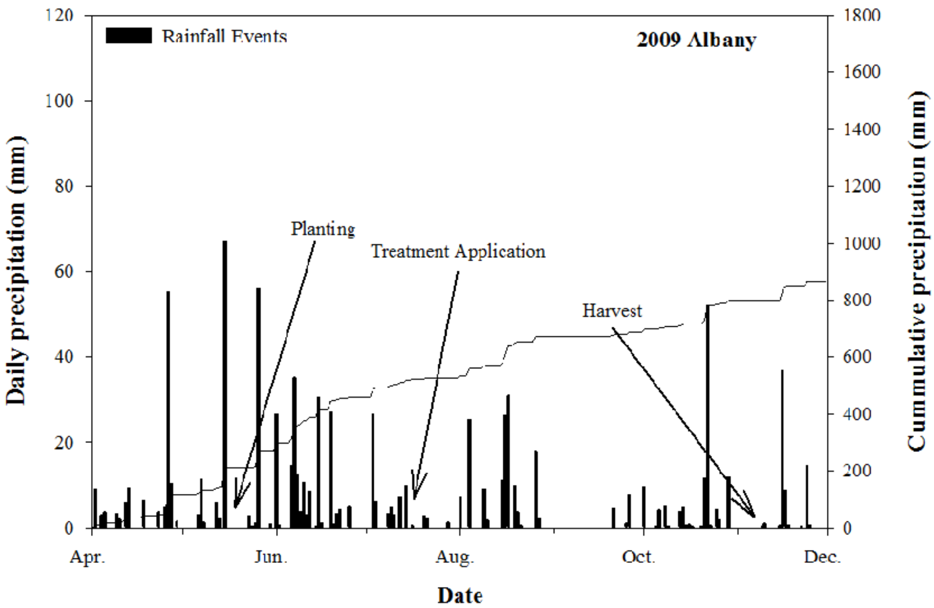


Figure 3. Precipitation, planting, treatment application, and harvest dates at Albany in 2008 and 2009. No supplemental irrigation was provided

3.2 Crop Injury

Crop injury, which occurred at all locations over both years, was generally persistent necrosis of leaf tissue depending on the treatment. Pyraclostrobin did not affect crop injury when compared to fertilizer-only treatments even though the treatments included surfactant (Table 5). Some fertilizer treatments caused injury. Overall and for all fertilizer treatments, crop injury was less than 10% at both Novelty and Albany for both years and less than 20% at Portageville (Table 6). The only treatment with consistently significant ($P \leq 0.05$) injury (4 to 20%) for all site-years was 0-0-30-0. The form of potassium in 0-0-30-0 is K_2CO_3 derived from potassium hydroxide (Nachurs Alpine Solutions, 2009). At high use rates, this fertilizer can cause necrosis to soybean (Nelson et al., 2012). When 0-0-30-0 was added to glyphosate, the pH of the solution was 12.0-13.0, which most likely elevated the injury levels (Nelson et al., 2012). Excluding 0-0-30-0, crop injury was inconsistent over the six site-years and was generally less than 10%. Inconsistencies most likely arose from varied environmental conditions during application, such as time of day, temperature, and relative humidity. Hotter and/or dryer conditions may have caused sprays to dry quicker on foliar surfaces, causing increased salt concentrations and leading to greater cellular damage (Neumann & Prinz, 1975; Gamble & Emino, 1987).

Table 5. Pyraclostrobin main effects for crop injury 7 to 14 days after treatment, grain yield, and quality characteristics averaged over fertilizer treatments and years (Albany) or site-years (Novelty and Portageville)

Fungicide treatment	Novelty and Portageville							Albany				
	Injury	Yield	Grain moisture		Protein	Oil		Injury	Yield	Moisture	Protein (2009)	Oil (2009)
			Novelty	Portageville		Novelty	Portageville					
	%	Mg ha^{-1}	g kg^{-1}	g kg^{-1}	g kg^{-1}	g kg^{-1}	g kg^{-1}	%	Mg ha^{-1}	g kg^{-1}	g kg^{-1}	g kg^{-1}
Non-treated	1	10.7	226	162	80.4	43.1	42.2	0.5	10.5	179	90.4	41.3
Pyraclostrobin	1	11.2	231	162	81.2	44.0	42.4	0.5	10.5	179	90.8	41.5
LSD ($P \leq 0.01$)	NS	0.4	3	NS	NS	0.9	NS	NS	NS	NS	NS	NS

Table 6. Foliar fertilizer main effects for crop injury, grain yield, and grain quality characteristics. Data were averaged over fungicide treatments and years unless designated otherwise

Fertilizer treatment	Injury				Novelty and Portageville				Albany				
	Novelty		Portageville										
	2008	2009	2008	2009	Protein	Oil	Moisture	Yield	Injury	Protein (2009)	Oil (2009)	Moisture	Yield
	------------------ % ----------------				--------------g kg^{-1}-----------			Mg ha^{-1}	%	--------------g kg^{-1}-------------			Mg ha^{-1}
Non-treated	0	0	0	0	80.7	43.1	197.0	10.8	0	91.0	41.5	179	10.6
3-18-18-0	0	0	0	10	79.3	42.4	196.3	11.0	0	90.8	42.0	179	10.3
0-0-30-0	7	9	20	10	80.8	43.0	198.3	10.1	4	91.2	41.1	183	10.5
22-0-2-1, 0.25% B	0	0	0	0	80.9	43.2	196.9	11.0	2	89.9	40.9	181	10.7
24-0-1-0.6, 0.25% B	0	0	0	0	80.4	43.1	200.6	11.0	0	90.4	40.9	180	10.7
25-0-0-0, 0.01% Cl	0	0	0	10	81.3	44.1	195.4	10.9	0	90.6	42.0	181	11.0
0-0-25-17	10	0	0	0	81.6	43.2	195.4	10.9	0	90.4	41.8	180	10.5
5-0-20-13	9	2	0	10	82.5	42.3	196.5	11.1	0	90.1	40.2	180	10.2
0-0-62-0	0	2	0	0	80.5	42.7	196.7	10.9	0	90.8	40.5	179	10.5
30-0-0-0	0	0	0	0	80.8	42.3	197.4	11.9	0	90.8	40.9	177	10.8
6-0-0-0, 10% Ca	1	0	10	10	80.5	42.3	197.8	11.0	0	89.2	42.0	176	10.4
B	0	0	0	10	80.4	43.4	198.7	11.4	0	90.6	41.1	179	10.4
Fe-Mo-Mn-B-Zn	0	0	0	0	80.5	43.0	198.4	10.7	0	91.8	42.7	179	9.9
Mn-chelate	0	1	0	20	81.4	43.0	196.4	10.7	0	91.2	41.9	177	10.2
LSD ($P \leq 0.01$)	1	1	10	NS	NS	NS	NS	0.9	1	NS	NS	NS	0.8

3.3 Disease Severity

Severity of foliar disease was less than 5% for all six site-years (Tables 7 and 8). Diseases varied by location and included: anthracnose leaf blight (*Colletotrichum graminicola*), bacterial stalk rot (*Erwinia dissolvens*), common rust (*Puccinia sorghi*), common smut (*Ustilago zeae*), gray leaf spot (*Cercospora zeae-maydis*), and northern corn leaf blight (*Exserohilum turcicum*). Neither pyraclostrobin, nor any of the foliar fertilizers had any significant effect on disease severity. For all treatments over all site-years, severity was quite low, despite elevated crop injury exhibited with some of the fertilizer treatments, optimal weather conditions for disease, and three of the six site-years having continuous corn (Table 1). The lack of disease may have resulted from using hybrids with good to excellent resistance to diseases present at all locations (Sweets & Wright, 2008; Poland et al., 2009). The disease-suppressing effects of pyraclostrobin and foliar fertilizers were not rigorously tested due to lack of disease, even in the non-treated controls. Therefore, yield response may stem from the effects of fungicide treatments or crop nutrition impacts of the fluid fertilizers.

Table 7. Disease severity ratings from each location combined over 2008 and 2009. Data were combined over fertilizer treatments

Fungicide treatment	Novelty			Portageville				Albany	
	GLS†	CR	NCLB	GLS	ANTH	BSR	CS	GLS	CR
	%								
Non-treated	1	0.2	0.3	1	2	1	1	1	2
Pyraclostrobin‡	1	0.1	0	1	2	1	1	1	2
LSD ($P \leq 0.05$)	NS	NS	NS	NS	NS	NS	NS	NS	NS

Note. †Abbreviations: ANTH, Anthracnose leaf blight (*Colletotrichum graminicola*); BSR, bacterial stalk rot (*Erwinia dissolvens*); CR, common rust (*Puccinia sorghi*); CS, common smut (*Ustilago zeae*); GLS, grey leaf spot (*Cercospora zeae-maydis*); LSD, least significant difference; NCLB, northern corn leaf blight (*Exserohilum turcicum*); and NS, non-significant; ‡Pyraclostrobin at 0.110 kg ha^{-1} plus non-ionic surfactant at 0.25% vol/vol.

Table 8. Disease severity ratings from each location were combined over years and fungicide treatments within a location

Fertilizer treatment†	Novelty			Portageville				Albany	
	GLS	CR	NCLB	GLS	ANTH	BSR	CS	GLS	CR
	%								
Non-treated	1	0.3	0	1	2	1	1	0	2
3-18-18-0	1	0.1	0	1	2	1	1	0	2
0-0-30-0	1	0.1	0	2	1	1	1	1	2
22-0-2-1, 0.25% B	1	0.1	0	1	2	1	1	1	2
24-0-1-0.6, 0.25% B	1	0	0.1	1	2	1	1	1	3
25-0-0-0, 0.01% Cl	1	0.3	0	1	2	1	1	1	2
0-0-25-17	1	0.2	0	1	2	1	1	1	2
5-0-20-13	1	0.1	0	1	2	1	1	1	2
0-0-62-0	1	0.2	0	1	1	2	1	1	2
30-0-0-0	1	0.1	0	1	2	1	1	1	2
6-0-0-0, 10% Ca	1	0.1	0.1	1	2	1	1	1	2
Boron	1	0.1	0.1	2	2	1	1	1	2
Fe-Mo-Mn-B-Zn	1	0.1	0.1	1	1	1	1	1	2
Mn-chelate	1	0.2	0	1	1	1	1	1	2
LSD ($P \leq 0.05$)	NS	NS	NS	NS	NS	NS	NS	NS	NS

Note. †Abbreviations: ANTH, Anthracnose leaf blight (*Colletotrichum graminicola*); BSR, bacterial stalk rot (*Erwinia dissolvens*); CR, common rust (*Puccinia sorghi*); CS, common smut (*Ustilago zeae*); GLS, grey leaf spot (*Cercospora zeae-maydis*); LSD, least significant difference; NCLB, northern corn leaf blight (*Exserohilum turcicum*); and NS, non-significant.

3.4 Leaf Nutrient Concentration

All ear leaf nutrient concentrations in the non-treated control were above the sufficiency levels at the time of application (Mills & Jones, 1996), with the exception of Ca concentrations (data not presented). Calcium levels were slightly below the sufficiency range. No interactions appeared between fungicide and foliar fertilizer treatment for any nutrient, so data main effects are presented. Interactions with year are presented separately for significant interactions. A significant interaction (P = 0.05) occurred between pyraclostrobin treatment and site-year for foliar nitrogen (N) concentrations; therefore, data were presented by individual site-years for ear leaf N concentrations (Table 9). All other foliar nutrient concentrations by pyraclostrobin treatment were averaged over all six site-years (Tables 9 and 10). Only one site-year (Novelty, 2008) showed differences (P= 0.1) between the pyraclostrobin-treated and non-treated corn for ear leaf N concentration. The pyraclostrobin treatment had 6.3 g kg^{-1} lower ear leaf N concentration. A decrease in tissue N levels may signify an increase in the activities of certain N-containing physiological processes that can increase with strobilurin fungicides (Glaab & Kaiser, 1998; Oerke et al., 2001; Köhle et al., 2002).

Table 9. Ear leaf B, Cu, and N concentration as affected by pyraclostrobin. Boron and Cu tissue concentrations were combined over all site-years. Nitrogen foliar concentrations are presented individually by site-year. Data were combined over fertilizer treatments

Fungicide treatment	Boron	Copper	Nitrogen					
			Novelty		Portageville		Albany	
			2008	2009	2008	2009	2008	2009
	--------mg kg^{-1}-------		----------g kg^{-1}----------					
Non-treated	65.1	30.7	39.4	42.4	47.5	47.4	40.3	35.3
Pyraclostrobin	76.3	32.7	33.1	44.5	46.1	49.9	40.5	33.0
LSD ($P \leq 0.1$)	8.3	1.9	5.6	NS	NS	NS	NS	NS

Table 10. The effect of pyraclostrobin on ear leaf P, K, Ca, Mg, S, Zn, Fe, Mn, and Cl concentration. Data were combined over site-years and foliar fertilizer treatments

Fungicide treatment	P	K	Ca	Mg	S	Zn	Fe	Mn	Cl
	-------------g kg^{-1}-------------				----------------mg kg^{-1}----------------				
Non-treated	2.08	22.6	1.69	2.33	290	48.8	332	124	5280
Pyraclostrobin	2.08	22.4	1.70	2.37	268	47.8	334	123	5270
LSD ($P \leq 0.1$)	NS	NS	NS	NS	NS	NS	NS	NS	NS

Boron concentrations in corn ear leaf samples increased with pyraclostrobin by 11.2 mg kg^{-1}. Strobilurin fungicides have altered various hormonal processes in plants, and B has been associated with various plant hormonal responses (Venancio et al., 2003; Marschner, 1995). In addition, B has been proposed to exhibit a role in respiration, which can be suppressed by strobilurin fungicides (Parr & Loughman, 1983; Nason et al., 2007). Strobilurin fungicides have also been shown to retard senescence in plants. Correlated with the impedance of senescence is an increase in indole-3-acetic acid (IAA) concentrations (Marschner, 1995; Grossmann & Retzlaff, 1997). Boron is closely associated with indole-3-acetic acid metabolism. Altering these biochemical processes with pyraclostrobin may explain differences in the observed B concentrations in the ear leaf.

Ear leaf Cu concentrations increased with pyraclostrobin 2.0 mg kg^{-1}. Köhle et al. (2003) showed that plants treated with pyraclostrobin had enhanced tolerance of oxidative stress. Copper is a component of superoxide dismutase, which along with other plant antioxidative enzymes detoxifies superoxide radicals (Taiz & Zeiger, 2006). Increased Cu in plant tissue may enhance tolerance of oxidative stress. All other nutrient concentrations showed no significant differences due to the main effect of pyraclostrobin treatment (Table 10).

Foliar fertilizer main effects are presented because limited 2- (year × fertilizer, fertilizer × fungicide) and no 3-way (year × fertilizer × fungicide) interactions were observed. A significant year × fertilizer interaction (P = 0.0009) occurred for ear leaf B concentrations; therefore, data are presented separately by site-year for B (Table 11). No consistent differences were found for ear leaf B concentrations between the non-treated control and the fertilizers that were applied. One foliar fertilizer (6-0-0-0) increased B concentrations over others in two site years. At Novelty in 2009, B concentration increased with 6-0-0-0 (P = 0.1) over 25-0-0-0 and Mn-chelate. At Albany that year, B concentration was greater than the non-treated control, 0-0-30-0, and 22-0-2-1. All other significant differences regarding foliar B concentrations were inconsistent and did not seem to show a pattern among site-years or with treatments that included B.

Table 11. Ear leaf B concentrations by site-year. Data were combined over pyraclostrobin treatments

Fertilizer treatment	Boron					
	Novelty		Portageville		Albany	
	2008	2009	2008	2009	2008	2009
	----------mg kg^{-1}----------					
Non-treated	53.5	26.8	45.8	49.3	72.8	54.2
3-18-18-0	42.8	33.0	75.8	37.0	91.2	205.8
0-0-30-0	35.3	31.2	80.3	63.5	85.8	63.8
22-0-2-1, 0.25% B	53.5	26.8	52.2	91.3	80.2	54.0
24-0-1-0.6, 0.25% B	34.8	23.5	61.5	58.3	106.0	100.8
25-0-0-0, 0.01% Cl	44.5	17.8	94.3	52.2	86.3	118.8
0-0-25-17	46.8	27.2	48.8	48.0	81.0	135.8
5-0-20-13	38.8	31.8	52.2	51.0	81.8	112.7
0-0-62-0	50.7	33.3	57.2	34.2	80.5	117.8
30-0-0-0	67.0	33.7	53.3	59.2	90.3	218.3
6-0-0-0, 10% Ca	57.0	38.7	54.3	59.0	79.8	276.5
Boron	45.3	27.3	56.5	33.7	86.2	256.0
Fe-Mo-Mn-B-Zn	41.2	29.2	75.0	33.0	85.7	136.0
Mn-chelate	87.7	21.2	72.5	51.3	98.0	116.8
LSD ($P \leq 0.1$)	39.6	17.4	37.7	30.5	30.5	119.0

Other ear leaf nutrient concentrations were averaged over all six site-years (Table 12) because 2- and 3-way interactions were insignificant. With the exception of Cu, all foliar nutrients exhibited significant differences (*P* = 0.1) among treatments. Ear leaf nutrient concentration with 24-0-1-0.6, 0.25% B increased over the control and/or other foliar fertilizers for several nutrient concentrations, but had lower ear leaf S concentrations compared to that of the 5-0-20-13 fertilizer treatment. The fertilizer treatment of 24-0-1-0.6, 0.25% B also had a higher (59 mg kg^{-1}) ear leaf Fe tissue concentration compared to the control. All other nutrient concentration increases were greater than the other foliar fertilizers for N, P, and Mg. Surprisingly, 25-0-0-0 increased ear leaf P concentration 0.19 g kg^{-1} compared to the control. Other differences for foliar nutrient levels were among other foliar fertilizers and were inconsistent. The fertilizer treatment of 5-0-20-13 resulted in the greatest concentrations and an increase over the control for sulfur levels with 369 mg kg^{-1}, a 114 mg kg^{-1} increase over the control. Conversely, 5-0-20-13 decreased foliar nutrient concentration of Ca and Mg compared to the control, and it decreased P and K compared to other foliar fertilizers. Ear leaf Mn concentration decreased 16 mg kg^{-1} with 0-0-62-0 compared to the control, while it decreased N, K, Mg, Fe, and Mn concentrations compared to several other foliar fertilizers. The micronutrient mix of Fe-Mo-Mn-B-Zn decreased Mg 0.20 g kg^{-1} and Zn 5.4 mg kg^{-1} compared to the non-treated control, while it decreased S and Fe concentrations compared to other foliar fertilizers. However, Fe-Mo-Mn-B-Zn increased Mn concentrations compared to other foliar fertilizers, which is expected with Mn in the formulation.

Table 12. Ear leaf nutrient concentrations seven days after treatment

Fertilizer treatment	Nitrogen	Phosphorus	Potassium	Calcium	Magnesium	Sulfur	Zinc	Iron	Manganese	Copper	Chloride
	$g\ kg^{-1}$					$mg\ kg^{-1}$					
Non-treated	41.8	2.05	23.0	1.72	2.41	255	51.1	319	132	30.3	5312
3-18-18-0	39.8	2.06	23.2	1.80	2.36	256	46.3	334	128	29.7	4966
0-0-30-0	42.4	2.12	23.7	1.75	2.31	258	46.6	341	128	32.0	5401
22-0-2-1, 0.25% B	43.1	2.03	22.6	1.68	2.37	293	50.4	339	125	30.2	5083
24-0-1-0.6, 0.25% B	44.5	2.19	22.7	1.74	2.47	268	50.3	378	131	31.1	5371
25-0-0-0, 0.01% Cl	41.5	2.24	22.1	1.71	2.44	265	46.6	327	121	31.6	5365
0-0-25-17	44.4	2.09	23.6	1.76	2.51	336	49.6	322	121	31.8	5307
5-0-20-13	43.0	1.94	21.8	1.50	2.28	369	48.3	342	116	32.3	5436
0-0-62-0	43.3	2.16	22.5	1.60	2.32	258	46.8	325	116	32.2	5195
30-0-0-0	42.7	2.03	22.2	1.72	2.36	263	46.9	325	120	31.4	5406
6-0-0-0, 10% Ca	40.8	2.01	22.1	1.71	2.24	249	46.7	321	119	32.3	5627
Boron	39.9	2.15	21.4	1.68	2.29	283	49.5	334	116	31.2	5216
Fe-Mo-Mn-B-Zn	41.2	2.07	22.2	1.66	2.21	243	45.7	333	135	34.4	5345
Mn-chelate	43.4	2.06	22.1	1.74	2.34	250	50.8	343	130	32.0	4891
LSD ($P \leq 0.1$)	4.17	0.17	1.65	0.18	0.18	91.9	5.20	43.7	14.5	NS	527

Note. Data were combined over site-year and pyraclostrobin treatment.

3.5 Grain Yield, Moisture, Oil, and Protein Concentration

In the absence of significant interactions, main effects were presented and data were combined over site-years. Overall, grain yield data for Portageville and Novelty could be combined over all four site-years, whereas Albany data were combined over years. This could be due to differences in seeding rates, presence of irrigation, and/or hybrids. Portageville and Novelty had slightly higher yields than Albany (non-irrigated) probably due to the lack of supplemental irrigation at the Albany site. Pyraclostrobin increased grain yield 0.5 Mg ha^{-1} at Portageville and Novelty, but showed no effect at Albany (Table 5). Pyraclostrobin did not affect grain moisture content at Albany and Portageville. However, grain moisture at Novelty increased 5 g kg^{-1} with pyraclostrobin compared to the non-treated control (Figure 4). This may be due to differences in environmental conditions at the time of harvest, timing of harvest, or the difference in corn hybrids. However, applying fungicide to corn can increase grain moisture (Bradley & Ames, 2010; Munkvold et al., 2001). Such increases may result from delayed senescence, similar to reports on other crops with pyraclostrobin and other strobilurin fungicides (Grossman et al., 1999; Ruske et al., 2003; Ypema & Gold, 1999). This experiment included no measures to support delayed senescence. Similar to wheat studies, pyraclostrobin did not increase grain protein concentration compared to the non-treated control (Köhle et al., 2002). Pyraclostrobin did not increase grain oil concentration at Portageville or Albany; however it increased grain oil concentration 0.9 g kg^{-1} at Novelty (Table 5). Soybean studies have shown similar effects of strobilurin fungicides on grain oil concentrations (Nelson et al., 2010).

Foliar fertilizers did not affect grain protein, oil or moisture concentrations (Table 6). One fertilizer, 30-0-0-0, increased yield 0.11 Mg ha^{-1} over the non-treated control. Sole liquid N sources (30-0-0-0 and 25-0-0-0, 0.01% Cl) had the highest overall yields at Albany. Although no fertilizer showed a significant yield reduction compared to the non-treated control, producers should avoid applying foliar fertilizers such as 0-0-30-0 at the rates this study evaluated, which caused injury and plant necrosis. With the timing of application at tasseling, plants may not be able to recuperate from injury as well as younger plants.

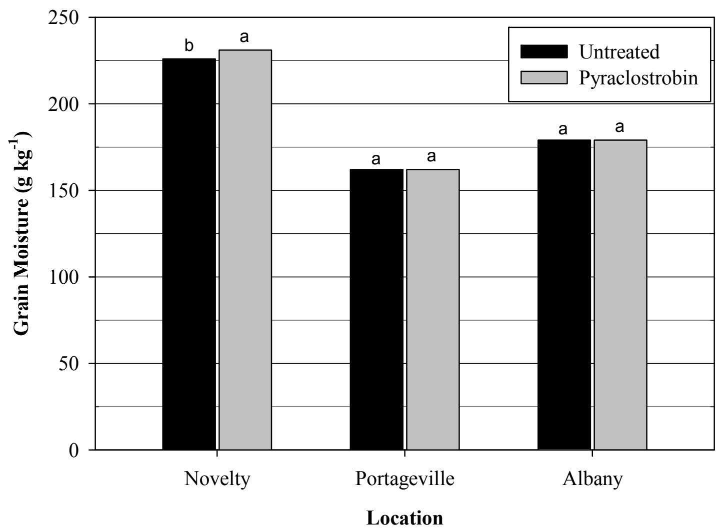


Figure 4. The effect of pyraclostrobin on harvested grain moisture at Novelty, Portageville, and Albany. Data were averaged over foliar fertilizer treatment and years

4. Conclusions

This study and previous research show that pyraclostrobin and other strobilurin fungicides interact with plants in multiple ways. Whether there is one interaction or many is unclear, but some factors such as the presence of pyraclostrobin lead to grain yield increases in corn crops over non-treated crops without visual differences in disease severity. During this study's six site-years, no differences appeared in disease severity among any treatments or controls. This may be a result of low levels of pathogenic inoculum, but is more likely a product of corn hybrid resistance to present pathogens. Of the 13 foliar fertilizers evaluated, 0-0-30-0 consistently caused injury over all six site-years. Ear leaf concentrations of nutrients varied widely over all macro- and micro-nutrients and site-years. Although many significant responses occurred from both main effects of pyraclostrobin and foliar fertilizers, few responses were consistent over all site-years, and no foliar fertilizer was responsible for more than one site-year's increase, except with regard to ear leaf S concentrations. Fertilizers, 5-0-20-13 and 0-0-25-17, both of which contain S, showed limited consistency in ear leaf nutrient concentrations seven DAT.

Neither pyraclostrobin nor any foliar fertilizer influenced grain protein concentrations at any of the site-years. Pyraclostrobin increased grain moisture and oil concentration at Novelty. Generally, pyraclostrobin had little effect on grain quality characteristics. Pyraclostrobin increased grain yields 0.5 Mg ha^{-1} at Novelty and Portageville, but showed no effect at Albany. An application of 30-0-0-0 was the only foliar fertilizer that significantly increased grain yields compared to the non-treated control. Sole N sources (30-0-0-0 and 25-0-0-0, 0.01% Cl) were the two highest yielding treatments at Albany. The combination of increases in yield from pyraclostrobin and 30-0-0-0 may provide tank mixture options for corn produced in high-yielding environments.

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