

# Integration of Row Spacing, Seeding Rates, and Fungicide Applications for Control of Sclerotinia Stem Rot in *Glycine max*

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## Abstract

Soybean (*Glycine max*) farmers in the Upper Midwest region of the United States often experience severe yield losses due to Sclerotinia stem rot (SSR). Previous studies have revealed benefits of individual management practices for SSR. This study examined the integration of multiple control practices on the development of SSR, yield, and the economic implications of these practices. Combinations of row spacings, seeding rates, and fungicide applications were examined in multisite field trials across the Upper Midwest from 2017 to 2019. These trials revealed that wide row spacing and low seeding rates individually reduced SSR levels but also reduced yields. Yields were similar across the three highest seeding rates examined. However, site-years where SSR developed showed the highest partial profits at the intermediate seeding rates. This finding indicates that partial profits in diseased fields were reduced by high seeding rates, but this

trend was not observed when SSR did not develop. Fungicides strongly reduced the development of SSR while also increasing yields. However, there was a reduction in partial profits due to their use at a low soybean sale price, but at higher sale prices fungicide use was similar to not treating. Additionally, the production of new inoculum was predicted from disease incidence, serving as an indicator of increased risk for SSR development in future years. Overall, this study suggests using wide rows and low seeding rates in fields with a history of SSR while reserving narrow rows and higher seeding rates for fields without a history of SSR.

**Keywords:** disease management, field crops, integrated management, oilseeds and legumes, *Sclerotinia sclerotiorum*, soybean, yield loss and economic impacts

Sclerotinia stem rot (SSR) is a devastating disease of soybean (*Glycine max*) caused by the fungal pathogen *Sclerotinia sclerotiorum* (Lib.) de Bary (Willbur et al. 2019a). Between 2015 and 2019, yield losses totaled >6.05 million metric tons in the United States and Ontario, Canada, with nearly 1.84 million metric tons of yield loss in 2017 alone (Bradley et al. 2021). Severe yield losses occur when SSR severity indices reach 68%, but yield impacts are observed with SSR indices as low as 20% (Fall et al. 2018a; Willbur et al. 2019b). *S. sclerotiorum* causes yield loss by decreasing seed number, seed weight, and seed quality of infected plants (Danielson et al. 2004; Hoffman et al. 1998).

This fungus is capable of surviving in the soil as overwintering structures called sclerotia for ≤5 years (Adams and Ayers 1979). Upon prolonged exposure to conducive environmental conditions such as cool temperatures between 8 and 21°C and adequate moisture, these sclerotia begin to germinate (Clarkson et al. 2004; Michael et al. 2020; Phillips 1986). Sclerotia germinate to form fruiting bodies called apothecia that forcibly eject ascospores into the soybean canopy when exposed to a slight decrease in moisture tension, such as drying dew in the morning (Abawi and Grogan 1979; Dillard et al. 1995; Phillips 1986). This apothecial germination typically occurs in sync with soybean flowering, when soybean is most susceptible to SSR infection (Abawi and Grogan 1979; Roth et al. 2020). Ascospores that land on

senescing flowers germinate and enter the plant, using a large repertoire of pectin-degrading enzymes and secreted virulence factors (Asoufi et al. 2007; Huzar-Novakowski and Dorrance 2018; McCaghey et al. 2019; Westrick et al. 2019). Infections by *S. sclerotiorum* are highly efficient if the ascospores are exposed to cool temperatures (15 to 25°C) and wet conditions for 2 to 4 h (Shahoveisi and del Río Mendoza 2020; Young et al. 2004). After successful infection, the fungus establishes itself throughout the vascular tissue, leading to water restriction, wilting, and premature death. At the end of the fungus' life cycle, the fungus will develop sclerotia to serve as overwintering structures and the source of inoculum in future years.

Severe SSR epidemics in soybean are inconsistent because *S. sclerotiorum* germination and development are highly dependent on weather conditions. Many methods for controlling SSR are focused on altering the microenvironments where apothecia develop, such as manipulating seeding rate and row spacing to reduce humidity in the soybean canopy (Carpenter et al. 2021; Fall et al. 2018b; Grau and Radke 1984; Lee et al. 2005). Fungicides are often used for SSR management (Willbur et al. 2019b), whereas tillage, cover crops, and genetic resistance are infrequently used because of their inconsistent management success (Roth et al. 2020).

Farmers have historically increased soybean seeding rates to maximize yields, but this practice can lead to conducive microenvironments for *S. sclerotiorum* and increase the risk of severe SSR epidemics. Seeding rates >432,250 plants/ha were shown to result in a greater disease severity index caused by SSR (Lee et al. 2005). As planting density increases, the airflow through the canopy is restricted, increasing moisture retention and humidity. Similarly, narrow row spacings (25 to 38 cm) accelerate canopy closure, which contributes to greater moisture, humidity, and SSR development compared with wider row spacings (76 cm) (Grau and Radke 1984). The number of developed apothecia peaks at 50% row closure, suggesting that canopy closure affects the production of apothecia, probably by affecting the quantity and quality of light reaching the soil (Fall et al. 2018b; Sun and Yang 2000; Thaning and Nilsson 2000). These conditions favor the germination of *S. sclerotiorum* sclerotia, apothecia development, ascospore release, and SSR development.

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Fungicides can be an effective method to manage SSR, but timing of application and selection of chemistry are important for successful control (Roth et al. 2020; Willbur et al. 2019b). Because soybeans are most susceptible during flowering periods, fungicide applications during these growth stages are most effective for controlling SSR (Mueller et al. 2002). In addition, boscalid and picoxystrobin fungicides were shown to provide the greatest levels of SSR suppression and the highest yields (Willbur et al. 2019b). Although the use of these fungicides can decrease the risk of SSR, fungicide applications may be unnecessary if weather conditions are not conducive to SSR development. Logistic regression models based on environmental factors such as 30-day moving averages of maximum air temperature, relative humidity, and maximum wind speed help provide information on the necessity of in-season fungicide applications for SSR management (Willbur et al. 2018a, b). When GPS coordinates, row width, presence or absence of soybean flowers, and irrigation information are provided, these models calculate local risk levels for the development of apothecia (Willbur et al. 2018a, b). Furthermore, these risk levels are used to recommend whether a fungicide should be applied on a specific date. These models are publicly available and currently trademarked as Sporecaster.

Although seeding rate, row spacing, and properly timed fungicide applications independently affect SSR incidence and severity, little is known about how effective these practices are when integrated. This study investigated the integration of these control practices and how they affect SSR development in soybean. Using a multistate, multi-year set of integrated management field trials, we attempted to determine which management practices or combination of practices led to the greatest reduction in SSR development, how these practices affect soybean yield, and which practices are the most economically profitable.

## Materials and Methods

**Locations and experimental design.** From 2017 to 2019, field trials at multiple locations across five states (Fig. 1 and Table 1) were used to examine management practices for controlling SSR. The three main factors investigated in this study were row spacing (38 and 76 cm), seeding rate (270,000, 345,000, 420,000, and 495,000 seeds/ha), and fungicide application programs, including a nontreated control. Fungicide applications were implemented under two programs, following “model applications” in Sporecaster based on SSR risk models (Willbur et al. 2018a, b) or “standard applications” based on soybean phenological growth stages, with applications at R1 (first flower present on the main stem) and R3 (pod development on one of the uppermost four nodes) (Fehr et al. 1971). Locations were examined for all three factors simultaneously ( $n = 10$  site-years; “fully integrated” locations) or for only two of the factors, seeding rate and fungicide application ( $n = 8$  site-years; “partially integrated” locations). The row width variable was excluded from the partially integrated trial locations because of equipment limitations. Trial details are presented in Table 1.

**Fully integrated trials.** Fully integrated locations were planted as split-split plots with a randomized complete block design, and treatments at all site-years were replicated four times. The whole plot factor was a planting width of either 38 or 76 cm, the subplot factor consisted of the four seeding rates, and the subsubplot factor consisted of the three fungicide application programs. Plots were planted with a custom plot planter, with 38-cm row width plots having six rows and 76-cm row width plots having four rows. All plots in the fully integrated trials were 6.4 m long. For the 38-cm row width plots, the inside four rows were assessed for SSR and yield, and for the 76-cm row width plots, the inside two rows were assessed to account for edge effects within these plots.

All fungicide applications were made with picoxystrobin (DuPont Approach, Corteva, DE). Applications were made with a backpack sprayer pressurized by CO<sub>2</sub>, equipped with series 8002 flat fan nozzles and calibrated to deliver 187 liters/ha, for a total application of 163 g of picoxystrobin per hectare. Standard applications were made based on soybean growth stage, with one application at R1 and a

second at R3. Model applications were made if the appropriate models indicated an apothecial risk level over a predetermined action threshold of 40% in nonirrigated conditions and 10% in irrigated conditions (Willbur et al. 2018a). If a fungicide application was recommended, an application was made with the same chemistry and rate as the standard applications, and the location was reassessed 14 days later to determine whether a second application was recommended by the models. For these models to appropriately recommend a spray, information about location, row spacing, flowering status, and irrigation conditions was entered into the Sporecaster application (beta to version 1.3) when soybeans reached R1 (Willbur et al. 2018a, b). Application dates for both standard and model programs for each site-year are provided in Table 2.

Beginning at the R6 growth stage (fully filled pod at one of four uppermost nodes on main stem), plots were scouted for SSR. In each plot, a total of 10 independent, 1-meter sections of inner rows were rated for disease incidence (DI). We determined DI by counting the total number of diseased plants in all 10 sections. We then calculated percentage DI by taking the number of plants with SSR and dividing by the total number of plants assessed. Disease severity (DS) was measured in 30 separate 0.3-m sections of inner rows, where the plant with the most severe SSR was identified and scored on a scale of 0 to 3 (0 = no disease present, 1 = mycelia present on lateral branch, 2 = mycelia present on main stem without girdling, 3 = mycelia present on main stem and full girdling has occurred), as described by Grau et al. (1982). We calculated average DS by taking the sum of all severity scores and dividing by the number of 0.3-m sections that had SSR present. At the Michigan trial location, we scored average DS by rating 30 consecutive plants within the inner rows on the 0 to 3 scale. We then calculated disease severity index (DIX) with the following equation, as described by Willbur et al. (2019b):

$$DIX = DI \times \left( \frac{DS}{3} \right).$$

All plots were harvested with small-plot harvesters. The four inner rows of the 38-cm row width plots were harvested, whereas only the two inner rows of the 76-cm row width plots were harvested to account for edge effect in the plots. Yields were then adjusted to 13% moisture.

**Partially integrated trials.** In partially integrated locations we examined only the four seeding rates and the three fungicide application programs described earlier. These trials were planted with a randomized complete block design, and fungicide applications and SSR ratings followed the same protocols as in the fully integrated trials. All plots in the partially integrated trials were 5.3 m long. All plots were harvested with small-plot harvesters, where only the two inner rows of the 76-cm row width plots were harvested to account for edge effect in the plots. Yields were adjusted to 13% moisture.

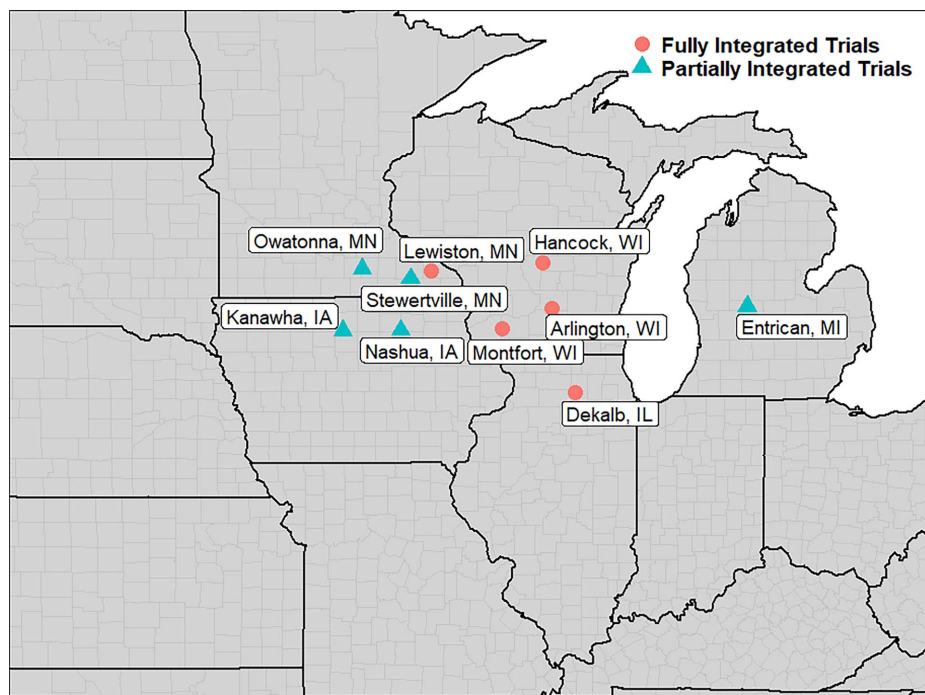
**Statistical analysis.** The fully integrated trials ( $n = 10$ ) and partially integrated trials ( $n = 8$ ) were analyzed separately. All data were analyzed by a generalized linear mixed model (GLMM) analysis of variance via the GLIMMIX procedure in SAS (version 9.4; SAS Institute, Cary, NC). For the fully integrated trials a single GLMM was created with row spacing, seeding rate, and fungicide program set as fixed effects. A heterogeneous variance structure was used in the analysis; thus, the random effects included the intercept, row spacing, and seeding rate nested within row spacing. Additionally, within the random effects statement, experimental repetition was defined as the subject effect, and site-year was defined as the grouping effect.

For the partially integrated trials a single GLMM was created, with seeding rate and fungicide program set as fixed effects. Again, a heterogeneous variance structure was used where the intercept was the only defined random effect, with experimental repetition defined as the subject effect and site-year defined as the grouping effect. A normal distribution was confirmed for all response variables except for DIX, which required logarithmic transformations to obtain a normal distribution. For significant main effects and interactions, differences between treatment means were determined via Fisher's least significant difference at  $\alpha = 0.05$  significance level with the “mult” macro in SAS (Piepho 2004).

**Partial profit economic analysis.** A partial economic analysis was performed to determine the degree of profitability for the various treatments. The cost of acquiring and maintaining planting equipment is variable and difficult to approximate; therefore, row spacing was not analyzed in this section. Seed cost was approximated at US\$62 for 140,000 seeds, based on regional seed costs, and this value was used to calculate a final seed cost for each of the four seeding rates. Cost of fungicide was approximated at \$60.77/liter (\$230/gallon) of commercially formulated product, and a \$17.30/ha (\$7/acre) applicator cost was added to each fungicide application. The nontreated plots had no associated fungicide costs, the cost for model applications was estimated with a single application, and the

cost for standard applications was estimated with two applications. Costs associated with fuel and general maintenance of other equipment such as applicators for fertilizer or harvesters are assumed to be equal across all treatments and were excluded from this analysis. Three grain sale prices (\$0.33, \$0.44, and \$0.55/kg) were examined to represent low, medium, and high grain sale prices.

All site-years from both the fully integrated and partially integrated trials ( $n = 18$ ) were combined for this analysis. Site-years were separated based on the presence ( $n = 10$ ) or absence of SSR ( $n = 8$ ). A mean DIX value of 0.5 was used for the nontreated plots as a threshold such that site-years in which the mean nontreated DIX was  $>0.5$  were designated as SSR present, and if the mean



**Fig. 1.** Location of experimental trials performed in this study from 2017 to 2019 throughout the Upper Midwest region of the United States. Locations indicated by a red circle are called fully integrated trials, where three management practices for controlling *Sclerotinia* stem rot in soybean were examined. Locations indicated by a blue triangle are called partially integrated trials, where two management practices were examined.

**Table 1.** Description of trials assessed in this study

	Site	GPS coordinates	Year	SSR presence <sup>w</sup>	Cultivar <sup>x</sup>	Planting date	Harvest date
Fully integrated <sup>y</sup>	Dekalb, IL	41.8442, -88.8491	2018	No	DSR-2330	5/8/2018	10/18/2018
	Lewiston, MN	43.9807, -91.9180	2018	No	DSR-2330	5/17/2018	10/22/2018
			2019	Yes	DSR-2330	5/14/2019	10/18/2019
			2019	Yes	DSR-2330	5/23/2019	10/19/2019
	Arlington, WI	43.3182, -89.3319	2017	Yes	AG2031	5/30/2017	10/18/2017
			2018	No	DSR-2330	5/7/2018	10/18/2018
			2019	No	DSR-2330	5/23/2019	10/19/2019
	Hancock, WI	44.1211, -89.5386	2017	Yes	AG2031	5/25/2017	10/9/2017
			2018	Yes	DSR-2330	5/3/2018	10/25/2018
			2019	Yes	DSR-2330	5/10/2019	10/18/2019
Partially integrated <sup>z</sup>	Montfort, WI	42.9589, -90.3990	2019	Yes	DSR-2330	6/3/2019	10/28/2019
			2019	Yes	DSR-2330	6/3/2019	10/28/2019
	Kanawha, IA	42.9327, -93.7976	2018	Yes	AG28X7	5/18/2018	10/23/2018
			2019	Yes	AG2733	5/16/2019	10/26/2019
	Nashua, IA	42.9379, -92.5698	2018	No	AG28X7	5/17/2018	10/23/2018
			2019	No	AG2733	5/13/2019	10/9/2019
	Entrican, MI	43.3516, -85.1755	2018	Yes	AG2535	5/17/2018	10/22/2018
	Owatonna, MN	44.0198, -93.3820	2018	Yes	AG28X7	5/22/2018	11/1/2018
	Stewartville, MN	43.8443, -92.3543	2018	No	AG28X7	6/1/2018	11/15/2018
			2019	No	AG2733	5/15/2019	10/29/2019

<sup>w</sup> *Sclerotinia* stem rot (SSR) presence was defined as an average disease severity index (DIX) value of  $>0.5$  for the nontreated plots within each site-year, such that if the average DIX value for the nontreated plots in any given site-year was  $>0.5$ , that site-year was designated as having SSR present. Conversely, if the average DIX value was  $<0.5$ , that site-year was designated as having SSR absent.

<sup>x</sup> Cultivars are denoted by abbreviations of their respective companies. DSR = Dairyland Seed, AG = Asgrow.

<sup>y</sup> Fully integrated site-years examined three *sclerotinia* stem rot management practices: row spacing, seeding rate, and fungicide application.

<sup>z</sup> Partially integrated site-years examined two *sclerotinia* stem rot management practices: seeding rate and fungicide application.

nontreated DIX was <0.5, that site-year was designated SSR absent (Table 1). We calculated income for each experimental unit by multiplying the yield (kg/ha) by each of the three soybean prices (\$0.33, \$0.44, and \$0.55/kg) to find an estimated income value (US\$/ha). Then, the cost of treatments was subtracted from income to determine the partial profit. Predicted production of *S. sclerotiorum* inoculum (e.g., sclerotia returned to the soil after harvest) was calculated from the DI for each experimental unit according to Lehner et al. (2017), where every 10% increase in DI produced 1 kg/ha of sclerotia. This calculation was used to determine additional benefits of controlling SSR, even if yield preservation was not significant between treatments.

Both partial profit and predicted return of *S. sclerotiorum* inoculum were analyzed similarly to the previous analysis using a GLMM analysis via the GLIMMIX procedure in SAS. Partial profits from the three grain sale prices (\$0.33, \$0.44, and \$0.55/kg) were examined separately. Two subgroups of analyses were performed for partial profit consisting of either SSR present ( $n = 10$ ) or absent ( $n = 8$ ) site-years. For each subgroup, one GLMM analysis was performed for each grain sale price, with seeding rate and fungicide application program set as the main effects. When we examined the predicted return of *S. sclerotiorum* inoculum, only one analysis was performed for the SSR present group and not the SSR absent group because of a lack of disease. Similar to the partial profit analysis, one GLMM analysis was conducted for the predicted return of inoculum with seeding rate and fungicide application program as the main effect. In all analyses a heterogeneous variance structure was again adopted, with the intercept defined as the sole random effect, experimental repetition defined as the subject effect, and site-year defined as the grouping effect. Significant main effects between treatment means were determined via Fisher's least significant difference at  $\alpha = 0.05$  significance level with the "mult" macro in SAS (Piepho 2004).

## Results

**Fully integrated trials.** In the fully integrated trials ( $n = 10$ ), the integration of all three main effects resulted in no significant

interaction for DIX ( $P = 0.31$ ) or yield ( $P = 0.63$ ). However, the main effects of row spacing ( $P = 0.03$ ) and fungicide program ( $P < 0.001$ ) and their interaction ( $P < 0.001$ ) were significant for explaining DIX (Fig. 2). Wide row spacings had significantly less SSR than the narrow row spacings, and standard applications of picoxystrobin at R1 and R3 significantly reduced SSR compared with the nontreated controls (Fig. 2A). Furthermore, the model applications reduced SSR compared with the nontreated in the narrow row spacings but not in wide row spacings (Fig. 2A).

Row spacing, seeding rate, and their interaction also had significant effects on yield responses ( $P < 0.001$ ). Yields were greatest in narrow row spacing at  $\geq 420,000$  seeds/ha. Yields were lower in the wide row spacing, with all seeding rates being statistically similar with the exception of the seeding rate of 270,000 seeds/ha, which had the lowest yield (Fig. 2B). Fungicide application also had a significant effect on yield ( $P < 0.001$ ), in which the standard fungicide program led to the highest yield, whereas the model program and nontreated control were lower and not significantly different from one another (Fig. 3).

**Partially integrated trials.** In the partially integrated trials ( $n = 8$ ), the interaction between seeding rate and fungicide programs was found not to be significant for DIX ( $P = 0.69$ ) or yield ( $P = 0.84$ ). Yet seeding rate had significant effects on both DIX ( $P = 0.002$ ) and yield ( $P < 0.001$ ). DIX was lowest at the seeding rate of 270,000 seeds/ha, and all other seeding rates had similar SSR levels (Fig. 4A). Yield followed a similar trend in which 270,000 seeds/ha seeding rate resulted in the lowest yields, and the three additional seeding rates yielded similarly (Fig. 4B). Fungicide programs affected both DIX ( $P = 0.06$ ) and yield ( $P = 0.01$ ). The standard fungicide program, but not the model program, reduced SSR compared with not treating with fungicide (Fig. 5A). Furthermore, the standard program also had higher yields than the nontreated plots (Fig. 5B).

**Economics of integrated management practices.** When we examined the partial profits, there was no significant interaction between seeding rate and fungicide application program for all three grain sale prices. Similarly, no significant interaction was found when we examined the predicted return of sclerotia. In site-years when SSR did not develop, the highest seeding rate (495,000 seeds/

**Table 2.** Irrigation status and fungicide application dates for the trials in this study

	Site	Year	Irrigated condition <sup>w</sup>	Application date <sup>x</sup>				
				Standard applications <sup>y</sup>		Model applications <sup>z</sup>		
				First application	Second application	38 cm, first application	38 cm, second application	76 cm, first application
Fully integrated	Dekalb, IL	2018	–	6/25/2018	7/17/2018	–	–	–
	Lewiston, MN	2018	–	7/2/2018	7/18/2018	–	–	–
		2019	–	7/10/2019	7/30/2019	7/10/2019	–	–
	Arlington, WI	2017	–	7/14/2017	7/28/2017	–	–	–
		2018	–	6/27/2018	7/13/2018	–	–	–
		2019	–	7/12/2019	7/31/2019	–	–	–
	Hancock, WI	2017	+	7/14/2017	7/28/2017	7/28/2017	–	7/28/2017
		2018	+	6/29/2018	7/13/2018	6/29/2018	7/13/2018	7/13/2018
		2019	+	7/8/2019	7/29/2019	7/8/2019	7/29/2019	7/29/2019
	Montfort, WI	2019	–	7/15/2019	8/5/2019	–	–	–
Partially integrated	Kanawha, IA	2018	–	7/9/2018	7/30/2018	–	–	–
		2019	–	7/22/2019	7/31/2019	–	–	–
	Nashua, IA	2018	–	7/10/2018	7/31/2018	–	–	–
		2019	–	7/23/2019	8/5/2019	–	–	–
	Entrican, MI	2018	+	7/6/2018	7/25/2018	–	–	7/13/2018
	Owatonna, MN	2018	–	7/9/2018	8/1/2018	–	–	–
	Stewartville, MN	2018	–	7/17/2018	8/9/2018	–	–	–
		2019	–	7/23/2019	8/15/2019	–	–	–

<sup>w</sup> +, irrigated. –, not irrigated.

<sup>x</sup> The date on which a fungicide treatment was applied at each site-year.

<sup>y</sup> Standard applications followed a growth stage-dependent program with applications at both R1 and R3 growth stages.

<sup>z</sup> Model applications followed an apothecial risk model-dependent program such that applications were made if the appropriate risk models reached 40% in nonirrigated conditions or 10% in irrigated conditions. If a fungicide application was made, the models were reassessed 14 days later to determine whether a second application was recommended. If no application date is listed, the risk models did not recommend an application at that site-year. The apothecial risk models for irrigated conditions depend on soybean row spacing; therefore, some differences of application dates are seen between 38- and 76-cm row widths.

ha) resulted in the highest profit, and the lowest seeding rate (270,000 seeds/ha) resulted in the lowest profit across all grain sale prices (Table 3). However, in site-years where SSR developed, the 495,000 seeds/ha seeding rate consistently resulted in the lowest partial profits (Table 3). The two intermediate seeding rates (345,000 and 420,000 seeds/ha) were the highest profiting across all three grain sale prices (Table 3). The highest seeding rate also had the highest levels of predicted return of *S. sclerotiorum* inoculum, and the lowest seeding rate of 270,000 was the only rate with significantly fewer returned sclerotia (Table 3).

Fungicide use resulted in similar partial profits across all treatments when used under high soybean market prices regardless of whether SSR developed (Table 4). However, when SSR did not develop, the use of fungicides reduced partial profits for both the \$0.33 and \$0.44/kg sale prices (Table 4). When SSR did develop, fungicide use reduced partial profits at the \$0.33/kg sale price, but at the \$0.44/kg sale price, the use of fungicides led to similar partial profits as the nontreated controls (Table 4). Additionally, the use of the standard fungicide program resulted in the lowest levels of predicted *S. sclerotiorum* inoculum produced, and the nontreated controls had the highest levels (Table 4).

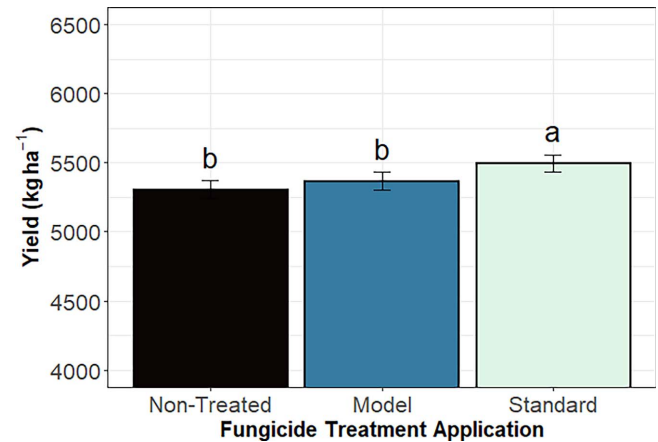
## Discussion

As shown by previous reports, SSR is a highly yield-limiting disease. However, a multitude of control methods are effective for reducing SSR (Roth et al. 2020). With options such as manipulating row spacing, seeding rates, or the use of fungicides, farmers have multiple tools for limiting the development of SSR and subsequent yield losses.

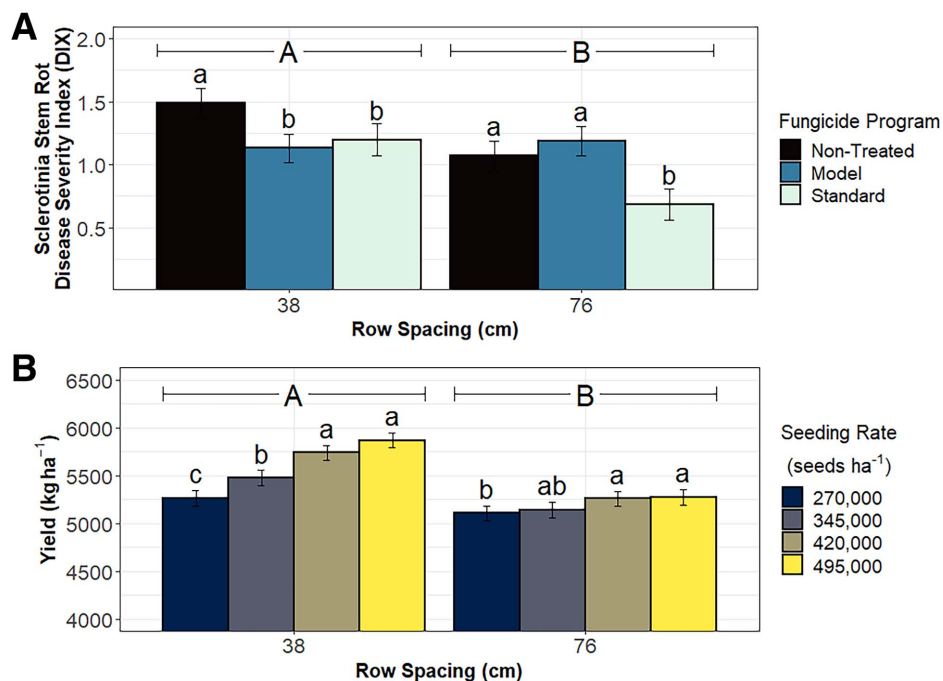
Our findings suggest that the use of wide row spacing without fungicide can reduce SSR to levels comparable to those of narrow row spacings coupled with two fungicide applications (Fig. 2A). This method would allow farmers to reduce their reliance on fungicides, leading to decreased costs and increased longevity of product efficacy. Our findings also suggest that lowering seeding rates could result in decreased SSR development (Fig. 4A), supporting previous work

(Carpenter et al. 2021; Lee et al. 2005). If a farmer were unable to transition toward wide row spacings, the use of lower seeding rates alone could be a suitable method for reducing SSR. Additionally, the use of these cultural management practices could be beneficial for organic production systems where pesticide use is limited.

Unfortunately, the use of either wide row spacing or low seeding rates is also associated with lower yields (Figs. 2B and 4B). When examining row spacing, we found a 391.2 kg/ha benefit of the narrow

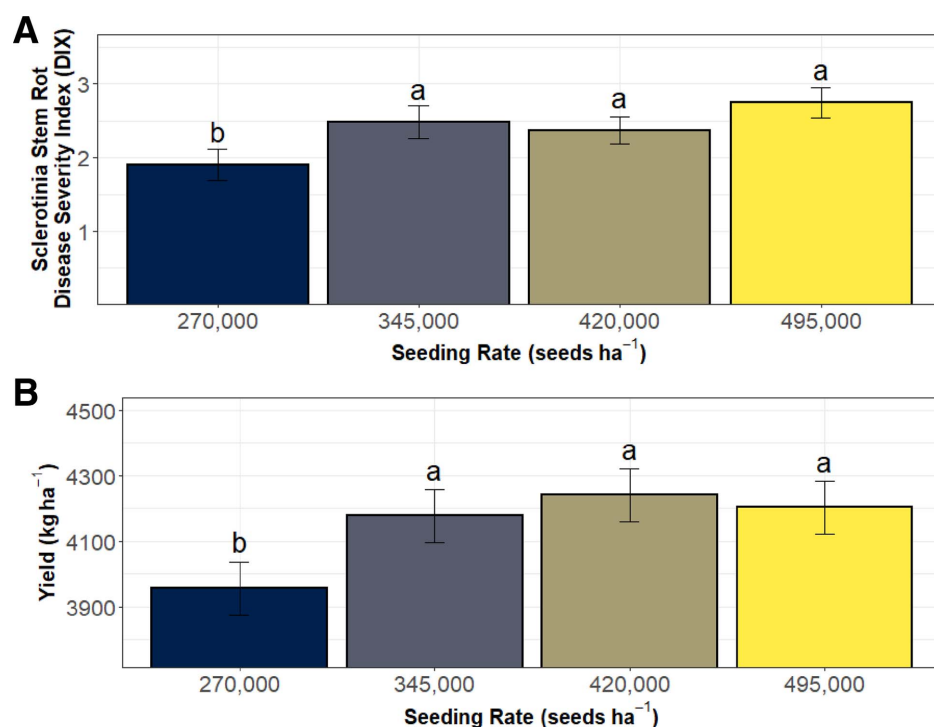


**Fig. 3.** Effect of fungicide treatments on soybean yield in fully integrated site-years ( $n = 10$ ). Trials examined the combined effects of two row spacings (38 and 76 cm), four seeding rates (270,000, 345,000, 420,000, and 495,000 seeds/ha), and two fungicide treatments (standard program of applications at R1 and R3 soybean growth stages and a model program of applications made when a risk prediction tool recommended a spray). The model program consisted of applications following *Sclerotinia* stem rot risk models called Sporecaster. Trials were performed across the Upper Midwest region of the United States between 2017 and 2019, and error bars represent the standard error of the mean. Factors sharing similar letters are not statistically different as determined by Fisher's least significant difference ( $\alpha = 0.05$ ).

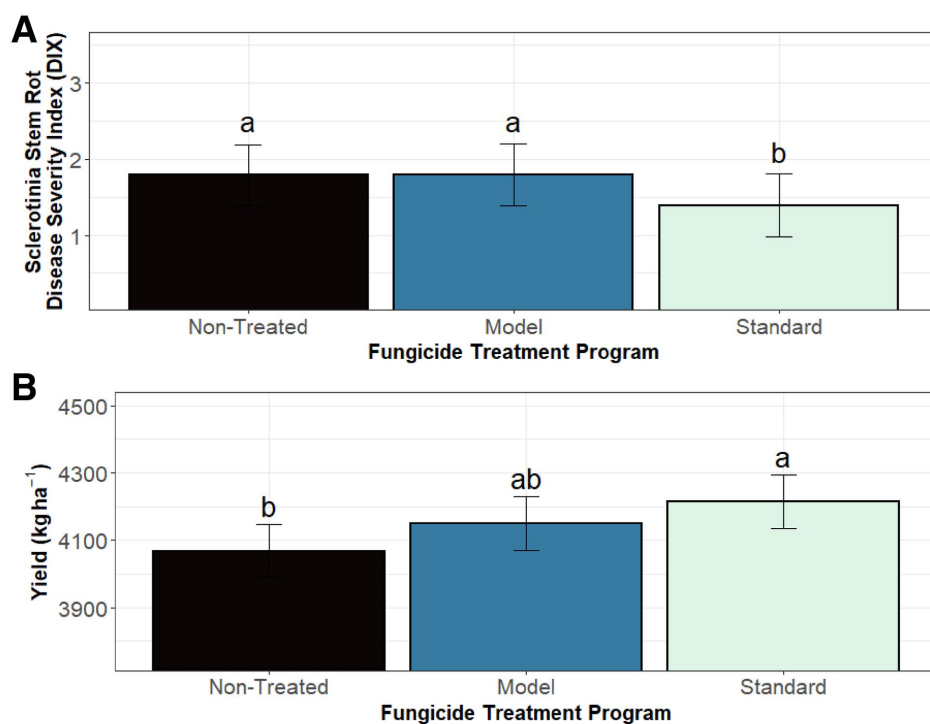


**Fig. 2. A,** Interaction of row spacing and fungicide applications on *Sclerotinia* stem rot (SSR) disease severity index (DIX) and **B,** the interaction of row spacing and seeding rates on soybean yield in fully integrated site-years ( $n = 10$ ). Trials examined the combined effects of two row spacings (38 and 76 cm), four seeding rates (270,000, 345,000, 420,000, and 495,000 seeds/ha), and two fungicide programs (standard program of applications at R1 and R3 soybean growth stages and a model program of applications made when a risk prediction tool recommended a spray). The model program consisted of applications following SSR risk models called Sporecaster. Trials were performed across the Upper Midwest region of the United States between 2017 and 2019. DIX values were subjected to a logarithmic transformation, and error bars represent the standard error of the mean. Factors within row spacings that share lowercase letters are not statistically different, and row spacings that share uppercase letters are not statistically different. These similarities were determined by Fisher's least significant difference ( $\alpha = 0.05$ ).





**Fig. 4.** Effect of seeding rate on **A**, Sclerotinia stem rot (SSR) disease severity index (DIX) and on **B**, soybean yield in partially integrated site-years ( $n = 8$ ). Trials examined the combined effects of four seeding rates (270,000, 345,000, 420,000, and 495,000 seeds/ha) and two fungicide treatments (standard program of applications at R1 and R3 soybean growth stages and a model program of applications made when a risk prediction tool recommended a spray). The model program consisted of applications following SSR risk models called Sporecaster. Trials were performed across the Upper Midwest region of the United States between 2018 and 2019. DIX values were subjected to a logarithmic transformation, and error bars represent the standard error of the mean. Factors sharing similar letters are not statistically different as determined by Fisher's least significant difference ( $\alpha = 0.05$ ).



**Fig. 5.** Effect of fungicide treatments on **A**, Sclerotinia stem rot (SSR) disease severity index (DIX) and on **B**, soybean yield in partially integrated site-years ( $n = 8$ ). Trials examined the combined effects of four seeding rates (270,000, 345,000, 420,000, and 495,000 seeds/ha) and two fungicide treatments (standard program of applications at R1 and R3 soybean growth stages and a model program of applications made when a risk prediction tool recommended a spray). The model program consisted of applications following SSR risk models called Sporecaster. Trials were performed across the Upper Midwest region of the United States between 2018 and 2019. DIX values were subjected to a logarithmic transformation, and error bars represent the standard error of the mean. Factors sharing similar letters are not statistically different as determined by Fisher's least significant difference ( $\alpha = 0.1$  when examining DIX and  $\alpha = 0.05$  when examining yield).

row spacing compared with the wide row spacing. This finding is consistent with previous reports showing a similar trend (Ablett et al. 1991; Andrade et al. 2019; Bullock et al. 1998; De Bruin and Pedersen 2008; Devlin et al. 1995; Grau et al. 1994; Lueschen et al. 1992; Oplinger and Philbrook 1992). Although the use of wide row spacing is shown here to reduce SSR levels, it has previously been shown that wide rows are also often associated with poorer weed control compared with narrow row spacings (Bradley 2006; Nelson and Renner 1999; Yelverton and Coble 1991).

When we examined seeding rates, yields were higher at higher seeding rates, and the 270,000 seeds/ha rate consistently yielded the lowest (Figs. 2B and 4B). Additionally, when we looked at the combined effect of row spacing and seeding rate, yield was more responsive to seeding rates in the narrow row spacing and more stable in the wide row spacing (Fig. 2B), with similar trends observed by Pedersen (2007). Our results suggest that the optimum seeding rate for maximizing yield in 38-cm rows is higher than in the 76-cm rows, which is consistent with previous reports (Devlin et al. 1995). This finding may reflect competition between seedlings, because the number of seeds planted within a row-foot under a constant seeding rate per area is higher in wide row spacings than in narrow row spacings. Therefore, the optimal seeding rate in wide row spacings may be lower than in narrow row spacings.

The data from our partial economic analysis indicate that the most profitable seeding rate depended on the presence or absence of SSR. In environments that did not favor SSR development, partial profits

increased with seeding rate, and at 495,000 seeds/ha the partial profits were greatest at all grain sale prices (Table 3). In environments where SSR did develop, a similar trend can be observed with increased profitability as seeding rate increased. Interestingly, partial profits of the highest seeding rate dropped sharply to levels similar to that of the lowest seeding rate across all grain sale prices (Table 3). This stark contrast between environments where SSR was present or absent suggests that the highest seeding rates have higher profitability, but only when there is no history of SSR. Additionally, predictions were made about the production of new *S. sclerotiorum* inoculum, serving as the inoculum source of future epidemics. Seeding rates of 270,000 seeds/ha were predicted to produce the lowest quantity of new sclerotia compared with seeding rates of 420,000 and 495,000 seeds/ha, which produced the highest quantities (Table 3). Because of the similar partial profits across the intermediate seeding rates and the increased risk of SSR development at higher seeding rates, a seeding rate of 345,000 seeds/ha may be the best option for balancing the goals of reducing disease levels, minimizing the production of new inoculum, and maximizing partial profits in environments where SSR development is favored.

Fungicides have been a valuable tool for controlling fungal diseases in crops and protecting yields. The use of fungicides, specifically for control of SSR, has been examined for their chemical efficacy and the optimal application timing (Mueller et al. 2002, 2004). Our data confirmed that chemical control is still a viable option for controlling SSR

**Table 3.** Effect of soybean seeding rates on economic partial profits and estimated *Sclerotinia sclerotiorum* inoculum returned to soil when Sclerotinia stem rot (SSR) was either absent or present

Disease presence <sup>x</sup>	Seeding rate (seeds/ha)	Partial profit (US\$/ha) <sup>y</sup>			Sclerotia produced (kg/ha) <sup>z</sup>
		\$0.33/kg	\$0.44/kg	\$0.55/kg	
No	270,000	1,257.4 b	1,735.9 c	2,214.3 c	–
	345,000	1,296.5 a	1,799.0 b	2,301.5 b	–
	420,000	1,306.0 a	1,822.8 ab	2,339.6 ab	–
	495,000	1,329.4 a	1,865.1 a	2,400.8 a	–
Yes	270,000	1,216.3 bc	1,680.7 b	2,145.1 c	0.08 b
	345,000	1,243.1 ab	1,727.5 a	2,212.0 ab	0.20 ab
	420,000	1,254.4 a	1,753.6 a	2,252.8 a	0.29 a
	495,000	1,192.7 c	1,682.4 b	2,172.1 bc	0.37 a

<sup>x</sup> Site-years were separately analyzed depending on the absence or presence of SSR. A site-year was labeled as SSR absent if the mean nontreated disease severity index was <0.5, and a site-year was labeled as SSR present if the mean nontreated disease severity index was >0.5.

<sup>y</sup> Partial profits were calculated by subtracting the cost of seed for each respective treatment level from the income determined by yield multiplied by the three presented grain sale prices. Within columns of each respective disease presence subgroup, means followed by similar letters are not statistically different as determined by Fisher's least significant difference ( $\alpha = 0.05$ ).

<sup>z</sup> Predicted quantity of *Sclerotinia sclerotiorum* inoculum produced and released back into the field. Quantities were predicted by 1 kg of produced inoculum for every 10% incidence of SSR as determined by Lehner et al. (2017). Within columns, means followed by similar letters are not statistically different as determined by Fisher's least significant difference ( $\alpha = 0.05$ ).

**Table 4.** Effects of fungicide programs on economic partial profits and estimated *Sclerotinia sclerotiorum* inoculum returned to soil when Sclerotinia stem rot (SSR) was either absent or present

Disease presence <sup>w</sup>	Fungicide program <sup>x</sup>	Partial profit (US\$/ha) <sup>y</sup>			Sclerotia produced (kg/ha) <sup>z</sup>
		\$0.33/kg	\$0.44/kg	\$0.55/kg	
No	Standard	1,256 c	1,769 b	2,282 a	–
	Model	1,301 b	1,809 ab	2,318 a	–
	Nontreated	1,336 a	1,838 a	2,339 a	–
Yes	Standard	1,216 b	1,716 a	2,153 a	0.11 b
	Model	1,214 b	1,695 a	2,175 a	0.26 ab
	Nontreated	1,250 a	1,723 a	2,196 a	0.34 a

<sup>w</sup> Site-years were separately analyzed depending on the absence or presence of SSR. A site-year was labeled as SSR absent if the mean nontreated disease severity index was <0.5, and a site-year was labeled as SSR present if the mean nontreated disease severity index was >0.5.

<sup>x</sup> All fungicide applications were made with picoxystrobin at a rate of 163 g/ha. The standard program consisted of applications at both the R1 and R3 soybean growth stages. The model program consisted of applications following SSR risk models called Sporecaster. A nontreated control was also included.

<sup>y</sup> We calculated partial profits by subtracting the cost of seed for each respective treatment level from the income determined by yield multiplied by the three presented grain sale prices. Within columns of each respective disease presence subgroup, means followed by similar letters are not statistically different as determined by Fisher's least significant difference ( $\alpha = 0.05$ ).

<sup>z</sup> Predicted quantity of *Sclerotinia sclerotiorum* inoculum produced and released back into the field. Quantities were predicted by 1 kg of produced inoculum for every 10% incidence of SSR as determined by Lehner et al. (2017). Within columns, means followed by similar letters are not statistically different as determined by Fisher's least significant difference ( $\alpha = 0.05$ ).

and that standard applications at the R1 and R3 growth stages are capable of managing SSR better than not treating with fungicides. However, the fungicide application programs based on Sporecaster models were inconsistent at reducing SSR levels and protecting yield (Figs. 2A, 3, and 4). The model application program resulted in similar SSR levels as the standard applications in narrow row spacing in the fully integrated trials but did not differ from the nontreated controls in the wide row spacing of the fully integrated trials or in the partially integrated trials (Figs. 2A and 5A). This finding suggests that the model program is capable of reducing SSR levels when an effective fungicide is used in conjunction with the Sporecaster tool. Preliminary observations have indicated that use of picoxystrobin has become increasingly inconsistent in reducing SSR compared with other fungicide options (Mueller and Smith 2019). The use of a different fungicide could have led to improved efficacy when the model program was used. The models used in this study showed high accuracy in predicting disease risk, especially in nonirrigated conditions, but accuracy levels lower than desired were seen in the irrigated conditions (data not shown). Thus, further adjustments to these models could increase accuracy in predicting disease risk, and this could better predict the optimal timing for applications. Ideally this method would prevent unnecessary fungicide applications, increasing financial savings and reducing selection pressure on *S. sclerotiorum* populations to develop fungicide resistance.

The finding of improved yields due to the standard fungicide application supports previous work (Willbur et al. 2019b). However, this improved yield did not always translate to increased partial profits, because of the exceptionally high cost associated with fungicide use. When SSR was either present or absent, the nontreated control profited the highest at the \$0.33/kg sale price, but as the grain sale price increased, both fungicide programs became as profitable as the nontreated. However, the standard application program became as profitable as the nontreated at a medium sale price (\$0.44/kg) when SSR was present and became as profitable as the nontreated only at the highest sale price (\$0.55/kg) when SSR was absent (Table 4). This finding shows that the use of the standard application program becomes more profitable at a much lower grain sale price when environmental conditions favor the development of SSR. Additionally, when SSR was absent, the standard program was less profitable than the model program at the \$0.33 sale price, because of the high cost of two applications in the standard program compared with the lower cost of potentially reduced applications by the model program (Table 4). However, this trend was not seen when SSR did develop because the standard and model applications profited similarly at all grain sale prices. Although there was a decrease in partial profit due to fungicide use at the lower grain sale prices, it must be noted that fungicide use for controlling SSR may be more economically beneficial when SSR incidence is at higher levels. As shown by Willbur et al. (2019b), yield losses decrease significantly at a 40% DIX and decrease more rapidly around 68%.

The use of fungicides also reduced the predicted production of sclerotia, providing the added benefit of reducing damage to future crops. The standard application program resulted in the lowest levels of sclerotia, because of the lower levels of SSR found from this treatment (Table 4, Figs. 2A and 5A). Minimizing the amount of new inoculum produced in a field is paramount in protecting future crops from this disease. As inoculum density increases, a higher percentage of mature apothecia will develop from sclerotia, leading to more ascospores released into soybean canopies and a greater number of infections, culminating in greater yield losses (Clarkson et al. 2014). As shown by Lehner et al. (2017), every 10% increase in SSR incidence led to mean yield losses of 172 kg/ha. As with all current management practices for controlling SSR, prevention of disease development is of the utmost importance.

Our analysis shows that wide row spacing, low seeding rates, and foliar fungicide treatments all individually reduced SSR levels. However, wide row spacing and lower seeding rates also decreased yields. A low seeding rate (270,000 seeds/ha) led to decreased partial profits across three grain sale prices in environments where SSR either developed or did not develop. A high seeding rate (495,000 seeds/ha) also

led to decreased partial profits across three grain sale prices, but only in environments where SSR developed. The use of fungicide programs was found to be less profitable than not spraying at a low grain sale price, but these costs were recovered in higher sale prices. It was also found that the use of lower seeding rates and the application of fungicides were capable of minimizing the predicted production of new inoculum. In order to balance these responses, environmental conditions and field history should be considered when management decisions are made. Fields with a history of high SSR pressure may benefit from the use of wide row spacing, using prescription seeding rate technology, or use of fungicides to minimize SSR development and protect yields. Fields without a history of SSR development continue to benefit from local planting recommendations in order to optimize yields.

## Data Availability

Data and analyses are available at <https://github.com/rwwebster/Sclerotinia-Stem-Rot-Integrated-Management>.

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