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## Integrated pest management strategies for the control of white mold (*Sclerotinia sclerotiorum*) in cultivated soybean [*Glycine max* (L.) Merr.]

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Integrated Pest Management Strategies for the Control of White Mold (*Sclerotinia sclerotiorum*) in Cultivated Soybean [*Glycine max* (L.) Merr.]

by

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A creative component submitted to the graduate faculty  
in partial fulfillment of the requirements for the degree of

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## DEDICATION

I dedicate this paper to my wife Norelys Walker for all her support for the many nights that I have spent putting into this paper, I could not have done this without you. I also dedicate this paper to the late Dr. Cam Perkins, who without your advice, I would not be here today.

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## **Abstract**

*Sclerotinia sclerotiorum*, a fungal pathogen in crops worldwide that causes white mold, can be devastating on yields if not controlled. Due to the complex nature of this disease, a well-defined integrated pest management, IPM, approach should be used to help minimize its impact on crops, more specifically, soybean. The most useful of these methods would include implementing wider row spacings, proper tillage practices meant to bury sclerotia, crop rotations to break pathogen cycle, planting partial resistant cultivars, chemical applications, and biological control methods.

## **Introduction**

White Mold, caused by *Sclerotinia sclerotiorum*, is a highly destructive disease. It infects numerous economically important crops, and overall 408 plant species worldwide (Workneh and Yang. 2000). This creative component describes different integrated pest management (IPM) strategies for the control of Sclerotinia stem rot, SSR, in soybean, [*Glycine max* L. (Merr.)]. Additionally, the need for novel approaches to control SSR in soybean are included that can reduce the devastating impact of the disease. Although, soybean growers want to buy commercial cultivars with high levels of disease resistance, no variety with complete resistance against SSR is available. Furthermore, limited fungicide options are available to control this disease only partially in soybean. Therefore, a balanced IPM approach is necessary to reduce the impact of this pathogen.

Integrated pest management is any broad-based approach that helps to suppress pest populations below the economic injury level (Willbur J et al. 2016). The foundation of IPM is to

control pest populations, not eliminate them. The main IPM approaches include tillage, mechanical cultivation, chemical applications, mechanical sanitation, and the use of resistant or tolerant cultivars. These approaches are grouped into three categories including cultural practices, varietal resistance, and chemical and biological control agents. The list can be quite extensive on available approaches, but with a disease such as SSR, no single approach can be used due to an incomplete understanding of resistance mechanisms (Peltier et al. 2012)

*Sclerotinia sclerotiorum* is a fungal pathogen found naturally in the soil. Also known as Sclerotinia stem rot, SSR of soybean is most prevalent in cool moist environments. Narrow spaced soybeans provide a quick canopy closure which lead to higher yields. These conditions



Figure1. Apothecium in north Nebraska field

also lead to a higher presence of the disease due to the moist environment it gives the pathogen to thrive in (Yang and Navi, 2006). The fungus produces a survival structure called a sclerotium.

This sclerotium structure is hard, black, and resembles a grain of rice. It is this structure that gives the fungus an excellent advantage to be able to survive numerous years in the soil before conditions allow it to germinate (Peltier et al. 2012). Proper conditions for germination include a cool and moist environment, often achieved in the North Central US under drill-seeded soybeans. Under these favorable conditions the sclerotium can germinate via two different ways, carpogenic or myceliogenic. Myceliogenic germination involves rapid mycelium growth throughout the soil while carpogenic germination is through the formation of an apothecium (Novakowski et al. 2017). Due to SSR's

infection of soybean via ascospores, this paper will focus on carpogenic germination. The apothecium will then germinate under cool and wet conditions and release ascospores. These ascospores land on senescing flowers and then colonize them saprotrophically. After colonization, mycelium will develop and can rapidly infect green tissues including stems, leaves, and pods that will eventually end in plant death (McDonald and Boland, 2004). Apothecium (Figure 1.) is a small, cupped-shaped mushroom-like structure that releases ascospores that infect the soybean plant directly on the dying leaves at the R2-R3 stages. McWilliams DA et al. (1999) provide an excellent reference on the different soybean growth stages. One important area to focus on when trying to control this disease is by removing apothecium structure from the soil surface by burying with tillage equipment. There are many factors that contribute to a sound control plan for SSR. One important control measure includes suppressing apothecium structure from producing ascospores, as it essentially removes the ability to cause infection. The disease cycle of Sclerotinia stem rot includes the following steps: (1) Sclerotia survive in the soil for up to 5 years; (2) under cool and wet environmental conditions, sclerotia germinate to produce apothecia; (3) apothecia produce sexual spores called ascospores; (4) ascospores are released into the air to infect senescing flowers; (5) symptoms include white tufts of mycelium found on the stem and leaves, bleached stems, heavy lodging and plant wilt, and plant death; (6) hard



*Figure 2. Sclerotia and mycelium at R5*

black sclerotia are formed on stems and pods and are dropped on the ground after they are dislodged by mechanical harvest, closing the cycle.

The first symptoms typically appear in the earlier stages of pod development during growth stages R3-R4 (Grau et al. 1994). Chlorosis and wilt are some of the early signs of this disease that can often be mistaken for other rots such as *Phytophthora* root rot and brown



*Figure 3. Shredded/bleached stem in center plant.*

stem rot. The vein tissues remain green while tissue between veins turn slightly grayish-green. The obvious signs around R5-R6 stages are the small, black sclerotia and fuzzy white mycelium on the stems of the plants shown in Figure 2. At this time, management can be difficult due to the disease's presence inside the stem. Late stages of the disease show stem collapse at the nodes and a very shredded appearance on the stems, with very poor pod fill (Figure 3), resulting in reduced yield. Diseased plants are easy to spot in late stages

with their bleached white stems and shriveled or empty pods as seen in Figure 4.

Danielson et al. (2004) concluded that for every 10% increment in incidence of SSR observed at beginning maturity in soybean, yield can be reduced by 0.13-0.34 tonnes per hectare. This is alarming considering the national soybean average for 2019 is right around 3.17 t/ha. If these average yields are expected in regions and fields where SSR damage occurs, for every 10% increment in the disease, yield can be reduced by 4.2-10.6%, which could be a heavy loss for a grower. While breeding for this disease has



*Figure 4. Diseased plant on left with healthy plant on right.*

been very challenging due to a lack of major gene controlling resistance to SSR, it is also possible that inadequate efforts are made to develop resistant cultivars due to the sporadic nature and irregular pattern of the disease. A “perfect storm” is needed to induce sexual reproduction of ascospores, which does not happen routinely each year. Furthermore, the complexity of the disease and a lack of knowledge of resistance mechanisms make control of this disease very cumbersome. These issues drive the need to conduct more research and hopefully provide growers with a cultivar with high genetic tolerance to give high yield in fields where the pathogen is present. In the next sections, major IPM strategies are explained to mitigate losses due to SSR in soybean.

## Row Spacing

SSR requires a cool and moist environment to develop. Row spacing can have an impact on disease incidence, especially using a narrower row spacing, which tends to give the cool and moist environment needed for pathogen infection. Soil temperatures are lower and soil moisture and humidity are higher in narrow spaced soybeans (Yang and Navi, 2006). Berglund et al. (1998) found a direct relationship with % sclerotinia and plant population density in narrow rows. As plant population density increased, % sclerotinia increased and seed yield decreased in one year. However, they did not observe this correlation in a different year, which could be explained by a dry and warmer period during flowering that year.

High-yielding soybean varieties excel on a narrow row-spacing (Pederson, 2007). This ensures a thick and dense canopy that is favorable for moist conditions allowing the germination of sclerotia into producing the apothecium which in turn infect soybean plants. However, in an experiment performed by Buzzell et al. (1992), quite the opposite occurred. While row width did not affect the incidence of stem rot, the 69-cm row width resulted in lower ( $P = 0.05$ ) yield than the 23-cm and 45-cm row widths (Buzzell et al. 1992). It should be noted that the cultivar 'Maple Arrow' was thought to be responsible for this lack of row width correlation and disease incidence due to its resistance mechanisms preventing the spread of SSR.

Often there will not be a single simple trend to follow as to why incidence occurs using any IPM approach. Their study also indicated that narrower row spacings did in fact, increase disease incidence in some cases and that there was a trend towards more stem rot in the 23-cm

spacing in the three cultivars, Evans, S1346, and Corsoy 79. These observations could be due to the tolerance to the pathogen in some of the cultivars, which would explain a lack of disease by row-width interactions. Yang and Navi (2006) found that despite row spacing, disease incidence can be quite similar between wide and narrow row soybeans, especially with susceptible varieties when cool and moist conditions are present. They concluded that it is best to completely avoid disease incidence by not planting soybean in row spacing less than 38.1 cm in areas where SSR has caused severe yield losses in the past. Even though yield is reduced by high incidence of disease in narrow-spaced plantings, yield has been found to be substantially greater, up to 20%, than in wide-row systems (Costa et al. 1980; Oplinger and Philbrook, 1992). Therefore, there is a conflicting choice for growers – plant in narrow rows to get high yield but with a higher disease risk or plant in wider rows to compromise yield, but reduce the disease risk.

In summary, it may be best practice to use narrow spaced systems due to higher yield potential that can overcome yield losses due to disease. If heavy disease incidence was present in the past, it is best to use wider row spacing to reduce moisture buildup in canopy.

## **Tillage**

A key focus of mitigating SSR is in the control of the sclerotia structures present in the soil. If the apothecia are below the soil surface, they are not able to disseminate their ascospores and infect the dying leaves of soybean after flowering, thus avoiding the disease. One method of control is to incorporate different tillage strategies in order to reduce the chance that sclerotia can germinate. Workneh and Yang (2000) conducted a study on the

prevalence of sclerotinia stem rot of soybeans in relation to tillage and concluded that, there were significant differences among tillage categories in prevalence of Sclerotinia stem rot. Most notably, the prevalence of the disease was far lower in no-till fields than in either conventional or minimum-till fields. Workneh and Yang (2000) found no significant differences between minimum and conventional till in relation to % of fields with SSR, but found a dramatic decline in SSR in fields with no-till showing a significant difference ( $p = 0.05$ ). Since there are many factors that influence the spread of the disease, a possible explanation for how a no-till system had far lower prevalence was due to less dense canopies associated with no-till compared to tilled fields. This less dense canopy provides less favorable conditions for germination. Another explanation offered was that in tilled systems the sclerotia are more evenly spread throughout the field by mechanical dissemination of the equipment and thus lead to a higher prevalence. Moreover, a lower incidence of disease was potentially found in no-till fields due to generally higher levels of microbial activity that would help to degrade the sclerotia (Workneh and Yang, 2000).

For the past three decades, a push from conventional tillage to conservation tillage has been implemented for soil conservation in the north-central United States. This, along with a shift from wider row spacing to narrower row spacing to help control weeds, has unfortunately given SSR more opportunity to infect soybeans. More information generate more questions about the best methods for control. For example, when burying the sclerotia under at least 10 cm of soil, which is common in conservation tillage, production of apothecia can be delayed, which in turn will reduce disease incidence. This method comes at the cost of risking higher soil

loss due to erosion and, as mentioned previously, is becoming less popular as soil conservation is paramount.

Dorrance and Mills (2008) recommend one year of moldboard plowing to bury sclerotia up to 10 cm deep upon the first encounter with the pathogen. A reduced tillage regimen is best for fields with a long history of the disease. The inconsistencies among all studies comparing tillage practices (Peltier et al. 2012) on disease incidence shows that more research needs to be done on this topic. A large-scale research project that would consider many different environments in the north central United States under many soil types and tillage plans would help provide a more consistent tillage control plan to help growers mitigate damage from this destructive pest.

## **Planting Date**

Most growers want to get their beans in the ground as soon as possible to maximize yield potential. Many row crops such as maize benefit greatly from an early planting date, soybean is no exception. The planting date for soybeans will vary from year to year and region to region (Pedersen, 2006). Pedersen states that regardless of diseases like SSR, planting should not be delayed because management practices such as variety selection, adjusting planting rate and row width will be sufficient to control this pest. However, this seems to be an oversimplification. Although it is true that early planting dates will typically lead to higher yield potentials to possibly offset the damage SSR can cause, white mold's ability to infect comes from climatic conditions during flowering. Only a full IPM approach that encompasses all the

cultivation, varietal, and chemical/biological agents, should be used to give the best chances for control of this pest.

Adjusting the planting date of soybeans can have a wide scope of impacts for many plant diseases other than SSR. Every state has an optimal planting date based on large experimental data and this information is disseminated by extension scientists and University researchers that promote soybean to be planted as early as possible to miss the spring frost and excessively cool soils. By planting early avoiding spring frost gives soybean the ability to take full advantage of the entire growing season and produce maximum yields. An early planting date may keep pathogens such as *Phytophthora* from infecting soybeans, due to its preference to warm soil. On the other hand, a delayed planting can help mitigate the impacts of pathogens that prefer cooler soils, such as some species of *Pythium* and *Fusarium solani* (Berglund et al. 1998). There are also many diseases that will occur despite when seeds are planted because they all infect throughout the season, such as: bacterial blight, brown spot, and stem canker. SSR, is in a different category altogether.

Instead of having a direct impact on disease occurrence, SSR has an indirect impact (Berglund et al. 1998) because it does not occur during the early stages, but rather during flowering. Planting early or late could be viable options to reduce the impact of SSR on soybeans, it all depends on whether cool and wet conditions persist during flowering. Yang (1998) listed the effects of planting date for soybean diseases in fields where disease is a concern. They noted that early diseases, before V2, such as damping off by *Pythium* (cool and wet soil), *Rhizoctonia* (warm soil), and *Phytophthora* (warm and wet soil) can be induced by either cool and wet warm and warm and wet soils, respectively. Yang concluded that an early

planting date in all regions will help lower the risk of infection in these three diseases mentioned above. Later diseases such as, brown stem rot, SSR, and pod and stem blight all have varied effects from altering plant date as they are all more complex diseases and growth state for infection cover later stages such as flowering and pod setting.

In summary, adjusting planting dates are important for reducing the effect of numerous diseases and maximizing yield potential in soybeans. However, adjusting planting dates has minimal impact on SSR suppression.

## **Crop Rotation**

Implementing a crop rotation with non-host crops can have a significant impact on increasing yields while minimizing inputs, reducing the cost of external inputs, and by promoting crop health through breaking pathogen and insect cycles (Maloney and Grau, 2001). For soybean specifically, studies have shown that continuous soybean can lower yields, especially for fields with prior SSR pressure. Although a corn and soy rotation allow corn to escape diseases by breaking insect and pathogen cycles, sclerotia can survive up to 5 years in the soil. This unique survival ability is another driving force as to why a full IPM strategy needs to be implemented for the best control of this pathogen.

Due to growers shifting their cultivation practices to maximize yields by planting earlier, reducing row widths, planting higher population densities, and using more reduced tillage methods, SSR has had the ability to proliferate, mainly in the northern states where cool and moist conditions occur more frequently. All these conditions aided infections with *Sclerotinia* stem rot. Mueller et al. (2002) designed an experiment to investigate the effect of crop

rotation under three tillage systems on the number and distribution of sclerotia and apothecia development as well as SSR incidence in soybean fields, along with a varied tillage system, from conventional to no-till. Plots were first grown in 1994 in Northern Illinois under a previous rotation of corn-soybean. The three rotations used were (corn-corn-soybean; soybean-corn-soybean; and continuous soybean). In 1997 plots were used to evaluate the effect of these three crop rotations using cultivars, 'BSR 101' (susceptible), 'NK S19-90' (moderately resistant) and 'NK S21-20' (unknown resistance). These cultivars were planted using a no-till drill at 19-cm row spacing at 494,000 seeds per hectare. The crop rotation regime did not influence the number of sclerotia or apothecia, SSR, or yield in 1996. This was likely a direct cause of having only a 1% incidence of SSR in all plants monitored in 1995 and 1996. However, 1997 showed a strong correlation between rotation and total number of apothecia, showing the highest incidence for continuous soybean, even though it had the highest overall yield. Mueller et al. (2000) concluded that crop rotation may not affect SSR incidence much because of sclerotia's ability to survive up to 5 years in the soil (Schwartz and Steadman 1978). Also, there are alternate hosts that *S. sclerotiorum* can infect such as different weed species that can lead to an infestation in soybean. This is yet another reason, why it is essential that an integrated approach must be taken to control SSR in soybean.

Measures like 2-3 years of non-host crops in rotations such as corn and small grains can reduce sclerotia numbers in soybean (Gracia-Garza et al. 2002). This is no surprise, as many different cropping systems benefit from utilizing crop rotations to break pest cycles and ensure healthy crops with maximum yields. Some growers are not able to use these rotations and will

have to rely on other cultivation approaches for white mold management, or the most significant method, using cultivars with higher levels of resistance.

## **Cultivar Resistance**

The dream for any grower today is to buy a bag of seed with full resistance to their problem disease. There are many cultivars today with partial or full resistance to different diseases. As mentioned before, SSR is not a simple disease that can be controlled by using one method of control. A fully resistant cultivar does not exist today for SSR mainly due to its complex nature and the fact that many genes control its function, so it is difficult to breed for complete SSR resistance (Arahana et al. 2001). Growers rely on partially resistant cultivars as well as a balanced IPM strategy to reduce the impact of SSR on their crop. Currently there are many different soybean cultivars available with a moderate amount of resistance that growers have at their disposal (Peltier et al. 2012). Variety selection should be the first line of defense to combat SSR (Yang and Navi, 2006). This statement has a lot of truth to it because as a grower, they want to rely on the simplest form of control to get the returns on their crops, and planting a resistant variety will be the most profitable way for soybean production.

Kim and Diers (2000) paper studied the inheritance of partial resistance to sclerotinia stem rot by mapping QTL responsible for resistance. The experiment involved 152 F<sub>3</sub>-derived lines from the cross of a partially resistant line and a highly susceptible line, Novartis Seeds S19-90 and Williams 82, respectively. Lines were tested from 1996-1997 at East Lansing, MI and Zilwaukee, MI. The East Lansing location was artificially inoculated prior to planting with sclerotia from screenings of dry bean and was sprinkler irrigated while the Zilwaukee fields

were naturally infested with *S. sclerotiorum* with no irrigation. Plots were rated for disease severity around the beginning of physiological maturity (R7) (Fehr et al. 1971). The disease severity between lines was significant at each environment and across environments, except Zilwaukee in 1996. Across all environments, the broad-sense heritability estimates for disease severity index (DSI) was 0.59, indicating that resistance can be selected for in the field effectively. DSI was also significantly correlated with lower yield. Due to the great difficulty of evaluating this disease in the field, breeders would benefit from the ability to select for resistance using genetic markers (Kim and Diers, 2000). They concluded that genetic markers on linkage groups K may hold the key for physiological resistance and would aid greatly in marker-assisted selection for future studies.

As mentioned before, the use of lines with a form of genetic resistance to SSR is paramount in building a solid IPM strategy for growers with this fungal pathogen. Further understanding of white mold resistance mechanisms in soybean must be a high priority for the improvement of commercially available resistance (Willbur et al. 2016).

For growers in eastern Nebraska, SSR resistance is not a topic often heard, this is likely due to the sporadic nature of infested fields and conditions that are not always met for the sclerotia to germinate. Regardless, this disease, when present, can lead to dramatic yield loss and further research on resistance mechanisms should be conducted to help these growers combat this pest. SSR proves to be an elusive disease and as climatic conditions vary every season across all environments, we must remain vigilant in using as many strategies as necessary to combat this highly problematic disease. Although the use of partial-resistance

cultivars is highly beneficial for growers in problem areas, there are still more IPM strategies at the disposal to reduce the impacts of this disease.

## **Chemical Applications**

The application of chemicals to fields to help control disease is still a highly popular method for control for a myriad of plant diseases today, and SSR is no different. One would think that just a simple fungicide application would be able to control a fungal pathogen such as *S. sclerotiorum*, but as we have learned before, this disease is more complicated than that. It is true that the use of different chemical applications can in fact lower disease incidence in soybean, however, the application window for use can be quite narrow (Dorrance and Mills, 2008). None of the current fungicides on the market provide complete control for SSR. Fortunately, there are multiple options for control via fungicides (Peltier et al. 2012). There could be several possible reasons leading to fungicides' inconsistent efficacy on SSR, but Peltier et al. (2012) reported that the inability of fungicides to move up or down plants could be the main reason behind this observation. This is an interesting point as many chemical classes are systemic, they can move inside the plant for more targeted specificity of diseases. The fact that fungicides' efficacy depends on contact with the pathogen, and symptoms do not typically show up until after the R3 stage, it can be a challenge for the fungicide spray to reach through a dense canopy where the pathogen resides. This is where early scouting is crucial to get ahead of this disease by observing the sclerotia and apothecia in and around the soil so that spraying regimes can be implemented before R3 stage, when symptoms start becoming visible.

The control of white mold using fungicide treatments were effective when applied at the correct time, an adequate volume, and using the appropriate spray nozzles (Mueller et al. 2002; Juliatti; Juliatti, 2010; Wutzki et al. 2016). Wutzki et al. (2016) verified the efficiency of using multiple modes of action (MOA) being applied alone and in rotation at different growth stages. They concluded that applications of the four fungicides: fluazinam, procymidone, methyl thiophanate, and carbendazim resulted in lower incidence levels of SSR on soybean. The most interesting finding in this study was the fact that no control was observed when applied after the R3 stage (beginning pod stage). This goes back to the narrow window of time when application of fungicide is effective. At the R3 stage, the pathogen has already inoculated in the dying flowers, and fungicide does no good at this point. Another reason why proper timing and knowledge of the disease are big factors when controlling *S. sclerotiorum*. Their conclusion stated that the use of fungicides is key in reducing sclerotia in the fields to reduce inoculum present in the soil for better control for future soybean crops. Willbur et al. (2016) stated that only a few products were effective in controlling SSR in soybean and timing of application is critical for maximum efficacy. There are even herbicides that can prove to be effective in reducing SSR incidence, including Cobra or Phoenix. These herbicides have an indirect impact on the disease incidence by modifying the canopy and delaying or reducing flowering, which is when the disease infects the plant. However, these herbicides can also reduce yields by causing crop damage, especially in years not conducive for disease (Dann et al. 1999; Peltier et al. 2012).

There are five main groups of fungicides effective in reducing SSR incidence levels including methyl benzimidazole carbamates (MBC), such as methyl thiophanate mentioned

above, demethylation, succinate dehydrogenase, and quinone outside inhibitors (DMI)(SDHI)(QoI) respectively (Armando et al. 2015; Di et al. 2016; Huzar-Novakowski et al. 2017; Liang et al. 2015; Peltier et al. 2012; Willbur et al. 2019). Also, Fluazinam, an uncoupler of oxidative phosphorylation has been found to be effective in inhibiting SSR (Liang et al. 2015). The use of different modes of action is often necessary when trying to control pests due to the unique ability for organisms to overcome, or build resistance, to a chemical. The MBC class of fungicides work by inhibiting fungal cell division. Other classes such as SDHI, QoI, and uncouplers work by inhibiting cellular respiration and energy production of *S. sclerotiorum* by interfering with its electron transport chain; while the DMI fungicides inhibit sterol biosynthesis that results in a change in fungal cell wall development (Peltier et al. 2012). All these classes of fungicides either work to inhibit spore germination or simply slow fungal growth, where the herbicides alter canopy development to promote systemic resistance (Willbur et al. 2019; Peltier et al. 2012; Dann et al. 1999).

The canopy can be quite dense in drill seeded high yielding soybeans. As mentioned before, this can lead to adventitious growing conditions for SSR to develop. When spraying fungicides in dense canopies to help control SSR application, coverage is particularly important. Canopy, spray volume, and droplet size all influence coverage (Derksen et al. 2008). A spray nozzle with a flat-fan with high-fine to mid-medium droplets often perform the best in penetrating through dense canopies (Willbur et al. 2019). When applied as early as the fourth trifoliolate stage (V4), Cobra (lactofen) has been known to suppress SSR for years (Yang et al. 1999) if there is a confirmed presence of the disease. Another useful IPM method of control for

SSR in soybeans, chemical application remains a highly popular option for growers for the control of this devastating pest.

Due to the popularity of using chemical control to combat diseases by growers, a cost-benefit analysis among various chemicals and disease incidence levels would be great information for any grower with SSR. SSR is considered problematic on approximately ~7.7 million hectares of the soybean production area in Brazil (Meyer et al. 2016a; Barro et al. 2019). Soybean yield losses from SSR in Brazil can reach 50 to 70%, in higher elevation regions (>600m), where the cooler weather conditions are conducive for infection (Lehner et al. 2016; Barro et al. 2019). Among all the various strategies mentioned here, the use of chemical fungicides still remains the most effective tool for protection of soybeans during flowering after primary infection occurs (Meyer et al. 2016a; Mueller et al. 2002; Sumida et al. 2015; Wutzki et al. 2016; Barro et al. 2019).

Madden et al. (2016) concluded that network meta-analysis (NMA) is a more powerful approach that provides a more precise estimate and has the ability to overcome limitations of the traditional pairwise meta-analysis (PMA) by using multiple comparisons among treatments of interest, in which PMA only compares two treatments at a time. The use of any of the applied fungicides resulted in reduced SSR incidence and sclerotia production compared to nontreated (Barro et al. 2019). The study concluded that the probability of breaking even on fungicide costs for the high-disease scenario was <65% for FLUZ, the more expensive and effective fungicide, as compared to the lower cost TMET fungicide. Profitability was less likely for the low-disease scenario and was more dependent on fungicide costs and soybean price.

This is good news for growers that have concerns about SSR in their fields. In situations where disease incidence is lower, they are less likely to benefit from a chemical application. In fields where SSR incidence is high, an application of FLUZ, when specifically applied at the recommended window, has the potential to be a cost-effective form of control of this pest.

## **Biological Control**

Biological control agents are the use of living organisms to lower pest populations to a more manageable level by often taking advantage of a biological's food source, in this case, the food source is *S. sclerotiorum*. A great benefit using biological controls for soybeans is that they can also be used on organic crops as well (Peltier et al. 2012). So far, little known information on the biological control of *S. sclerotinia* in soybean are available. Pest control methods that are safer to use on the environment are preferable over highly toxic chemical treatments that are currently applied. Fortunately, there are multiple different organisms that feed on *S. sclerotiorum* that growers can use to manage the impact that SSR has on soybeans (Bailey et al. 2010).

*Trichoderma* species are one such soil-borne saprobe that acts as a mycoparasite on *S. sclerotiorum* (Harman et al. 2004). By secretion of chitinases,  $\beta$ -1,3-glucanases, proteases and secondary metabolites, *Trichoderma* species help to control SSR of soybean (Geraldine et al. 2013; Monte, 2001; Shirmbock et al. 1994). Sumida et al. (2018) conducted research on two *Trichoderma* strains (T25 and T42) on the *in vitro* antagonism and the effects of crude organic solvent extracts from these two strains against nine strains of *S. Sclerotiorum*. The antagonism towards *S. sclerotiorum* by *T. asperelloides* T25 and T42 extracts showed high potential as a

biological control agent, especially T25, being highly effective against all nine strains. Sumida et al. (2018) also found that control of SSR in soybean plants using T25 and T42 extracts using commercial products containing *T. harzianum* spores shows a strong promise for potential use on the market in this region. Geraldine et al. (2013) also found the two strains to be effective in reducing apothecia density and the severity of SSR.

*Bacillus subtilis* is another soil-borne microbe that has shown promise in controlling SSR in several crops in the U.S. Zhang and Xue, (2010) tested the *in vitro* antagonistic activities of SB24, a *B. subtilis* strain against *S. sclerotiorum* under control conditions as well as observing *B. subtilis* population dynamics in both controlled and field conditions in 2006 in Canada. All three preparations (cell suspension, broth culture, and cell-free filtrate) provided significant suppression of SSR in the greenhouse. However, each of the three preparations reduced in effectiveness over time, probably due to rain washing the bacteria off the leaves, making a reapplication necessary after rainfall. Without rain, the bacteria were able to survive on the leaf surfaces for up to 5 weeks. The authors concluded that a cell suspension preparation was the most likely candidate for a commercial formulation because it does not cause the environmental contamination caused by the foul odor of broth cultures. In soybean growing regions in the north U.S. states, rainfall is needed for high yielding conditions. For the growers who receive steady rainfall in the growing season may not benefit from using this biological agent for control. However, for growers who use a form of row irrigation in drier environments, that does not splash off the bacteria on the leaves, could possibly benefit from using *B. subtilis* strains, like SB24, for control.

Another biological control, that is the most widely used today, is *Coniothyrium minitans*. This fungal organism is a pathogen of *S. sclerotiorum*, (Huang and Hoes, 1976) which parasitizes the hyphae and sclerotia of white mold (Willbur et al. 2019). When compared to *S. lydicus* (43.1%) and *T. harzianum* (35%), *C. minitans* was observed by Zeng et al. (2012) to produce SSR DSI reductions in soybean by 68%. This fungal pathogen must be incorporated to a depth of 5 cm to be effective and provide contact with sclerotia for its degradation to occur (Dorrance and Mills 2008). A big advantage this control method has over fungicides is its ability to provide long-term control of the primary inoculum as opposed to just in-season control that fungicides have (Waldron et al. 2013).

There are limited research studies on the use of biological control for SSR in soybean by *C. minitans*. One such study was conducted by Waldron et al. (2013) from Cornell University, titled, "Enhancing integrated options to better manage soybean white mold using a biological fungicide." The fields that were chosen for the study had previous severe economic losses due to SSR. However, this study was unable to give useful results due to an unusually dry season, which is very unfortunate due to a lack of similar studies on the use of *C. minitans* for control of SSR in soybean. Waldron et al. (2013) also tried to create an environment conducive for infestation by using irrigation, with no success. This study shows the complexity of a white mold and how difficult it can be to study. Specific conditions must be met for infestation to occur each year, regardless of past severe losses due to the pathogen.

With such a high potential of these biological parasites for lowering SSR damage in soybean, more research is needed. Zhang and Xue, (2010) suggested that there could be many

more microorganisms that could be advantageous for controlling SSR, they just need to be found.

## Recommendations

All the mentioned IPM strategies are highly useful to aid in control for SSR. Since SSR development depends on multiple complex factors, (weather conditions, apothecial germination, flowering time, etc.), fungicide and biological applications can be ineffective, and even unnecessary (Willbur et al. 2019). The one key element that must be known before any method can be implemented is whether an infestation is going to occur, and it can often be too late if symptoms are detected. Of course, if it is known for certain that an infestation is going to occur, the pathogen would likely not be much of a problem, as with any crop disease. If only there was a way to forecast when the right conditions are present for white mold to develop.

Forecasting models of *S. sclerotiorum* have been developed for peanut, carrot, lettuce, and canola to determine if or when a fungicide treatment is necessary. There is currently one app-based phone model for soybean developed at the University of Wisconsin, called “Sporecaster.” This app was just developed in 2018 and is a huge step towards forecasting *S. sclerotiorum* in soybean in the US. Next necessary step forward is to begin with validation of this app in states outside of Wisconsin. As growers often depend on extension agents and crop advisors on handling disease problems, these apps need to be in the hands of every grower in the country that has problems with SSR. Nearly every person in America has access to a cell phone and more app-based models like this will have a dramatic impact on the control of complex diseases like SSR. There is a need in exploring different microorganisms and

hyperparasites for the control of SSR. Regardless of the methods used to help in the control, growers and researchers need to remain vigilant in the proper scouting of fields, recording information based on field history and previous infestations, and the consulting of your local extension agencies and crop advisors on how to best handle this disease.

## **Conclusion**

*S. sclerotiorum* can only be effectively managed using a diverse integrated pest management approach. Since there are so many different environmental and agronomic factors that can influence germination of sclerotia, a thorough knowledge of all the different IPM strategies for growers is crucial. As a first step, a grower needs to assess the level of damage experiences at their farm and the value of control. An integrated approach of control along with newer methods will be desirable, if needed for disease control. The potential of biological control for white mold suppression is high, as studies have shown with multiple different hyperparasites, and more biologicals should be tested for their efficacy in control of this devastating disease. Forecasting models, such as Wisconsin's "Sporecaster" app will be important in helping growers get ahead of SSR before it becomes a bigger problem. In the coming years growers across the north-central states should begin validating this model in hopes that updated models can be created for future control. In addition to IPM strategies, further breeding efforts need to be conducted to obtain a cultivar with higher level resistance to SSR; this can only be achieved with a more thorough understanding of resistance mechanisms to *S. sclerotiorum*. To an extent, we will always be at the mercy of mother nature,

but with the help of technology and efforts of researchers, yield losses from white mold may become a problem of the past.

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