REVIEW: DISEASE PROFILE

Soybean target spot caused by *Corynespora cassiicola*: a resurgent disease in the Americas

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Abstract

Soybean target spot, caused by *Corynespora cassiicola*, has historically been considered a minor disease for soybean crops since it was first reported in the USA, Brazil, and Argentina. However, changes in the agricultural practices, such as cultivar selection, crop succession, and the intensive use of fungicides of the same mode of action have favored inoculum survival and build-up. A resurgence of target spot has been reported in the main soybean-growing areas of the Americas where yield losses between 10 and 42% have been reported. The high frequency and severity of recent target spot epidemics have challenged farmers and ag-allied professionals for management practices to compensate for the resurgence of the disease. Even though resistance to the fungus within experimental soybean germplasm was documented during the late 1950s and early 1960s by soybean breeders, susceptible cultivars are dominant in the marketplace, and, as a result, soybean farmers have come to heavily rely on the use of foliar fungicides to minimize the potential economic losses associated with the disease. The limited number of studies assessing soybean yield losses caused by target spot or absence of environmental-based forecasting systems for this disease leave soybean farmers with difficult decisions regarding fungicide application. The high genetic diversity provides the pathogen, *Corynespora cassiicola,* an enhanced ability to adapt to different environments and infect a considerably broad range of plants. *C. cassiicola* is a multifaceted pathogen that can establish necrotrophic interactions with soybean by extracting nutrients from leaves, stems, pods, and seed, and to a much lesser extent roots, or even surviving in an endophytic relationship with soybean depending on environmental conditions. The current review is focused on the history of target spot in the Americas, fungal life cycle and disease symptoms, host range, the impact on soybean yield, and specific disease management measures. Research insights focusing on integrated disease management are proposed to improve target spot management in the future.

Keywords *Glycine max* · *Corynespora cassiicola* · Disease management · Disease cycle · Host range · Yield loss

Introduction

Soybean [*Glycine max* (L.) Merril] is considered the most important oilseed crop worldwide. Botanically, soybean is an annual legume plant in the family Fabaceae (Hymowitz 2008). The first historical evidence places the emergence of soybean as a food crop in northeastern China between 1700 and 1100 B.C (Hartman et al. 2011). The associated evidence is based on the extensive distribution of semi-natural wild soybean in the geographic area, but not in other regions (Fukuda 1933). Soybean has many commercial uses and is an important crop for human consumption as well as livestock. In addition, soybean is currently used to produce oil, which can be used to produce biodiesel, a common biofuel source. While soybean products are important and have numerous end-uses, the nutritional content of soybean

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is important as a source of food. One source of nutrients produced by the soybean plant is the grain that is harvested. Grain from soybean seed is composed of approximately 40% protein and 20% oil; such characteristics represent the greatest protein content and the greatest gross output of vegetable oil among the cultivated crops in the world (Singh 2010). The global soybean production for 2021 was estimated to be 385 M ton (USDA 2021). The USA (29%), Brazil (34%), and Argentina (17%) are responsible for approximately 80% of the total production (Faostat 2021).

Several important abiotic and biotic stresses threaten soybean production by reducing grain yield, threatening the production of soybean seed, and impacting the overall quality of either grain or seed (Hartman et al. 2011). Some of the major abiotic stress issues include improper fertilizer application that results in excess nutrients or nutrient deficiencies, heat, cold, drought, or excessively wet soils. Damage by plant pathogens and pests are the main biotic factors that can lead to reductions in soybean production throughout the world. In addition, and where some specific plant diseases occur, biotic stresses tend to be geographically and environmentally restricted. However, there are examples of disease-causing organisms that occur wherever soybean is grown. Previous estimates suggest that approximately 11% of attainable soybean production is negatively impacted by plant pathogens (Oerke 2006; Allen et al. 2017; Savary et al. 2019; Bradley et al., 2021). More than 40 fungal organisms have been reported to cause significant yield losses to soybean (Hartman et al. 2011) and are closely associated with the environmental conditions that occur within a cultivated region (Yang and Feng 2001). However, some soybean pathogens are more of a concern than others. For example, the downy mildew causal pathogen (*Peronospora manshurica* (Naumov) Syd.) is one that is widely observed throughout the global soybean-producing area; however, the fungus that causes downy mildew is not reported to result in significant yield losses (Yorinori 1992). Conversely, losses attributed to the soybean rust pathogen (*Phakopsora pachyrhizi* Syd.) can be severe given the ability of the disease to rapidly increase given the proper environment especially in unmanaged field situations (Kelly et al. 2015). Moreover, even though the losses associated with soybean rust have been more frequent in South America, the potential for soybean rust to cause significant yield losses is oftentimes most closely associated with the environmental conditions that occur prior to and immediately after infection. Curiously, the importance of target spot in soybean production has been reported on the North and South American continents, most specifically in the USA, Canada, southern Brazil, Paraguay, Uruguay, and Argentina (Savary et al. 2019).

Over the last several decades, the importance of some plant pathogens has been observed to increase and has generally been termed to be the result of a resurgence of plant diseases by previously reported, important plant pathogens. In the past, the specific definitions related to disease observations and how those situations should be reported by the scientific community. According to Milgroom (2017), the reemergence of a disease is characterized by the rapid increase of disease levels, such as disease incidence or disease severity. Oftentimes when a disease outbreak is attributed to the reemergence of a specific plant pathogen, these disease outbreaks are the result of a genetic mutation or selection of new pathogen races and/or more aggressive strains. For example, the reemergence of potato and tomato leaf blight has been reported in several countries throughout the world. The reemergence of potato and tomato late blight is mainly due to changes in the *Phytophthora infestans* pathogen populations on a worldwide basis that have occurred as a result of sexual recombination (Fry et al. 2015). In addition, some more important food crop diseases have been observed to be more severe. Most recently, the widespread reemergence of yellow rust has been attributed to new races of *Puccinia striiformis* f. sp. *tritici* which have been determined to be more prevalent as a result of sexual recombination within the pathogen population (Ali et al. 2014, 2017; Hovmøller et al., 2016). A similar situation was observed for stem rust with the emergence of the new strain, Ug99 (Singh et al. 2011). *Puccinia graminis tritici* strain Ug99, named after the initial observations of the pathogen in Uganda in 1999, emerged through somatic hybridization (Li et al. 2019). Ug99 has been considered a huge threat to food security due to the susceptibility of approximately 90% of all commercially available wheat cultivars to the stem rust fungus on a worldwide basis (Singh et al. 2011). In addition, in soybean production, outbreaks of target spot have been reported on the North American continent in the last decade leading to estimated yield losses between 10 and 42% (Allen et al., 2017; Bradley et al., 2021). However, based on the published literature and the specific situation as it relates to target spot, the situation as it relates to target spot in soybean production systems is hereby considered to be a resurgence. At present, the importance of target spot has been elevated from one of "minor" importance to a disease that may in fact result in substantial yield losses if the environmental conditions are conducive and prevalent over a substantial period of time.

The increase in the incidence of plant pathogens, the large cultivation of cultivars with narrow and limited sources of resistance, as well as the misuse and over application of fungicides contribute to the difficulty in managing plant diseases. In addition, major changes in the tillage practices employed by soybean farmers in some production regions have likely contributed in part to the importance of some specific soybean diseases. The use of reduced tillage, or no-till systems, tends to promote retention of crop residue and reduces the breakdown of crop residue which can factor into

the survival of some specific fungal pathogens. Maintaining crop residue on the soil surface is a beneficial production practice for several reasons including the reduction of erosion that results from rain and wind and the improvement of water conservation (Nunes et al. 2018). However, a slow degradation of crop residue favors the substrate to serve as a reservoir for numerous necrotrophic fungi to survive, reproduce, and serve as a source of primary inoculum to incite disease in the field in subsequent seasons (Baird et al. 1997). Countless organisms can survive as a result of this production practice, most of which are foliar disease-causing organisms as well as important root and stem disease-causing fungi. Additionally, the successive cultivation of a single crop without rotation can allow fungal pathogens to increase in their importance. More specifically, one of the important organisms that appears to have increased over the past several years is *Corynespora cassiicola* (Berk and M.A. Curtis) C.T. Wei, the causal agent of target spot. Current information regarding the pathogen history, the occurrence of target spot in the Americas, fungal life cycle and disease symptoms, host range, the impact on soybean yield, and insights on disease management are presented in this review.

History regarding the etiology of target spot

The mycological history of *C. cassiicola* is rather murky. In the early 1900s, cowpea [*Vigna unguiculata* (L.) Walp] and soybean in China were reported to be infected by a fungus that produced large and slender, pale olivaceous brown conidia (Wei, 1950). In the 1930s, the fungus was identified as *Cercospora vignicola* Kawamura (Tai, 1936; Teng, 1939). A decade later, Liu (1948) identified isolates purported to be the same fungus as *Helminthosporium vignae* Olive on cowpea in Japan. One of the first reports of target spot in the western hemisphere was made in 1944. Olive et al. (1945) attributed the defoliation of cowpea at La Place, Louisiana and Florida, to a previously undescribed species of *Helminthosporium* (*H. vignae* Olive). In addition, *H.*

vignae had previously been observed to be associated with fungal specimens that originated from cowpea in North and South Carolina and soybean in Florida in 1943. In summary, the genus that was initially reported to cause the leaf spots was misclassified as *Helminthosporium*. In the subsequent research efforts that have been conducted since the 1940s, the causal agent of target spot was reported to be *Corynespora cassiicola* (Berk and M.A. Curtis) C.T. Wei. Later a phylogenetic study determined that individuals from the genera *Corynespora* and *Helminthosporium* are polyphyletic, meaning they have a different ancestor (Voglmayr and Jaklitsch 2017). Since the initial report of target spot in the USA, the disease has been detected in several regions of the American continent, particularly where soybean is grown.
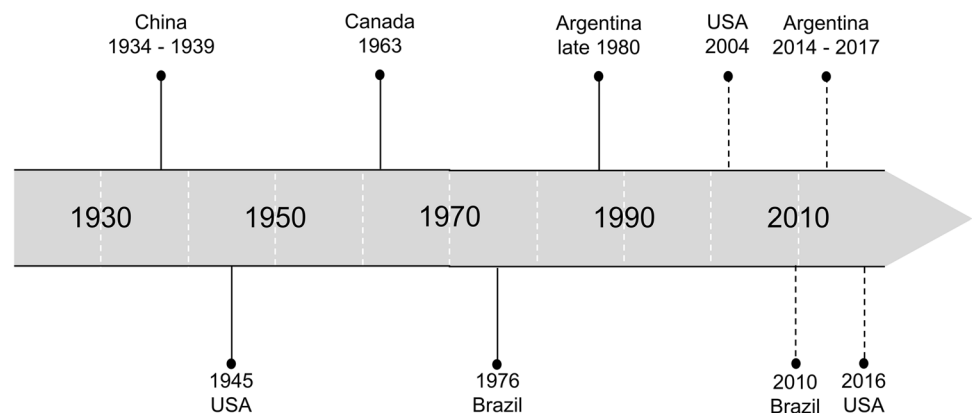
Target spot epidemics in the Americas

In general, a relatively extended period of time was necessary for *C. cassiicola* to become endemic in the main soybean-producing countries in South America. Since first reported from the southern US, approximately 60 years have passed before it emerged as a widespread threat across a larger geographic area to soybean production throughout the southern US. In Brazil, this period required from initial observation to endemic spread of the disease throughout the soybean-producing areas took approximately 30 years and in Argentina approximately 20 years. However, the specifics as related to epidemics report on each continent have been rather different and bare a separate presentation.

North America

The first report of target spot on soybean from the USA was in 1945 (Fig. 1) (Hartwig, 1959). In the southeastern US, target spot was also reported to occur on additional hosts including cotton (*Gossypium hirsutum* L.), cowpea, and sesame (*Sesame indicum* L.) (Jones 1961). Due to the initial misclassification of the causal organism as

Fig. 1 First reports of soybean target spot (solid black lines) and reemergence reports (dashed black lines)

China 1934 - 1939 | Canada 1963 | Argentina late 1980 | USA 2004 | Argentina 2014 - 2017

1930 1950 1970 1990 2010

1945 USA | 1976 Brazil | 2010 Brazil | 2016 USA

Helminthosporium gossypii, Jones (1961) proposed that the pathogen may have occurred in cotton in the USA at least 25 years before the first published report. Thus, it is suggested that the appearance of target spot on cotton was concomitant with the occurrence in soybean. Some years later, following the initial reports of target spot from several states in the USA, the first report of target spot in Canada was made in 1963. However, as opposed to the observation of target spot on foliage as was reported from the USA, the causal organism was identified from roots of mature soybean plants grown at three locations in Ontario (Seaman et al. 1965).

In the 1950s, Hartwig (1959) observed target spot causing significant yield losses in soybean in the Mississippi Delta ranging from 18 to 32%. Significant yield losses were reported in years that the accumulated rainfall was greater than normal. Hartwig's respect of the yield losses that could result from severe target spot epidemics led him to include resistance traits in the lines that he developed in the USDA soybean breeding program (M. Kenty, pers. comm.). As a result, the bulk of his breeding lines were released with resistance to target spot; however, they typically did not provide good agronomic traits and as a result were generally considered to be low yield-producing cultivars (L. Heatherly, pers. comm.). Even though Hartwig focused his efforts on breeding soybean lines with resistance to target spot, historically, soybean improvement programs have likely bred soybean lines without a focus on this specific disease. Breeding lines without a focus on target spot as an important plant disease could explain the high susceptibility to the disease within the currently available commercial germplasm. In addition, one situation that contributes to a dilution of traits within breeding programs, by apparently losing track of important plant diseases, has been the increasing rate of turnover within commercially available germplasm as a result of the need for inserting newly developed herbicide tolerance traits (e.g., Enlist and Xtend) to stay ahead of the increasing concerns that have resulted from important weed pests developing resistance to commonly used herbicides. Major shifts in herbicide trait tolerance genes (e.g., the Xtend trait associated with Dicamba tolerance) may have acted as driver of the high soybean cultivar turnover rate and consequently the continued resurgence of target spot in the USA.

As a brief history to the continued situation throughout the southeastern US, in 2004, a limited survey of soybean pathologists from the southern US estimated yield losses between 20 and 40% resulted from *C. cassiicola* in several commercial fields in Alabama and North and South Carolina (Koenning et al. 2006). More recently, during 2016, the environmental conditions were extremely conducive for the development of target spot across areas of the southern Mississippi River valley, which allowed the detection of

extremely susceptible cultivars. Even one of the most popular and best yield producing cultivars for the region (Asgrow 4632) proved to exhibit severe levels of lower canopy defoliation as a result of target spot. The problem ranged from the south Delta of Mississippi, in western Mississippi, to northeast Arkansas and extended north and east into northeast Mississippi (Allen 2017). The widespread nature of target spot throughout the mid-southern US in 2016 resulted in severe lower canopy defoliation on a substantial number of hectares within the region. Even though yield losses were presumed to result from the defoliation, judging the approximate yield losses that resulted from the epidemic were difficult since additional soybean diseases occurred across much of the geographical area. Diseases such as pod and stem blight (caused by *Diaporthe sojae* Lehman) were commonly observed in fields impacted with target spot since the two diseases prefer a similar set of environmental conditions. In many cases, and where target spot was severe, pod and stem blight may have gone undiagnosed, and therefore losses may have resulted from the co-infection of the two diseases. Many substantial yield loss situations, on the order of 2,017.5 kg/ha, were blamed solely on target spot, and therefore the concern regarding a lower canopy disease was likely greatly exaggerated across the region and lead to some significant expenditures at attempting to manage target spot since 2016 on an almost annual basis even though significant losses have not generally been attributed to target spot. In addition to the widespread outbreak of severe target spot in the mid-southern US, since the early 2000s, there has been a general increase in the frequency of target spot reports on additional hosts, such as cotton which continues to be grown on a substantial number of hectares in the mid-southern US. Pathogen occurrence since 2000 on cotton has notably been reported from Georgia (Fulmer et al., 2012), Alabama (Conner et al. 2013), Arkansas (Mehl et al. 2020), Florida (Sumabat et al. 2018b), Louisiana (Price et al. 2015), Mississippi (Schultz et al. 2017), North Carolina (Edmisten 2012), Tennessee (Butler et al. 2016), as well as Virginia (Mehl et al. 2020), indicating its endemic nature as well as general geographic distribution across the southern US.

South America

The first observations of target spot throughout South America followed a general progression across the continent. Even though target spot was first officially reported in Brazil on soybean in the São Paulo state in 1976 (Almeida et al. 1976), Yorinori et al. (1977) stated that the disease had initially been observed in the state of Mato Grosso in 1974. In the Rio Grande do Sul state, target spot was identified in 1978 by Veiga (1978) in experimental plots at the Federal University of Santa Maria. In 1986, root rot symptoms and leaf spots were observed in Castro county, Paraná state, and in

the following year, the disease was detected in Mato Grosso, Mato Grosso do Sul, and Rio Grande do Sul states (Yorinori, 1988). In 1995, the disease was reported on cotton in Mato Grosso state and in 2012 in Mato Grosso do Sul state (Goulart and Lams, 2016).

Whereas target spot has been a reported disease of soybean in Brazil since the 1970s, target spot has been considered a disease of limited importance since its first report in 1976 (Godoy et al. 2016). However, due to the adoption of susceptible cultivars and the loss of sensitivity by the pathogen to some of the major fungicide active ingredients currently available, the disease increased in importance between 2010 and 2020. Currently, target spot is considered an important disease and responsible for yield losses especially when susceptible cultivars are used. In 2006, an important target spot epidemic was reported with yield reduction estimated to be between 10 and 20% (Silva et al. 2008a, b). A meta-analysis based on data obtained from fungicide field trials across Brazil estimated the potential yield losses to be approximately 24%. However, yield losses were significantly moderated by cultivar, since some, e.g., BMX Potência RR, resulted to be highly tolerant to the disease, with potential maximum yield losses of 8%. Conversely, the potential yield losses on susceptible cultivars, e.g., M9144RR, were estimated at 42% (Edwards Molina et al. 2019b). Since 2010, target spot has been considered to be an endemic disease-causing yield loss not only in soybean fields but also in cotton production in the central and northeastern regions of Brazil (Galbieri et al. 2014). The disease has been reported in early stages, more specifically before and during flowering stage (R1) of soybean or cotton crops in the last years when sowing cotton in succession to soybean in the Cerrado region.

In Argentina, target spot was first detected in the northern region in the late 1980s (Ploper and Ramallo 1988). The prevalence as well as severity of target spot increased during the late 1990s. The disease was mainly observed by the beginning pod stage (R3) when pod formation is initiated. As a result of the observations of target spot at R3, this initially included the grouping of target spot with those diseases that impact soybean as a broad category of "late-season" diseases (Ploper et al. 2011). The grouping of plant diseases as a late-season disease is an artificial classification for diseases that can potentially cause premature senescence and/or reduce grain yield and quality and occur during late reproductive growth stages. However, following that general distinction, some plant pathologists have considered target spot in the group of diseases occurring in the early stage of the crop cycle, as initial symptoms can be detected as early as the initial flowering stages (R1). At the R1 stage, canopy closure has been observed to increase periods of high humidity favoring disease occurrence (Teramoto et al. 2013). However, the general period of canopy

closure typically depends on the prevailing row spacing used in a given region. Moreover, the presence of a lower canopy disease would depend on the environment encountered prior to disease observations.

In Argentina, target spot of soybean was first detected in the northwestern subtropical region. Beginning in 2000, target spot prevalence and severity increased throughout this geographical area, probably due to successive years of continuous soybean mono-cropping and a general increase in no-till cultivation practices (Ploper et al. 2011). Between 2014 and 2017, target spot was considered the most prevalent soybean disease in northwestern Argentina with mean severities ranging from 20 to 30% (Reznikov et al. 2019). More than 80% of Argentina's total soybean production is concentrated in the Pampas region, a temperate area, with cold winter temperatures located at the center-east of the country. Since 2015, observations of target spot have expanded from the subtropical region to the main soybean-growing area (Distéfano et al. 2017).

In Bolivia, approximately 95% of soybean production occurs in the Santa Cruz de la Sierra state (Wrather et al. 2001). Based on climate and soil types, soybean-producing areas in Bolivia could be divided into two different regions: Northern and Southern Santa Cruz. Since 2001, target spot has been reported to be an endemic disease mainly in the northern region. In situations where intensive rainfall occurs, the disease may appear during the flowering stages (R1 to R2) in this region, which is considered to be early compared to other regions where target spot may occur more regularly at mid-reproductive growth stages. In situations when target spot is observed during early, before flowering stage (R1), the disease can reach high levels of severity, causing significant reductions in yield due to premature defoliation (D Baldelomar, pers. comm.). In Southeastern Santa Cruz, target spot is more often observed during the grain filling stages (R5), due to environmental conditions that generally coincide with those growth stages. Presentation of symptoms during more advanced growth stages is probably the result of soybean being grown during the summer months in the southeastern region. Since the summer of 2005 and 2006, the high prevalence of *C. cassiicola* in combination with soybean rust have guided the fungicide application decisions to avoid yield and economic losses (D Baldelomar, pers. comm.).

In Paraguay, *C. cassiicola* was reported in soybean-producing areas in the Corpus Christi state (Xavier et al. 2013), and yield losses were estimated to be up to 405% (Arrúa et al. 2021). The occurrence of target spot in soybean has been reported in other countries in Latin America, such as Mexico (Onesirosan et al. 1974) as well as Colombia (López-Cardona et al., 2020). In Colombia, disease incidence within a limited geography, reported to be a commercial soybean production field with a specific cultivar (Soyica

P-34), was reported to be 50% (López-Cardona et al., 2020). Experiments conducted in Colombia in 2018 evaluated the effect of natural infection on four local soybean cultivars. Disease severities were observed between 1.4% and 52% leading to yield losses between 0 and 18%, which indicates that the cultivars have different levels of tolerance to target spot (Flórez-Gómez et al. 2021).

Epidemiology of target spot of soybean

Much like many of the organisms that cause foliar diseases in soybean, the overwintering strategy for *C. cassiicola* is an important method for survival between seasons. In general, *C. cassiicola* has been reported to overwinter on infected soybean debris, weeds, and seed which can all serve as a source of primary inoculum for the subsequent growing season (Fig. 2). The fungus can colonize residues of a wide range of plant tissue on the soil surface and has previously been isolated from soybean residue at least 6 months following harvest (Almeida et al. 2001). Survival on residues other than soybean allows the fungus a greater level of adaptability to surviving between seasons and infecting soybean in subsequent seasons. In addition, in some plant systems, *C. cassiicola* has been observed to have the ability to produce survival structures such as chlamydospores which can allow survival for more extended periods of time in the soil

profile. Chlamydospores, which are thickened hyphae that may be melanized and can aid in long-term survival, have been reported to aid in survival for extended periods of time in the soil profile. To date, chlamydospores have not been observed to be produced by isolates of *C. cassiicola* originating from soybean (Oliveira et al. 2012). However, the initial report of target spot as a disease of soybean from the 1940s made by Olive et al. (1945) does in fact mention the production of chlamydospores from agar culture that is in fact carried through the literature cited in the Compendium of Soybean Diseases as late as the Fifth Edition, published in 2015. With this discrepancy in mind, the information as it relates to this part of the life cycle has been omitted since in vitro as well as in vivo research has not widely indicated this to be an important aspect of the fungal life cycle in soybean; however, given the more widespread interest in target spot as a disease of soybean, it is likely a matter of time before this important mycological statement changes. Conversely, chlamydospores have been reported to occur in isolates recovered from cowpea, eggplant, cucumber, and lantana (*Lantana camara* L.), wild crapemyrtle or Barbados cherry (*Malpighia glabra* L.), and ash colored fleabane (*Vernonia cinerea* L.) (Oliveira et al. 2012; MacKenzie et al. 2018). Even without the production of chlamydospores, *C. cassiicola* has been reported to survive in fallow soil for more than 2 years (Sinclair et al. 1999). Alternatively, the pathogen has also been isolated from cysts of the soybean
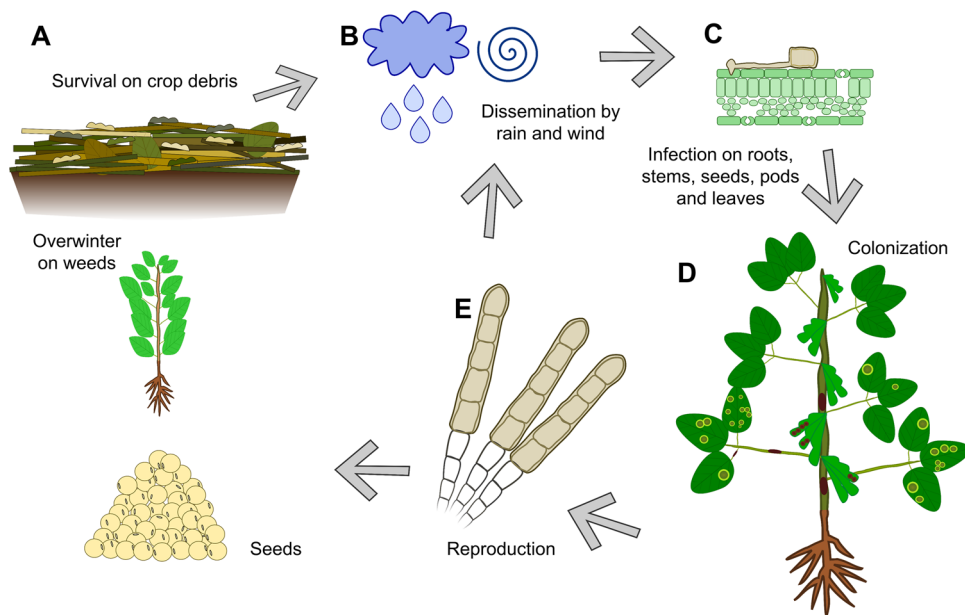

Fig. 2 Macrocyclic disease cycle of *Corynespora cassiicola*, the causal agent of target spot in soybean. The pathogen forms survival structures (A) and overwinters in crop debris, seed, or additional host plants that in some cases can include important weeds. Conidia are disseminated by rain or wind (B). Conidia germination is stimulated by high humidity conditions. An appressorium is formed in the end

of the germination tube during penetration. Infections can occur on leaves, roots, stems, pods, and seeds (C). The pathogen is necrotrophic and may release cassicolin, a non-host specific toxin, responsible for the production of the chlorotic halo (D). Under conducive environmental conditions, the pathogen forms conidiophores and releases conidia responsible for the secondary disease cycle (E)

cyst nematode (Carris et al. 1986). The fungal colony, conidiophore, conidia morphology, and typical symptomatic plants confirmed that the strain isolated from the nematode cysts was able to infect soybean (Carris et al. 1986).

Soybean seed has long been considered a reservoir of fungal inoculum to initiate target spot. Molecular research has confirmed the presence and quantification of *C. cassiicola* using multiplex qPCR assay (Ciampi-Guillardi et al. 2020). The associated molecular assay is highly sensitive and has been reported to detect one infested seed out of 1,599 healthy ones or an overall incidence level of 0.06%. The same study reported that 17.3% soybean seed sampled in the Brazilian market between 2013 and 2015 contained propagules of *C. cassicola* (Ciampi-Guillardi et al. 2020).

In situations where residue may not be the primary source of inoculum, fungal conidia can be transported distances from the primary source and infect soybean in either adjacent fields or fields at a greater distance. Wind is responsible for the release and dispersion of propagules under dry weather conditions. The so-called dry spores (Maude 1996) are passively liberated into the air within or above the canopy and can be disseminated long distances. Rainfall is usually responsible for the pathogen spread over relatively short distances within the same field, and most often mainly results in secondary infections. The distance of splash dissemination is usually dependent on wind velocity (Maude 1996). Infection of foliage is favored when free moisture is present on leaves, typically in the form of dew or when extended periods of rainfall maintain extended periods of leaf wetness, and the relative humidity within the soybean canopy is greater than 80% (Sinclair 1999). In vitro experiments reported that *C. cassiicola* conidia germinated within a temperature range of 7 to 39 °C, with an optimal temperature at 23 °C (Melo and Reis 2010; Sinclair 1999). Conidia germination rates of 16%, 33%, and 90% were observed following 12, 24, or 48 h of leaf wetness, respectively (Mesquini et al. 2011). Under the conducive environmental conditions, the conidia germinate and an appressorium is formed at the end of the germ tube, allowing the fungus to penetrate directly into host tissue (Mesquini et al. 2011). A series of fungus-specific enzymes help the fungus gain entry into the host plant tissue rather than entry through natural openings on the leaf surface. Oliveira et al. (2012) identified the production of eight enzymes by *C. cassiicola* isolates, which may be related to the pathogenic process. Some of the detected enzymes, such as α-glucosidase, β-glucosidase, and α-mannosidase, are important for cell wall degradation (Oliveira et al. 2012). In general, dry weather inhibits infection and colonization in the leaves. The temperature range conducive for mycelial growth was normally between 7.2 and 32.5 °C (Mesquini et al. 2011), similar to the temperature range which stimulates germination. An epidemiological study on leaf infection indicated that disease can develop

under temperatures between 18.0 and 32.7 °C, the optimal temperature was 26.7 °C (Mesquini et al., 2011), and symptoms could be visualized 5 to 7 days after infection (Agrios 1988).

One important aspect to note regarding target spot as a soybean disease, there are both leaf and petiole phases of the disease that differ in symptom expression from stem and root infection. However, reports of stem and root infection have tended to be rare as indicated by a limited number of reports in the literature. In instances where infection of either the stem or the root occurs, the infection has been reported to occur at the cotyledonary stage (Raffel et al. 1999). Soil temperatures between 15 and 18 °C are optimal for root infection and subsequent disease development on the roots and stem. Reports of the root rot associated with *C. corynespora* were most commonly reported from areas where no-till is a more common production practice. The symptoms associated with root infection were observed to include a dry rot, which began as a red to purple spot on the cortical tissue, evolving into a black color. The occurrence of leaf and root symptoms has been reported to be independent, as they have not frequently been observed simultaneously in the same field (Yorinori 1992). As a result of the two different disease types, leaf lesions and root lesions, research has been conducted to determine the specific types of isolates associated with the two discriminate symptom types. Snow and Berggren (1989) reported the existence of at least two different races of *C. cassiicola* affecting soybean. Based on their research, one race was detected that infected the hypocotyl, roots, and stem and was responsible for root rot, while the second race infected leaves, pods, and seed causing the symptoms that are more synonymous with the name target spot. In general, morphological differences between isolates that cause root and stem rots from those that cause target spot on soybean plant foliage could indicate that two separate pathogen species may be responsible for these two distinct disease symptoms (Sinclair and Shurtleff 1975). However, Yorinori (1992) stated that isolates from soybean roots were capable of causing lesions in leaf tissue that were regularly associated with target spot under artificial inoculations.

In addition, a specific area associated with the production of target spot symptoms on soybean plant material has to do with the production of a toxin that may be important in causing disease. Cassiicolin, a toxin that kills tissues adjacent to the infection site, has been reported to be released by the pathogen under in vitro conditions, when cultivated in Czapek medium, and in vivo conditions (Barthe et al. 2007). The toxin can diffuse through the plant tissue and include necrosis, similar to cell wall degrading enzymes which cause plasmolysis (Barthe et al. 2007). During in vitro testing, several plant species, including soybean, exhibit symptoms similar to the lesions associated with target spot when leaves

were either injected with the purified toxin or inoculated with conidia (Barthe et al. 2007). Additionally, cassiicolin behaves as a host-specific toxin sharing the same host range from which the strain originated (Barthe et al. 2007).

Symptoms caused by *C. cassiicola* on soybean leaves include roughly circular to irregular necrotic lesions, which may have alternating light and dark rings surrounded by a dull green or yellowish-green halo (Snow and Berggren 1989). In reference to these symptoms, the disease was commonly designated "target spot" (Fig. 3). Target spot is a typical representative of the "light stealer" disease group (Boote et al. 1983) since production of lesions can result in a reduction of the photosynthetic leaf area by the production of symptoms themselves in addition to leaf senescence that results from the disease and occurs prior to soybean plants reaching physiological maturity (R8). The damage on the photosynthetic leaf area is associated with a decrease in the plant's concentration of chlorophyll a and b (Fortunato et al. 2018). The capacity of electron transport is reduced as the thylakoid structural integrity is damaged by the pathogen,
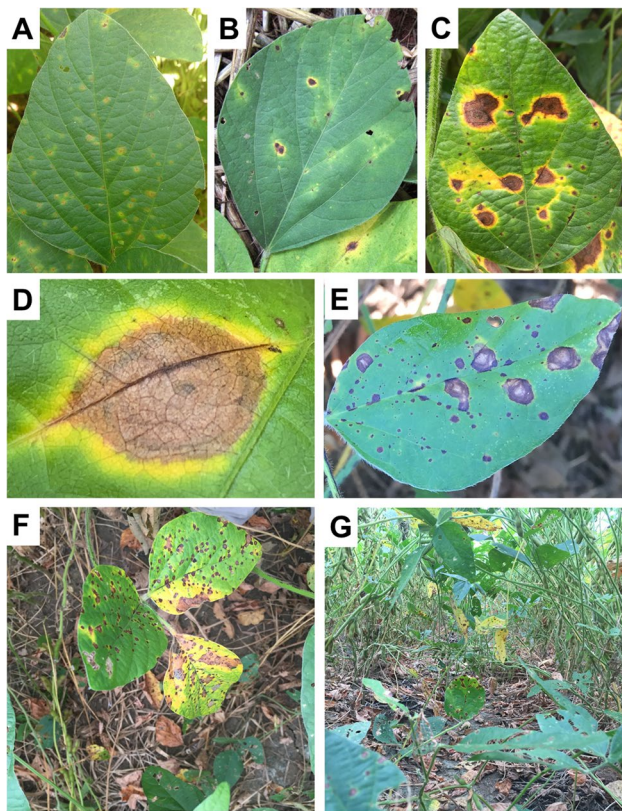
probably by a general release of the toxin produced by the fungus into the surrounding plant tissues (Fortunato et al. 2018). One key factor about target spot epidemics is that symptoms are most commonly observed in the lower part of the canopy. However, in situations where extended periods of conducive environmental conditions prevail, target spot symptoms can move up into the upper canopy. In situations in which target spot is observed in the upper canopy, yield losses can prove to be significant (JP Edwards Molina unpublished data). Target spot lesions can also be observed on pods, petioles, and stems similar to some of the additional foliar diseases that impact soybean production systems. As one example, the lesions on other plant parts can be mistaken for frogeye leaf spot, caused by the ascomycete *Cercospora sojina*. However, in situations where lesions are produced on plant parts other than leaf tissue, some laboratory diagnosis may be necessary to distinguish between diseases. Even though the symptoms associated with target spot at the field level are fairly well-known, there can be some subtle differences between symptom expression in the field and the symptoms that present following inoculation with fungal material in the greenhouse. In general, three different leaf lesion morphologies are observed as a result of *C. cassiicola* inoculations in greenhouse settings when conidia are used for inoculations: a dark infection point surrounded by a chlorotic halo, a necrotic spot without a chlorotic halo, and brown-reddish specks restricted to the infection point.

Host range

In general, *C. cassiicola* has been reported to have an extremely broad host range (Dixon et al. 2009). In fact, Dixon et al. (2009) reported that 530 plant species from 380 genera, including monocots, and dicots, can be infected by *C. cassiicola*. Additionally, *C. cassiicola* has been reported to grow as an endophyte or non-specific saprophyte on some hosts (Gond et al. 2007; Promputtha et al. 2007). Several authors have performed cross-inoculation assays with *C. cassiicola* and different host species in order to verify host specificity on species of economic importance. Several recent sources in the literature regarding soybean target spot contain extensive lists including host species for which *C. cassiicola* has not even been reported to be cross-pathogenic with soybean; therefore, the following information presented is based on compatible soybean hosts.

Olive et al. (1945) reported positive cross infections between *C. cassiicola* (named *Helminthosporium vignae* Olive, at that time) strains obtained from soybean and cowpea collected from different areas of the southern US. Isolates from soybean, sesame, cowpea, and cotton in Mississippi did not show differences in pathogenicity on different hosts (Jones 1961). However, Spencer and Walters

Fig. 3 Target spot foliar symptoms in soybean under field conditions. Typical leaf spot with black center (A), infection site surrounded by chlorotic halo (B), concentric rings of necrotic tissue and chlorotic halo (C), close-up image from a typical lesion surrounded by chlorotic halo (D), most advanced and recognizable lesion type (E), and induced premature senescence (F), and defoliation in the lower canopy (G)

(1969) confirmed a cross host susceptibility between cotton and soybean. Onesirosan et al. (1974) reported that soybean isolates from the southern US and Mexico were highly virulent on soybean, sesame, eggplant, and cotton. In general, different isolates from a single host can show differences in virulence on additional hosts. For example, one out of two isolates of *C. cassiicola* obtained from tomato were able to infect soybean (Cutrim and Silva 2013). In additional pathogenicity experiments, 15 *C. cassiicola* isolates were obtained from several hosts and only three isolates from Japanese cucumber hybrids (*Cucumis sativus* L.) (Oliveira et al. 2006). In the same experiment, one isolate from pumpkin was able to infect the four tested cucumber hybrids, whereas only two hybrids were infected by the two isolates obtained from soybean (Oliveira et al. 2006).

Regarding soybean and cotton, two important crops in the USA and Brazil, cross-inoculations performed with three cotton isolates and two soybean isolates on six cotton cultivars and six soybean cultivars in fact showed that the two groups of host-specific isolates were virulent to all hosts (Galbieri et al. 2014). Additionally, molecular studies identified similar banding patterns for cotton and soybean isolates using the ERIC/REP-PCR method (Galbieri et al. 2014). A genotypic variation within the cotton and soybean isolates was identified; however, there was little variation between isolates from different hosts. Similarly, results from ERIC/ REP-PCR and rDNA molecular techniques indicated that *C. cassiicola* isolates infecting cotton and soybean in Brazil are not genotypically distinct, which belong to the same strain of the pathogen (Galbieri et al. 2014).

A summary of five cross-inoculation studies on *C. cassiicola* and hosts showed that the greatest compatibility with soybean (100% of crossed infections) was observed with isolates that originated from cotton, eggplant, and sesame. Reduced compatibility levels, from 0 to 50%, were recorded for isolates that originated from papaya, tomato, cowpea, and cucumber (Table 1). One important thing to consider is that soybean cultivation near or in sequence with cotton may represent a high risk for target spot epidemics to both crops. The latter is a common context in the Mato Grosso state in Brazil and to a lesser extent in the mid-southern US, where the greatest target spot epidemics have resulted in reports of substantial yield losses in cotton and soybean in the recent past (Bowen et al. 2018; Galbieri et al. 2014).

Molecular and virulence studies demonstrated some host specialization within members from the genus *Corynespora* (Dixon et al. 2009; Sumabat et al. 2018a, 2018b). Phylogenetic analyses were performed using the nucleotide sequences of four loci with 143 isolates of *Corynespora* spp., and a lack of recombination within the species was observed (Dixon et al. 2009). Six phylogenetic lineages among the tested isolates correlated with host of origin, pathogenicity, and growth rate, but there was no correlation with the geographic origin of the isolates included in the study (Dixon et al. 2009). A second, more recent phylogenetic analysis with 23 isolates of *C. cassiicola* from several hosts was clustered according to the host of origin regardless of the geographic location (Sumabat et al. 2018b). Pathogenicity and virulence tests provided evidence of host specialization as isolates that were recovered from a specific host were more aggressive to the hosts they originated from (Sumabat et al. 2018b). Conversely, genome sequences and single polymorphism analyses of isolates collected from the southeastern US have shown a limited amount of genetic diversity within cotton isolates and little differentiation from the soybean isolate (Shrestha et al. 2017). Another study with 265 isolates from cotton, soybean, and tomato demonstrated that the population originated from soybean had a regional geographic structure (Sumabat et al. 2018a). In general, a high level of genotypic diversity detected within *C. cassiicola* populations leads to speculation of a cryptic sexual reproduction (Sumabat et al. 2018a).

Table 1 Cross-inoculation studies for *Corynespora cassiicola* isolates obtained from soybean, cotton, cowpea, cucumber, eggplant, papaya, sesame, or tomato with the references included for each of the respective research studies

	Inoculation direction[a]	Cotton	Cowpea	Cucumber	Eggplant	Papaya	Sesame	Tomato
Soybean	→	13/13	5/5	9/21	7/7	8/9	8/8	10/11
		(2–5,10,11)	(1,3,5,9)	(5,7,8,9)	(5)	(5,8)	(3,5)	(5,7,9)
	←	6/6	2/8	7/14	1/1	0/10	1/1	4/20
		(2–5,10,11)	(1,2,9)	(5,8,9,10)	(5)	(5,9)	(5)	(5,6,8,9,10)
	Host compatibility	100%	54%	46%	100%	42%	100%	45%

[a]The inoculation direction refers to instances where the right arrow (→) indicates studies where *Corynespora cassiicola* strains isolated from soybean were inoculated on the alternative hosts and leaf arrow (←) indicated where *Corynespora cassiicola* strains isolated from alternative hosts were inoculated on soybean

References: [1]Olive et al. 1945; [2]Jones, 1961; [3]Seaman et al., 1965; [4]Spencer and Walters, 1969; [5]Onesirosan et al., 1974; [6]Cutrim and Silva, 2003; [7]Oliveira et al., 2006; [8]Oliveira et al., 2007; [9]Dixon et al., 2009; [10]Teramoto et al., 2013; [11]Galbieri et al., 2014

Disease management

The effect of target spot on soybean yield can vary greatly from region to region or annually within the same region. The convergence of at least three factors, in situations where target spot may result in yield losses, comprises a group of "risk factors" which appear to be important and necessary for yield losses to result from target spot infection (Fig. 4). The risk factors required for yield losses to occur include (i) cultivars with low tolerance or a general high susceptibility to the disease, (ii) intense periods of rainfall during important soybean reproductive stages (Sinclair 1982; Teramoto et al. 2013), and (iii) significant levels of viable inoculum in the field that results from several years of soybean production and poor residue management practices (e.g., no till) that lead to infection of the soybean crop and the general production of lesions and subsequent premature defoliation in the mid-to-upper canopy (JP Edwards Molina, unpublished data) (Fig. 4). Even though target spot appears to be a common disease in some soybean-producing regions, disease management may not always be an important issue especially given that an ultra-specific environment appears to be required for the disease to manifest itself into yield-limiting situations. However, in situations where target spot may be yield-limiting, a fungicide application may be necessary to prevent the disease from defoliating much of the mid-to-upper canopy and reducing the photosynthetic leaf area of the soybean canopy especially prior to full seed (R6). Moreover, any target spot management program should be based on an integrated disease management
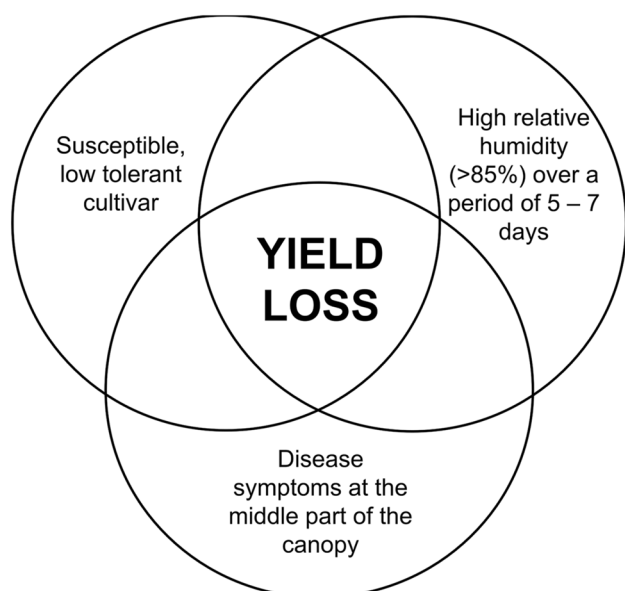
program. Several different methods can likely be employed to effectively manage target spot. One potential management practice that may be rather difficult given the history of soybean production practices in a given area would be based on the principle of evasion that consists of selecting the most adequate area with a reduced history of the disease (Agrios 2005). Additional important agricultural practices are based on crop rotation and selection of resistant cultivars. In situations where all three of the outlined risk factors are in convergence, soybean yield losses can be minimized by means of chemical control. However, additional factors should be considered when choosing specific fungicide materials for managing target spot as not all fungicides are efficacious on the organism and some work better than others on reducing defoliation in the mid-canopy.

Crop rotation

In general, soybean is cultivated in areas with a history of continuous soybean production; however, one way to manage the disease would be avoiding soybean cultivation in areas with historically high disease pressure. In terms of disease management that results from crop rotation, this practice consists of cultivating plant species that are not infected by a given organism. The principle of crop rotation aims to reduce the initial sources of inoculum and has been reported to be a satisfactory method when used to manage pathogens that can rely on survival in soil (Agrios 2005). In general, crop rotation can reduce pathogen populations in the soil and has been proven effective in situations whereby the organism in question causes a root or stem disease. As an example from a cotton production system, crop rotation to a non-host has been observed to reduce the number of microsclerotia which are the survival structures produced by *Verticillium dahliae*, which in some cotton-producing areas can result in substantial yield decreases (Wheeler et al. 2012). In the case of reniform nematodes on cotton, the cultivation of soybean after cotton allowed a greater reproduction rate of nematodes when compared to maize and peanut that are non-hosts (Holguin et al. 2015). The cultivation of camelina (*Camelina sativa*) and winter cereal rye (*Secale cereale*) as cover crops before soybean cultivation has been reported to reduce soilborne populations of *Pythium* (Acharya et al. 2020). Additionally, cereal rye has been reported to provide several benefits to soybean, by reducing soilborne populations of the soybean cyst nematode and suppression of *Rhizoctonia solani* and *Fusarium virguliforme* (Leandro et al. 2018; Wen et al. 2017). Crop rotation has shown to increase yield for soybean and maize. The relative abundance of *Fusarium* spp. was high in the system of continuous soybean production when compared to the system of rotation with maize. However, there were no differences in the relative abundance of leaf pathogens between continuous and

Fig. 4 Risk factors associated with soybean yield losses due to target spot

rotation systems of soybean and maize production (Chamberlain et al. 2021). In regard to foliar diseases, crop rotation has a positive effect on the disease severity of leaf pathogens on barley when compared to barley grown for 2 consecutive years (Kutcher et al. 2011) and oilseed rape in a 4-year rotation when compared to continuous oilseed rape cultivation (Kutcher et al. 2013).

Crop rotation is an important cultural method for the control of several soilborne pathogens of soybean and cotton. Nevertheless, the efficiency of crop rotation as a control practice for the management of *C. cassiicola* is a controversial topic. Almeida et al. (2001) suggested the adoption of monocot crops to reduce the primary inoculum sources of *C. cassiicola*; however, the control efficiency of such a management practice is reduced due to the non-specific saprophytic activity of the pathogen. The survey conducted by Dixon et al. (2009) indicated an extensive list of hosts for this pathogen. Over the last several decades, the increasing demand for commodities especially for soybean and the increased prices of cotton have resulted in a need for row crop farmers to continue to produce crops in a continuous fashion as a result of continued high commodity prices. However, the cultivation of soybean in fields following cotton has previously been demonstrated to be problematic since both are 100% compatible hosts for *C. cassiicola* infections (Teramoto et al. 2013; Galbieri et al. 2014). The extensive and successive cultivation of soybean and cotton has likely been a contributing factor at least in part to the resurgence of target spot in Brazil. However, there are likely additional contributing factors that are much more difficult to elucidate since no commercially resistant cultivars are available in either production system, and the complete impact of the environment on disease incidence and progression, in the case of the USA, as it relates to specific soybean growth stages is poorly understood. Moreover, documenting the differences in yield losses between fields that may have been in continuous soybean as compared to those that were recently rotated between crops is difficult to capture in a brief survey especially in a single year.

Breeding for resistance

One important pillar of integrated disease management for any specific disease is the selection of resistant cultivars. In general, the target spot resistance carried by soybean cultivars is quantitatively inherited, and the low heritability indicates that the effect of environment on the resistance phenotype is high (Soares and Arias 2020). Breeding for resistance to target spot began in earnest in the 1950s. Hartwig (1959) reported that soybean lines cultivated in the Mississippi Delta, in western Mississippi, were resistant to target spot, indicating the importance of resistance in the control of this disease. Additionally, the author reported that

soybean cultivars within maturity groups that reached physiological maturity in October or later than October were at greater risk to target spot than cultivars that reached physiological maturity at earlier calendar dates (Hartwig 1959). Since Hartwig's efforts in the 1950s and 1960s to observe the differences between soybean cultivars from a breeding standpoint, little effort has been continued. However, more recently, field experiments have considered the response of several different cultivars across multiple locations in Argentina and determined that disease pressure may vary according to location. During 2018 and 2019, research was conducted using the same genotypes planted in two different regions. Target spot severities between 25 and 40% were recorded in San Agustín, whereas disease severity in La Fragua ranged from 5 to 15%, both in northwest region of Argentina (Escobar et al. 2019).

The increase in target spot in Brazil may be related to the genetic improvement of new cultivars. One example, as stated by Silva et al. (2008a, b), suggested that since nematodes have become such a widespread issue that breeding for resistance to nematodes has been successful; however, those cultivars with enhanced resistance to the nematode *Heterodera glycines* may be more susceptible to *C. cassiicola*. Recent cultivar screenings to target spot resistance showed variability in the current commercial Brazilian germplasm, such as BRS360RR, which is a highly resistant genotype (JP Edwards Molina, unpublished data) or additional highly disease-tolerant cultivars. Histological studies on leaves of the resistant cultivar TMG 132 demonstrated an accumulation of phenolic-like compounds, which contributes to the death of fungal mycelia and to the maintenance of cell wall integrity. Thus, soybean resistance to target spot may be associated with the activation of the phenylpropanoid pathway (Fortunato et al. 2017).

Fungicides

The use of fungicides has previously been reported to be an effective practice for managing target spot. The bulk of the field trials conducted with fungicides to manage target spot have been conducted in South America with only a limited number of published trials that generally include data for a single year from a single location as well as unpublished trials conducted in the southern US. Under field conditions, experiments conducted during three seasons in northwestern Argentina demonstrated that under environmental conditions that were believed to be conducive for the development of high levels of target spot, all tested fungicides were able to reduce disease severity from 10 to 70% when compared to disease severity levels of the non-treated plots (Reznikov et al. 2019). Fungicide application timing was made at either the beginning pod (R3) or beginning seed (R5) timings with products that contained pre-mixes of multiple fungicide

modes of action. The specific fungicides were either composed of a quinone outside inhibitor (QoI) and a demethylation inhibitor (DMI) or QoI and a succinate dehydrogenase inhibitor (SDHI) as two-way modes of action or a three-way combination that included a QoI, DMI, and an SDHI. Target spot control was observed to differ between the 3 years of the study; however, in general, control ranged from 10 to 70% for the most efficacious fungicide mixtures as compared to the non-treated. A limited number of fungicide trials have been conducted in the mid-southern US to determine the most efficacious fungicide products on target spot. In general, fungicide applications for disease management have tended to be made as specifically timed applications based on growth stage, generally between the R3 and full pod (R4) stages.

Additional fungicide trials conducted in South America have produced similar results, with the best fungicide products tending to be composed of the mixture of a QoI and a SDHI. A meta-analysis was conducted with data from field trials whereby labeled fungicides were tested between 2012 and 2016 in Argentina (Edwards Molina et al. 2019a). The associated analysis separated fungicides into three groups according to their efficacy: the high efficacy group consisted of fluxapyroxad + pyraclostrobin and fluxapyroxad + pyraclostrobin + epoxiconazole ($\approx$76% of control efficiency), intermediate control efficacy with prothioconazole + trifloxystrobin (66%), and low efficacy with mancozeb (49.6%), azoxystrobin + bixafen (46.7%), and carbendazim (32.4%). Additionally, the study determined that yield responses were dependent on target spot pressure (assessed from the non-treated plots at the R5-R6 growth stage). When target spot severity (mean of the whole plant) was greater than 35%, yield responses to fungicides based on fluxapyroxad + pyraclostrobin had the best performances increasing yield relative to the non-treated check fluxapyroxad + pyraclostrobin + epoxiconazole 503 kg ha^{-1} (+ 20.2%) and fluxapyroxad + pyraclostrobin 469 kg ha^{-1} (+ 19.1%) (Edwards Molina et al. 2019a).

A limited number of published fungicide trials have been conducted in the southern US with target spot being the main disease of interest. Two foliar fungicide trials were conducted in eastern MS during 2016, a year when a fairly severe target spot epidemic occurred throughout the mid-southern US, to determine the impact of the application of multiple products on target spot (Brochard et al. 2017a, b). One of the main variables evaluated in the two fungicide trials was the impact of fungicide on lower canopy defoliation that resulted due to target spot; however, differences were not significant between fungicide products applied and the non-treated. In most cases, the lower canopy defoliation that was observed to have resulted due to target spot 42 days post-application was reduced by between 2.5 and 20.8% with fungicide products that contained multiple modes of action

typically consisting of a QoI and DMI mixture. Notably, azoxystrobin on its own resulted in numerically greater defoliation than the non-treated. However, target spot was not the only disease observed in these two trials, and as a result, it is likely that either frogeye leaf spot or Septoria brown spot (caused by *Septoria glycines* Hemmi) may have contributed to any potential yield losses as well as overall defoliation since the environment was conducive for additional diseases. Additional fungicide trials aimed at target spot management have been conducted in western MS as well as some additional areas across the southern US; however, data associated with those trials have not been published (TW Allen, unpublished data). Even though managing the fungus requires the proper fungicide application in the field, determining if the fungicide is efficacious is sometimes important in a laboratory setting. In vitro evaluation of six fungicides at two concentrations (50 and 100 ppm) revealed that four fungicides, i.e., fluxapyroxad, propiconazole, tebuconazole, and hexaconazole completely inhibited the growth of the pathogen (Kurre et al. 2017).

Research suggests that one additional area whereby managing the target spot fungus can be achieved is through the use of seed treatments. However, research studies evaluating fungicide efficacy to control *C. cassiicola* on seed are scarce. Moreover, the number of research studies that have determined the presence of *C. cassiicola* from soybean seed as well as the importance of this particular source of inoculum are even more limited. *C. cassiicola* has previously been reported as an organism isolated from seed (Ramiro et al. 2019); however, seed is oftentimes not considered a major source of inoculum for the initiation of target spot epidemics. A recommended treatment for soybean seed consists of a mixture of thiabendazole + thiram or carbendazim + thiram (Reis et al. 2010). However, low efficiency control of carbendazim has been reported (Xavier et al. 2013); therefore, the use of this active ingredient may not be effective as a seed treatment to control *C. cassiicola*. Research to elucidate the potential efficacy of seed treatment products and how the source of seed may serve as an important inoculum source has not been conducted in the USA.

Impact of fungicide resistance

As mentioned above, fungicides have become an important management consideration for soybean farmers, especially in areas where disease threatens yield on an almost constant basis. However, the increased reliance on fungicides comes with the omnipresent risk of resistance development, especially in situations where fungicides are used for improving yield in the absence of disease. Since the initial observation of soybean rust in the contiguous USA in late 2004 (Schneider et al. 2005), fungicide applications in the greater US soybean production system have increased whether as a disease

management practice or in situations where increased yield is perceived to occur as a result of "plant health" (Bandara et al. 2020; Wise and Mueller 2011). One important aspect of fungicide applications to consider would be the impact of plant-health-based fungicide applications, generally considered to be those made at a specific growth stage in the absence of any yield-limiting disease, which would be rather difficult to measure since fungicide sales are generally estimated and not based on actual purchase and/or application data, at least in the US production area. Moreover, since 2010, the increase in documented cases of fungicide resistance within the *Cercospora sojina* pathosystem, the causal organism of frogeye leaf spot, may provide some foresight into what could be expected within the broader soybean production system when it comes to foliar disease management using fungicides. Since 2010, when QoI resistance was initially documented in Tennessee (Zhang et al. 2012), QoI-resistant *C. sojina* has been documented from an additional 20 total states ranging from as far east as Virginia, to as far north and east as Ohio and as far north and west as South Dakota with the most northern observations from Michigan, Minnesota, and Wisconsin (Neves et al. 2020, 2021; Zhang et al. 2018). In at least one instance from a single state, the documented instance of the G143A substitution from the *C. sojina* population within Mississippi was observed to be widespread (Standish et al. 2015). The documentation continues throughout the US soybean production area where frogeye is observed. A similar correlation can be made between *C. sojina* and *Corynespora cassiicola*. Similar to *Cercospora sojina*, *Corynespora cassiicola* is classified as a high-risk pathogen when considering the potential development of fungicide resistance (FRAC, 2019). In the USA and Brazil, reports of the G143A substitution in *C. cassiicola*, which confer complete resistance to members of the QoI (Quinone outside Inhibitors) fungicide class, have occurred from a limited number of locations (Rondon and Lawrence 2019; 2021; FRAC 2020). Additional reports of QoI resistance within *C. cassiicola* have been made from as many as three additional states at regional meetings (TW Allen, personal communication); however, no official reports of fungicide resistance outside of Alabama have yet to be published. In addition, in vitro experiments detected reduced sensitivity of Brazilian isolates to carbendazim and benzimidazole, both from the group of methyl benzimidazole carbamates (Xavier et al. 2013; Avozani et al. 2014). In Brazil, isolates containing the sdhB-H278Y and sdhC-N75S substitutions that confer reduced sensitivity to the SDHI (succinate dehydrogenase inhibitors) fungicides were reported by FRAC in 2018 (FRAC 2020). One outcome of some of the documented regional fungicide resistance within the important soybean pathogen populations has apparently been an increased reliance on fungicide chemistries with a more multi-site mode of action. For example, mancozeb has gained a newfound importance in South America as a result of increased target spot (Godoy et al. 2015).

Future direction

The importance of target spot has increased as a potentially yield-limiting disease throughout the Americas over the past decade. In addition, there is a perception that target spot has become a more important and widespread disease in situations where crop rotation includes cotton as can most often occur throughout the southeastern US as well as parts of Argentina and Brazil. Additionally, the use of susceptible cultivars increases the risk of yield losses that could occur as a result of target spot. Thus, experiments focusing on cultivar resistance as well as tolerance should be studied in areas where target spot is considered an important disease. The incorporation of parental lines with good resistance to target spot may be fundamental for the improvement of breeding programs focusing on the conditions of each region specifically. Efforts on research should be focused on the determination of conducive weather conditions, mainly focused on accumulated rainfalls (Hartwig 1959), which has been indirectly reported as a risk factor (Sinclair 1982; Teramoto et al. 2013). A better understanding of disease epidemiology, such as genetic variability within the pathogen population and the potential occurrence of sexual reproduction, may help to improve disease management and predict future disease outbreaks. There is a need for conducting crop rotation experiments that, in the future, will provide a better series of disease management suggestions as they relate to crop rotation strategies. Determining the best fungicide application timing strategies as well as the specific application of fungicide mixtures that contain the most efficacious commercially available active ingredients should be taken into consideration in the management of foliar diseases of soybean, as target spot has been observed to occur earlier in the soybean plant phenology than some of the foliar diseases that are more inherently considered to be late season diseases. However, much of that research will need to be related to the prevailing environment and how that impacts disease incidence, severity, and overall disease progression during important soybean growth stages. Fungicide resistance monitoring studies using isolates of *C. corynespora* originating from soybean continue to be conducted in multiple states throughout the southern US where the disease has been observed to be most damaging (Rondon and Lawrence, 2019; 2021). Information regarding fungicide resistance will ultimately aid in the identification of loss in fungicide efficacy, which may contribute to a better management of target spot through the use of fungicide applications.

Declarations

Conflict of interest The authors declare no competing interests.

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