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Review Article

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Fungi Causing Leaf Spot Diseases of Soyabean: Their Epidemiology and Integrated Management Strategies

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Abstract

Majority of the leaf spot diseases on soybean crop is caused by different pathogenic fungi. Under favourable conditions and severe infection, it may result in heavy defoliation. It decreases the production efficiency of the crop by reducing the surface area available on leaves for photosynthesis. With less photosynthetic activity, the yield may be reduced significantly. Thus, making it a production constraint by reducing the economic viability of the infected soybean crop. Cercospora leaf spot, Target leaf spot, Alternaria leaf spot, Brown Spot, Phyllosticta leaf spot are the major leaf spots caused due to fungus in the soybean crop worldwide. In this review, we are discussing about economic importance, symptomatology, causal organism, disease cycle, epidemiology and integrated management of leaf spots caused due to fungal infection in the soybean crop.

Keywords: Epidemiology, fungi, integrated management, soyabean

1. Introduction

Among oilseeds, soybean covers a leading role at the global scale. As of now, soybeans represent about 35% of total harvested area committed to annual and perennial oilseed crops. The crop is highly valued and a profitable crop as well. The commercial utilization of both its sub products, meal and oil, respectively account for two thirds and one third of the crop's economic value thereby determining its economic feasibility. A specific feature of the soybean economy is that substantial value addition occurs at the downstream phases of the production and processing chain. At the worldwide level, the majority of soybeans delivered is put away and dispatched in mass to large scale industrial units for further processing into oil and meal. As one of the world's major and quickest extending crops, soybean contributes altogether to in general human sustenance regarding both calorie and protein consumption; the crop appears to be well placed to meet the fast-growing demand for vegetable oil and animal feed in developing countries.

Currently, India ranks fourth in respect to production of soybean in the world. The crop helps earn valuable foreign exchange (Rs. 62000 million in 2012-13) by way of soya meal exports. Soybean has largely been responsible in uplifting farmer's economic status in many pockets of the country. It usually fetches higher income to the farmers owing to the huge export market for soybean de-oiled cake.

Soybean crop is host to more than 100 pathogens. In India, loss due to various diseases has been estimated to an extent of 12% of total production. The major economically important diseases are rust, wilts, leaf spot, rots, powdery mildew, bacterial and viral diseases (Jahagirdar, 2019). Alternaria leaf spot at vegetative stage and Cercospora leaf spot at flowering and pod formation stage in the soybean fields of Assam were reported by Borah (2019).

2. Cercospora Leaf Spot

Frogeye leaf spot caused by *Cercospora sojina* is one of the most conventional and prominent soybean infections. As of now, it is the most damaging disease in Central Brazil. Frogeye leaf spot is a common fungal disease which is recorded every year with a severity on an average of 2.50 to 15.75% (Huilgol, 2020). For many decades, FLS has been prevalent in the southern U.S. and, more recently, has become endemic throughout the Midwest and upper Midwest. Significant yield losses of soybean (10–60%) from this disease have occurred and severity has increased in the last five years due to continued cropping of susceptible soybean varieties over large acreage. Since 1999, increased severity and prevalence of FLS have been reported in some southern and north central areas of the U. S. Outside the U. S., *C. sojina* has been reported in at least 27 countries spanning North and South America, Europe, Africa, and Asia (Lin and Kelly, 2018).

2.1. Symptoms

Leaf spots are circular to angular in shape. Leaf symptoms begin as dark brown, water-soaked spots and mature into lesions with tan or brown centers and a narrow reddish brown to purple margin. Older lesions are translucent and have whitish centers containing black dots. In severely infected plants, several lesions may coalesce into larger irregular shaped spots (Huilgol, 2020).

2.2. Causal organism

Kingdom: Fungi

Division: Ascomycota

Class: Dothideomycetes

Order: Capnodiales

Family: Mycosphaerellaceae

Genus: Cercospora

Species: *C. sojina*

Conidia are produced on light to dark brown conidiophores borne in fascicles arising from a thin stroma formed on infected leaves, stems or seeds, or infested crop residue. Conidia form on tips of conidiophores and are pushed aside as the conidiophores continue to grow. Conidia are hyaline, and cylindrical to fusiform, tapering toward the tip, and 6 to 8 µm by 40 to 70 µm in size with up to 10 septa. The size and shape of conidia and conidiophores vary with the substrate on which the fungus grows (Lin and Kelly, 2018).

2.3. Disease cycle

Frogeye leaf spot is a polycyclic disease in which infection, symptom development, and production of conidia are repeated throughout the growing season. The amount of disease is influenced by the primary or first inoculum (the amount of initial inoculum) and the number of disease cycles that occurs in the field. *C. sojina* overwinters as mycelium in infected seeds and infected soybean residue. Initial infection occurs as conidia produced on infected residues or cotyledons are dispersed by splashing rain or wind. Dew and rain events produce favorable environmental conditions to promote secondary infections as conidia are dispersed to other areas on the plant or are carried in rain and wind to surrounding fields. Warm (25-30°C) and wet weather (e.g., rain, heavy dew or >90% relative humidity) conditions favor infection and disease development. Following infection, lesions are visible after 7 to 14 days depending on the conditions. Leaves are most susceptible to infection as they are emerging and become less susceptible as they mature. Mild winter temperatures and conservation tillage favor pathogen survival. Diseased soybean residue (leaves, stems and pods) left on the soil surface provides inoculum to continue disease cycle in next soybean crop. If the first symptoms of this disease are detected late in the season (at or after growth stage R5/beginning seed) there is very little impact on the plant. However, if this cycle begins prior to or at flowering, then substantial amounts of disease can develop that will impact yield. Under conducive conditions, the fungus will continue to produce conidia and secondary infections, which increases disease severity (Lin and Kelly, 2018).

2.4. Epidemiology

Its wind and seed-borne nature offer great advantage over most soybean pathogens for quick dispersal. Once introduced, the fungus can survive on crop residues or on stored seed. For an outbreak to occur, the fungus requires high temperature (25-35°C) and humidity, and susceptible cultivars (Yorinori, 1989).

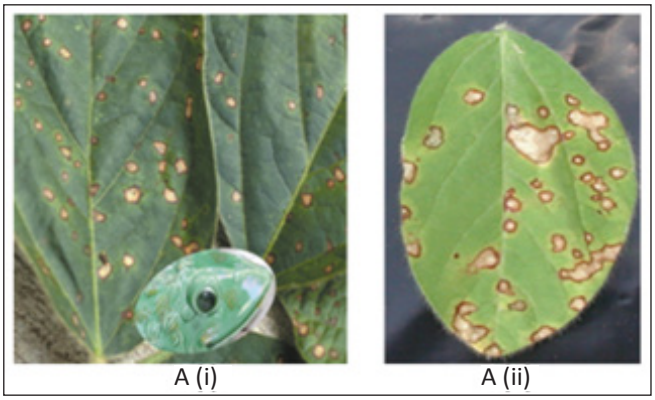
2.5. Integrated management

Crop rotation and tillage can be effective means of reducing the population from season to season. Two-year rotations with crops other than soybean may help to reduce the level of *C. sojina* inoculum. Host resistance is the most effective and economical management practice for frogeye leaf spot. Several different genes for frogeye leaf spot resistance have been discovered and described in soybean. Currently, Rcs1, Rcs2, and Rcs3 are three named genes by the Soybean Genetics Committee for frogeye leaf spot resistance, and additional sources of resistance have been reported (Lin and Kelly, 2018).

Mancozeb, a dithiocarbamate fungicide with contact activity, and two benzimidazole fungicides, benomyl and carbendazim, with systemic activity, were known to be effective against Frog eye leaf spot (FELS). Galloway (2008) reported that Flusilazole and the mixture of flusilazole+carbendazim were more effective against FELS than benomyl (Figure 1 and B).

3. Target Leaf Spot

The disease recorded for the first time in Karnataka by Huilgol, 2020 with maximum disease severity was noticed in Haveri district (39.05%) followed by Belagavi (28.02%) and Dharwad



A (i) A (ii)

Figure 1: (A) Characteristic frogeye leaf spot lesions resulting from Cercospora sojina infection of soybean leaves. i) The lesions start as dark water-soaked spots and develop into well-defined lesions with light centers and dark borders; ii) Several adjacent lesions may coalesce and form larger irregular spots (Mian et al., 2008)

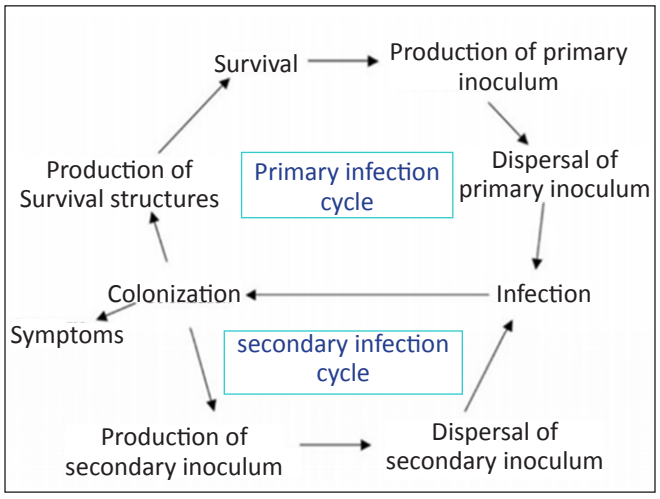


Figure 1 (B): Disease cycle of Cercospora sojina (Lin and Kelly, 2018)

district (19.18%). In India, Target leaf spot reduced yield but do not occur regularly or occur in small areas. Target leaf spot is one of the most damaging diseases of Soybean in Bolivia (Wrather et al., 2010).

3.1. Symptoms

On leaves spots are rounded to irregular and dark brown in colour and size varies from small specks to big mature spots. These spots are surrounded by a dull green or yellowish green halo. At later stages the leaves become yellow and drop prematurely. On stem and petiole, the spots are dark brown and spindle shaped (Huligol, 2020). Infected areas on stems and petioles were dark brown elongated and spindle shaped. Pod spots were mostly circular with slightly depressed and dark brown in colour (Kurre, 2016).

3.2. Causal organism

Target leaf spot caused by *Corynespora cassiicola*

Kingdom: Fungi

Phylum: Ascomycota

Subphylum: Pezizomycotina

Class: Dothideomycetes

Order: Pleosporales

Family: Corynesporascaceae

Genus: Corynespora

Conidiophores arising singly from mycelium, 3-7 septate, unbranched, erect, straight to slightly curved, sub-hyaline to pale-olivaceous, brown, smooth walled, 0-16 pseudosepta, hilum at the base (Kurre, 2016) (Figure 2).

3.3. Epidemiology

Temperature range of 25˚C-30˚C and RH of 80% was optimum for initiation and development of disease, with maximum severity at 100% RH and 25˚C (Sharma, 2005). Twelve-hour alternate light and dark period influenced the disease

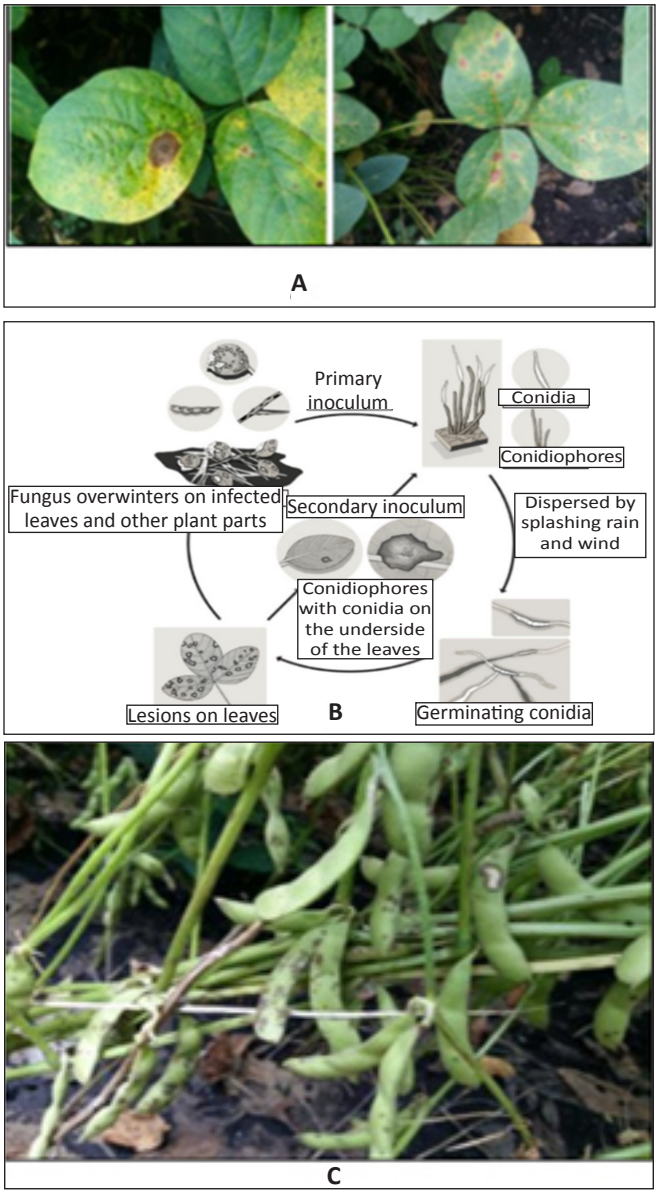


Figure 2: (A) Dark brown spots surrounded by a dull green or yellowish halo; (B) Life cycle of Corynespora cassiicola; (C) Symptoms on stems and petioles. The spots are dark brown and spindle shaped (Huligol, 2020)

development while complete light and dark induced less disease.

3.4. Integrated management

Late sowing could reduce the disease severity compared to early sowing and Hexaconazol was highly effective in reducing the disease (Kurre, 2016). Intercropping of soybean with maize resulted in less disease as compared to pure crop of soybean (Sharma, 2005). He also found biopesticide Wanis (0.5%) to be highly effective in inhibiting mycelial growth of the pathogen followed by Neemgold (0.5%) and Achook (0.5%). *Trichoderma harzianum* and *T. pseudokoningii* were highly antagonistic to the pathogen.

4. Alternaria Leaf Spot

The roving survey was conducted during Kharif season 2017–18 in Vidarbha region of Maharashtra in order to know the per cent disease incidence of Alternaria leaf spot of soybean and results revealed that maximum mean per cent disease incidence (41.66%) was observed in Walgoan village of Amravati district followed by in Mozari (40.19%) Tahsil Tiwsa district Amravati (Zade et al., 2018). Soyabean is widely cultivated oil seed crop in Marathwada and rest of Maharashtra region. The leaf spot of soyabean caused by *Alternaria alternata* is becoming a common disease on soyabean crop (Gawai and Mangnalikar, 2018). Disease incidences of up to 30% were recorded in Turkey (Ustun et al., 2019).

4.1. Symptoms

Infected plants displayed necrotic, circular to oval, and dark brown spots on the upper surfaces of the lower leaves (Ustun et al., 2019). Diseased lesions are round or restricted by a major vein or merge with another lesion. Some have brown concentric rings with a well-defined border. The lesions expand and may combine to yield larger dead areas on the leaves. Infected leaves eventually dry out and fall.

4.2. Causal organism

Alternaria alternata specifically lives in the soil as a saprophyte and decomposition organic matter.

Domain: Eukaryota

Kingdom: Fungi

Phylum: Ascomycota

Subphylum: Pezizomycotina

Class: Dothideomycetes

Subclass: Pleosporomycetidae

Order: Pleosporales

Family: Pleosporaceae

Genus: Alternaria

Species: *Alternaria alternata*

4.3. Disease cycle

Spores after dispersion by wind or water and on further germination develop into long chains that start producing spores from the tips of the hyphae, which is known as a conidiophore. The air borne conidia infects the plant, thereby producing host specific toxins leading to colonization and symptom development.

4.4. Epidemiology

Temperature 35˚C and soil pH 6.5 is optimum for the growth *Alternaria alternata* (Gawai and Mangnalikar, 2018). Dispersion of the conidiospores is by wind or by water, landing in a suitable environment such as plant parts like leaves, fruits, or seed. This enables the spore to start germinating when there is enough moisture and temperatures of 31-32°C (Mokobi, 2020).

4.5. Integrated management

Planting high quality, certified, disease-free seed that has been treated with a strobilurin fungicide is critical to managing this disease.

4.6. Future approaches in disease management

Dip treatment in different concentration of angiospermic plant extracts (10, 15 and 20%) *Allium sativum* and *Allium cepa* brought about significant reduction in diseases intensity caused by *Alternaria alternata* leaf spot on the soybean (Bhosale et al., 2014). Organic compounds in the angiospermic plant extracts like phenolics, quinones, flavones, flavonoids as well as aromatic oils and nutrients accelerate development and growth of the plants and helping plants against fungal attack. Significant improvement was observed in plant growth of soybean due to reduction in pathogenic fungal growth.

5. Brown Spot

Brown spot disease occurs at the end of the soybean (*Glycine max* (L.) Merrill) cycle in all cropping regions in Brazil. Under severe conditions, end-of-cycle leaf diseases can reduce crop yield by over 20% (Jung et al., 2002). Estimated yield loss in soyabean (thousand metric tonnes) in 2006 due to brown spot were 1176.5 (Argentina), 340 (Brazil), 0.8 (Canada), 2186.3 (China), 19.6 (India), 536.6 (USA) (Wrather et al., 2010).

5.1. Symptoms

The disease is most noticeable upon the foliage, where it is characterized by brown or reddish - brown angular spots 2 mm in diameter. It is primarily a leaf - spot disease and causes severe defoliation. It appears also on the stems and pods as the plants approach maturity. The first evidence of disease is apparent early in the season, when the first pair of true leaves have formed. At this time there are irregular dark - brown patches, varying in size from minute specks to areas 4 mm in diameter on the cotyledons. The lesions on first true leaves are conspicuously reddish brown on both leaf surfaces. They are angular in outline, being limited by the small veins, and vary in size from 1 to 5 mm in diameter. The tissues surrounding the diseased areas are pale green at first and then they become decidedly chlorotic, after which the leaves fall off. The disease progresses upward on the plants from the lower leaves. The disease on the stems manifests itself by the presence of indefinitely margined, brown discolorations. Tissues adjacent to these discolorations are less green than normal tissues and as chlorosis advances, the diseased areas become more conspicuous. The spots on pods are similar in all respects to those on stems. The smaller areas are pinpoint like, whereas the larger ones may come to involve more than half the surface of the pods (Wolf and Lehman, 1926).

5.2. Causal organism

Brown spot is an early season fungal disease caused by

Septoria glycines.

Kingdom: Fungi

Division: Ascomycota

Class: Dothideomycetes

Order: Capnodiales

Family: Mycosphaerellaceae

Genus: Septoria

Species: *S. glycine*

5.3. Disease cycle

The fungus survives on infected leaf and stem residue. Warm, wet weather favors disease development. Disease usually stops developing during hot, dry weather but may become active again near maturity or when conditions are more favorable.

5.4. Epidemiology

Plants as young as the V2 growth stage show symptoms. The fungus spreads from the soil to soybean plants by splashing rain. Frequent rainfall was the primary reason for the occurrence of this disease (Yang, 2002). Infection and disease development can occur over a range of temperatures (60 to 85°F), but is greatest at 77°F; increasing leaf wetness (up to 36 hours) is associated with increased disease infection and severity. Disease development is severely hindered during hot, dry weather, but will resume when conditions again favor the disease and/or the crop approaches maturity. Infections early in the season are frequently the source of late-season infections. As a consequence, the disease almost always exists first in the lower canopy and then moves into the upper canopy as the season progresses (UK Cooperative Extension Service, 2012).

5.5. Integrated management

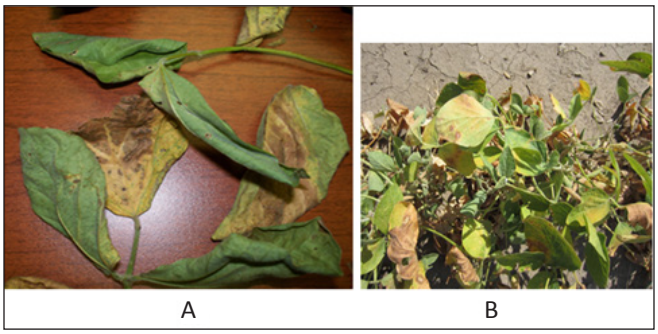
The best results for reducing disease incidence and severity were obtained with the strobilurin fungicides pyraclostrobin plus epoxiconazole, trifloxystrobin plus cyproconazole, and azoxystrobin. The best yields were obtained with chemical and biological treatments at high doses applied at R3; these treatments appeared to have a direct relationship with reduced development of foliar symptoms (Mantecon, 2008) (Figure 3, 4 and 5).

6. Phyllosticta Leaf Spot

The disease has caused severe defoliation in certain areas such as the Eastern Shore of Maryland and Southeastern Missouri. In general, the disease does not persist long after the third - or fourth - leaf stage of the soybean plant (Dunleavy et al., 1966).

6.1. Symptoms

Symptoms may look like those caused by Phytophthora root rot, but close examination reveals leaf scorch symptoms without root rot. New lesions are round or oval. Infected leaves often have lesions starting at the leaf margin and



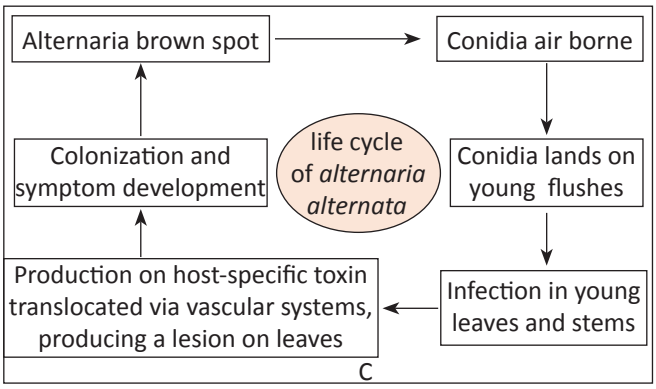
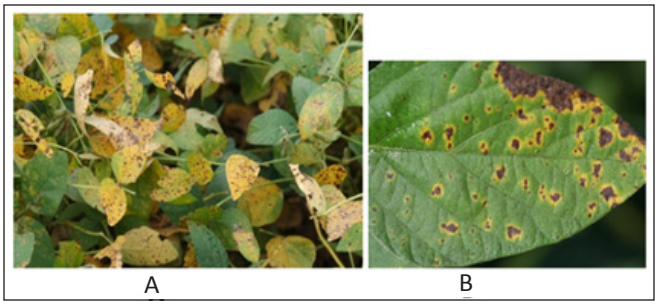


Figure 3: (A) Soybean leaves with symptoms; (B) Soybean plants with symptoms Image (Liu et al., 2012); (C) Life cycle of Alternaria alternata (Chung, 2012)



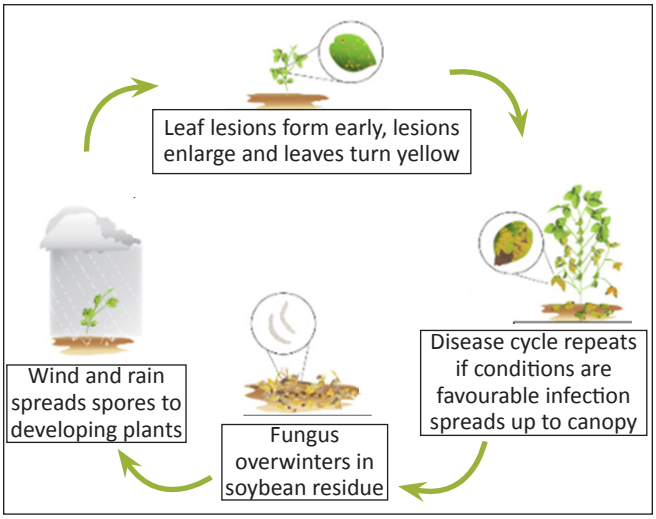


Figure 4: (A)-(B) Septoria brown spot lesion; (C) Disease cycle of *Septoria glycine*

A B

Figure 5: (A) Phyllosticta leaf spot lesion; (B) Pycnidia in older lesion of leaf

progressing inward. As the disease progresses, lesions have an irregular or V shape pointing inward. When severe infection occurs, diseased tissues extend through petioles to stems, killing the plants and causing stand reduction. Sometimes, canker type symptoms can be observed on lower portions of stems in severely infected plants. With cool and moist conditions, pods and seeds can be infected, causing seed discoloration (Yang, 2002).

6.2. Causal organism

Phyllosticta leaf spot is caused by the fungus *Phyllosticta sojicola*.

Kingdom: Fungi

Division: Ascomycota

Class: Dothideomycetes

Order: Botryosphaeriales

Family: Botryosphaeriaceae

Genus: Phyllosticta

Species: *Phyllosticta sojicola*

Conidia mainly aseptate, rather variable in size hyaline, sub globose to ellipsoidal oblong, with small guttules, occasionally one-septate. Chlamydospores are absent (Irinyi et al., 2009).

6.3. Disease cycle

The fungus produces numerous small spores, which can spread to healthy leaves and plants, thereby causing new infection. The fungus can survive on seeds and can spread with infected seeds.

6.4. Epidemiology

Cool, moist conditions favor disease development. Crop rotation and tillage will reduce survival of the fungus.

6.5. Integrated management

Use of pathogen free seed, in production fields where disease is severe, use of rotation and tillage, use of infested residues for next soybean crop as infested crops are possible source of inoculum. So, crop rotation and tillage will reduce the source of inoculum (Yang, 2002).

7. Conclusion

In this review, the economic importance, symptomatology, causal organism, epidemiology, disease cycle and integrated management strategies followed for every disease has been discussed. The causal agents differ for every leaf spot in soyabean and thus correct identification is necessary for efficient management strategies of the crop.

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