Fungal Biology

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Fungal diversity, ecology and control management

Springer

Fungal Biology

Series Editors

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Fungal biology has an integral role to play in the development of the biotechnology and biomedical sectors. It has become a subject of increasing importance as new fungi and their associated biomolecules are identified. The interaction between fungi and their environment is central to many natural processes that occur in the biosphere. The hosts and habitats of these eukaryotic microorganisms are very diverse; fungi are present in every ecosystem on Earth. The fungal kingdom is equally diverse, consisting of seven different known phyla. Yet detailed knowledge is limited to relatively few species. The relationship between fungi and humans has been characterized by the juxtaposed viewpoints of fungi as infectious agents of much dread and their exploitation as highly versatile systems for a range of economically important biotechnological applications. Understanding the biology of different fungi in diverse ecosystems as well as their interactions with living and non-living is essential to underpin effective and innovative technological developments. This series will provide a detailed compendium of methods and information used to investigate different aspects of mycology, including fungal biology and biochemistry, genetics, phylogenetics, genomics, proteomics, molecular enzymology, and biotechnological applications in a manner that reflects the many recent developments of relevance to researchers and scientists investigating the Kingdom Fungi. Rapid screening techniques based on screening specific regions in the DNA of fungi have been used in species comparison and identification, and are now being extended across fungal phyla. The majorities of fungi are multicellular eukaryotic systems and therefore may be excellent model systems by which to answer fundamental biological questions. A greater understanding of the cell biology of these versatile eukaryotes will underpin efforts to engineer certain fungal species to provide novel cell factories for production of proteins for pharmaceutical applications. Renewed interest in all aspects of the biology and biotechnology of fungi may also enable the development of "one pot" microbial cell factories to meet consumer energy needs in the 21st century. To realize this potential and to truly understand the diversity and biology of these eukaryotes, continued development of scientific tools and techniques is essential. As a professional reference, this series will be very helpful to all people who work with fungi and should be useful both to academic institutions and research teams, as well as to teachers, and graduate and postgraduate students with its information on the continuous developments in fungal biology with the publication of each volume.

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Preface

Food security is one of the major concerns in the world today. The continuously increasing world population coupled with global climate change poses a grave challenge to feed a billion mouths. Fungi play a significant role in modulating plant's environment, in both rhizosphere and phyllosphere resulting in growth changes that affect agricultural productivity significantly. Fungi have serious consequences on food security in the changing global environmental scenario.

Fungi play key roles in the ecosystem as decomposers, mutualists and pathogens and represent the second most diverse group after insects. Fungal diversity is not only reflected in its morphological characters but also in bioactive molecules produced, their pathogenicity and virulence, and impact on crop production. The increasing number of infectious fungal diseases is regarded as a global threat to agricultural productivity and food security. Therefore, it is important to document the fungal diversity and inventorize it. Further, to sustain agricultural productivity, it is important to mitigate or control the plant diseases. Plant pathogens cause severe losses to crops and significantly reduce the quality and quantity of agricultural products. The global tendencies are shifting towards a preferred use of various biocontrol agents in plant disease management. Fungal antagonists are widely used as biocontrol agents to control plant diseases globally. Biological control mechanism, however, needs an understanding of the complex interactions among plants, pathogens, and the environment.

This book provides a consolidated and comprehensive account of research being conducted by scientists all over the world in the areas of fungal biodiversity, fungal ecological services, fungal biology and ecology, and biological disease control and provides perspectives on crop protection and management and control of various fungal pathogens. The book also serves as an invaluable resource for researchers and educators working in the above fields. It will be useful to students studying mycology, plant pathology, crop protection, agricultural sciences, and plant sciences. Students will find this book handy to clear their concepts and to get an update on the recent research conducted in this area. Also, scientists involved in biological and

agricultural research, crop management and environmental sciences and industries that manufacture agrochemicals as well as small- to large-scale growers and producers will find the book useful.

Delhi, India — Vijay Rani Rajpal
Delhi, India — Ishwar Singh
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The editors gratefully acknowledge their families for their understanding, patience, and emotional support during the course of this book. Our sincere thanks are due to the whole Springer team involved in the production of this book. We especially appreciate Ms. Aakanksha and Ms. Priya for their continued support.

We are sure that this book will attract scientists, undergraduates, graduates, and postdocs who are working on fungal diversity, ecology, and biocontrol mechanisms.

Contents

Part III Disease and Control Management

About the Editors

Vijay Rani Rajpal M.Sc., M.Phil (Botany), Ph.D., is a Professor in Botany Department, Hansraj College, Delhi University, Delhi, India, and has ~25 years of research experience working on population biology, phylogenetics and diversity of plants. She has 45 published research papers, review articles and book chapters and has edited four books on molecular diversity, abiotic stress mitigation and improvement of plants published from Springer Switzerland. She has guided Ph.D. students and successfully completed a number of research projects funded by various agencies of the Government of India. She is a reviewer of several reputed national and international research journals and invited guest editor for a thematic issue of *Frontiers in Genetics*.

Ishwar Singh M.Sc. (Botany), Ph.D., a trained mycologist, has been researching in the field of Microbiology with special focus on mycotoxins, characterization of phytopathogens and their biocontrol for the last 20 years. He was a Visiting Faculty at the Department of Plant Pathology and Microbiology, Iowa State University of Science and Technology, Ames, Iowa, USA (2014–2015, Raman Research Fellowship). He has also been a recipient of UGC Research Award (2013). Currently, he is working as an Associate Professor in the Department of Botany at Hansraj College, University of Delhi, Delhi, India. He has published 30 research papers and articles and successfully completed a number of research projects. He is a member of the editorial board of many journals.

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Part I

Biology and Diversity of Fungi

Biology and Management of Spot Blotch Pathogen *Bipolaris sorokiniana* of Wheat

1

Rashmi Aggarwal, Shweta Agrawal, Malkhan Singh Gurjar, Bishnu Maya Bashyal, and M. S. Saharan

Abstract

Bipolaris sorokiniana (Teleomorph: *Cochliobolus sativus*), causal agent of spot blotch of wheat, emerges as a serious concern for yield losses to wheat crop in warm and humid regions of the world. Due to global warming and late sowing of wheat crop, spot blotch becomes a major concern worldwide. Spot blotch mainly occurs in North eastern plain zone in India as well as in other South Asian countries. To meet escalating demand of wheat crop in near future due to increasing global population growth rate and nutritive changes, management of this disease is necessary. This review summarizes the biology of the pathogen (*Bipolaris sorokiniana*) and an overview of distribution, impact, and management of the disease. In addition, it also provides insights into wheat—*B. sorokiniana* pathosystem at histological and molecular level. There are several approaches for the management of spot blotch disease, by way of identifying QTLs (quantitative trait loci) for spot blotch resistance, exercising marker-assisted selection and integrated approaches such as agronomic practices, proper crop rotation, biological control, and seed treatment with fungicides.

Keywords

Bipolaris sorokiniana · Spot blotch · Biology · Host–pathogen interaction · Management · Wheat

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1.1 Introduction

Wheat (*Triticum aestivum* L.) is one of the main cereal crops all around the world. It is grown in an area of about 220 million hectares with 763.06 million tons of production globally. Utmost area covered under wheat cultivation is in India (14%) shadowed by Russia (12.43%). India is the second major producer of wheat (98.5 mt) with the state of Uttar Pradesh being the largest producer (28 million tons) and holding largest share area wise (9.75 million hectare) in the country (Sendhil and Singh 2019). Improving yield and quality of wheat crop is necessary to cope up with the massive challenge of meeting future wheat demands with the same cultivable land and water available as on today.

The limitations in wheat production system include various pathological wheat diseases, among which fungal diseases like spot blotch has arisen as the most devastating disease in warm and humid environments of South Asian countries. Spot blotch/Leaf blight/foliar blight is caused by *Bipolaris sorokiniana* (Teleomorph: *Cochliobolus sativus*). Yield losses through *B. sorokiniana* have been reported to be highly variable from 2.7 to 100% depending on varieties across different countries (Mehta 1993 Duveiller and Gilchrist 1994; Villareal et al. 1995). The average yield loss has been estimated to be 15.5% in India due to spot blotch, which can be worst in severe conditions of infections (Joshi and Chand 2002). In addition to spot blotch, *B. sorokiniana* also causes common root rot in wheat in southern Brazil and Australia (Diehl et al. 1982; Tinline et al. 1988). Spot blotch disease is favored by many conditions like continuously increasing global temperature, rice–wheat cropping system (wheat sowing is delayed), and stress conditions (fertility is reduced) (Duveiller et al. 1998). At present *B. sorokiniana* is defined as the major pathogen in India and its frequency is highest in north eastern plains zone due to warm and humid weather conditions. *B. sorokiniana* infection increases with temperature, bright sunshine hours and in the morning. However, it is also observed that relative humidity (both maximum and minimum) and pathogen infection progression shows negative correlation (Devi et al. 2018).

In wheat cultivars, resistance to spot blotch is generally unsatisfactory due to environmental alteration, application of extreme or little fertilization, development of new races, and lack of effective durable resistance, therefore identification and characterization of new sources of resistance are helpful to overcome this disease (Mahapatra et al. 2020). Studies have been conducted on the biology and management of spot blotch fungal pathogen, over the last few decades which include: its life cycle, reproduction, survival, source of inoculum, symptoms developed on host plant, host range, diagnosis through various methods and management *via* integrated approaches and genetic resistance. A comprehensive information gathered from all these studies in the present genomics era is explicitly presented in this chapter.

1.2 Worldwide Distribution of the Pathogen

Spot blotch is common in the Mega Environment 5 (ME5) where humid and warm weather prevails during the growth of wheat crop. Surveys indicated that spot blotch has become a serious disease of wheat in several parts of the world, particularly in those areas characterized by moderate temperature and high humidity during the late growth stage such as eastern India, Bangladesh, Tarai of Nepal, and Brazil. Therefore, causal organism *B. sorokiniana* is considered as most destructive fungal pathogen in the warmer and humid areas mainly for wheat crop. Wheat grains are rich in carbohydrates, energy, dietary fiber, fat, riboflavin, protein, thiamine, niacin, vitamin B6, folate, pantothenic acid, calcium, magnesium, iron, phosphorus, zinc, potassium, and manganese (Gebhardt et al. 2006). Due to its high nutritive value, wheat grains are consumed in different forms across the world. *B. sorokiniana* also affects wheat in other warmer parts of the world like Asia, Latin America, Africa, Southern Asia, etc. The disease has been under investigation since it was first recorded in 1914 by Mohy in India (Joshi et al. 1986), but recently it has been recognized as a major concern. Severity of spot blotch has increased many fold after green revolution in India and in many countries of tropical and sub-tropical climate where rice–wheat cropping system is commonly practiced. Worldwide distribution of *B. sorokiniana* according to https://www.cabi.org/isc/datasheet/14694 is elaborated in Table 1.1.

Table 1.1 Worldwide distribution of *B. sorokiniana*

Continent	Country
Africa	Nigeria, Angola, Algeria, Cameroon, Ghana, Egypt, Kenya, Uganda, Ethiopia, Morocco, Libya, Malawi, Tunisia, Mauritius, Zambia, South Africa, Zimbabwe, Sudan, Tanzania
Asia	Thailand, Saudi Arabia, Azerbaijan, Afghanistan, Japan, Israel, Bangladesh, China, Bhutan, India, Kazakhstan, Indonesia, Iran, Myanmar, Iraq, Kyrgyzstan, Taiwan, Laos, Lebanon, Malaysia, Nepal, Oman, Uzbekistan, North Korea, Pakistan, Philippines, South Korea, Turkey, Sri Lanka, Syria
Oceania	Solomon Islands, American Samoa, Australia, Kiribati, Tonga, New Zealand, Papua New Guinea, New Caledonia
Europe	Belarus, Czechoslovakia, Belgium, Poland, Bulgaria, Croatia, Greece, Cyprus, Czechia, Federal Republic of Yugoslavia, Union of Soviet Socialist Republics, Serbia, Denmark, Estonia, Finland, France, Austria, Germany, United Kingdom, Italy, Norway, Hungary, Ireland, Lithuania, Moldova, Netherlands, Ukraine, Romania, Montenegro, Slovakia, Latvia, Spain, Russia, Sweden, Switzerland
North America	United States, Nicaragua, Canada, Guatemala, Costa Rica, El Salvador, Jamaica, Cuba, Mexico
South America	Venezuela, Argentina, Brazil, Colombia, Paraguay, Peru, Bolivia, Uruguay

1.3 Naming and Taxonomy of Spot Blotch Pathogen

B. sorokiniana (Teleomorph: *Cochliobolus sativus*) causing spot blotch disease was initially named as *Helmithosporium sorokinianum* Sacc. by Trans. Soc. Nat. Univ. Kazan 22:15 in Sorokin in 1890. In 1934, conidia at ascigereous stage (teleomorph) were first observed in the lab by Ito and Kurib and they named it as *Opliioholus sativus*, which was later renamed as *Cochliobolus sativus* by Drechsler ex Dastur in 1942.

In 1959, Shoemaker suggested the generic name *Bipolaris* by observing fusoid, straight, or curved conidia in helminthosporium species having one germ tube germinated from each end (bipolar germination). He renamed the spot blotch pathogen as *Bipolaris sorokiniana* (sacc.) shoem syn. *Drechslera sorokiniana* (Sacc.) Subrm and Jain. The genus *Bipolaris* belongs to Ascomycota, Pleosporales, Dothideomycetes, and Pleosporaceae. This pathogen has many names like *Helminthosporium sorokinianum* Sacc., *Helminthosporium acrothecioides* Lindfons, *Helminthosporium sativum* Pammel, *Helminthosporium californicum* Mackie and Paxton, C.M. King and Bakke, *Drechslera sorokiniana* (Sacc.) Subram. & B.L. Jain but valid name currently is *B. sorokiniana*. On the basis of character of conidial germination, it is named as *Cochliobolus sativus* at teleomorph stage and *B. sorokiniana* at anamorphic stage.

1.4 Reproduction of the Pathogen

B. sorokiniana causing spot blotch disease is the asexual state of the pathogen. *Cochliobolus sativus* (teleomorph) is its sexual stage, which is hardly perceived in nature but forms in pure culture with compatible mating types. Consequently, this pathogen reproduces mainly through asexual state through conidia. In *C. sativus* and some species of *Helminthosporium* sexuality and a parasexual cycle has been reported (Nelson 1960; Tinline 1962; Chand et al. 2003). One such study in eight different species of *Helminthosporium* confirms viable ascospores progenies from nine of the 13 interspecific crosses. In four crosses, perithecia were also observed signifying sexuality presence in some forms, and also presence of some pathogenicity genes, which segregate in these crosses. However, within or between *B. sorokiniana* isolates derived from wheat, sexuality has not been reported. *B. sorokiniana* is heterothallic and exhibits variability (Tinline 1951). Sexual reproduction of this pathogen is still baffling (Rapper 1966). Although, in Zambia, sexual reproduction of *B. sorokiniana* has been observed in the field conditions (Raemaekers 1987). Sexual reproduction leads to genotypic diversity which eventually helps the progeny to better fit into the changing environment (Colegrave 2002; Goddard et al. 2005; Heitman 2006, 2010; De Visser and Elena 2007). Historically, *B. sorokiniana* is a variable fungus (Christensen 1925; Tinline 1962) due to sexual reproduction, heterokaryosis and parasexual recombination (Tinline and Dixon 1958; Burdon and Silk 1997). Inheritance of the variability has been studied using two methodologies: crosses among two different genotypes trailed by an analysis of

segregation pattern (Mendelian approach), and another using molecular markers which are involved in a quantitative genetics approach (Gupta et al. 2017).

1.5 Pathogen Survival and Source of Inoculum

As the pathogen is seed borne, its survival on the host or in the soil as source of inoculum depends on several factors. In nature, the sources of inoculum of this pathogen are collateral hosts, infected seeds, free dormant conidia in the soil, and crop residues (Reis 1991). The presence of *B. sorokiniana* in seeds of wheat correlated with higher disease (Mehta 1981). The pathogen has been generally introduced through infected seeds into new wheat growing areas. In eastern India, seeds are considered as the most important source of inoculum for the reoccurrence of spot blotch in rice–wheat cropping system (Pandey et al. 2005). Sporulation of *B. sorokiniana* occurs on necrotic tissues of leaves and it reaches to the spike and finally moves to seeds. This sporulation continues on the residue till it gets completely decomposed. Pathogen can also survive in soil as free dormant conidia. Ability of fungus to colonize in diseased wheat straw results in pathogen survival in the soil, and the inoculum density of the pathogen in soil is associated to the quantity of sporulation occurring in crop residues (Burgers and Griffin 1968; Reis and Wunsche 1984). Melanin content in pathogen has a direct association with conidiogenesis, signifying that melanin formed by the pathogen neutralizes antimicrobial activity of the host cells, accordingly contributing to the pathogen survival (Henson et al. 1999; Aggarwal et al. 2011a). Large number of plant species act as collateral hosts for the pathogen such as weeds that are ubiquitous in different cropping systems and also present on adjacent uncultivated land (Neupane et al. 2007). *B. sorokiniana* infects a wide host range differing from isolate to isolate. *B. sorokiniana* mainly infects Poaceae members including *Triticum aestivum*, *Bromus erectus*, *Secale cereale*, *Hordeum vulgare*, *Alopecurus pratensis*, *Hordeum murinum*, *Avena sativa*, *Agropyron pectinatum*, *Agropyron repens*, *Beckmannia eruciformis*, *Poa pratensis*, *Pennisetum villosum*, *Bromus inermis*, *Festuca heterophylla*, *Festuca ovina*, *Lolium perenne*, *Setaria virdis*, and *Dactylis glomerata*. However, it is also reported in some dicot crops including Beans, Alfalfa, Red and Yellow clover, and Buck wheat. Chinese isolates have been shown to infect 29 graminaceous hosts. *Zizania caduciflora*, *Saccharum officinarum*, *Paspalum thymbergii*, *Apluda mutica*, *and Ischaemum ciliare* have been reported as new hosts for the pathogen (Naitao and Shenyang 1987). Therefore, the knowledge of inoculum survival of a pathogen in off-season through diverse sources is very vital to develop suitable disease management approach.

1.6 The Pathogen: Primary and Secondary Disease Cycle and its Morphology

B. sorokiniana (Sacc.) can survive in dormant phase on stubbles, seeds, and roots; but once it becomes active, primary infection initiates. Pathogen can survive on stubbles or soil for many months and infects the healthy wheat plants through conidial spore germination followed by germ tube formation (Aggarwal et al. 2008; Acharya et al. 2011), which ultimately produces an appressorium within 8 h, which later on develops hyphae. Within 12 h of hyphae formation, it penetrates into the cuticle of hosts (Sahu et al. 2016). In host, the hyphae multiply rapidly and spread into intercellular space infecting the mesophyll tissue of the wheat leaf. After 48 h of multiplication in the intercellular space, conidiophores are formed which are 100–150 × 6–8 μm long and integrate a new generation of conidia in it. The conidia (60–120 × 15–20 μm in size) are olive brown in color, thick walled, and tapered toward the end (bipolar) with 5–9 septa; which is inclined for secondary infection (Fig. 1.1). Conidia mainly disperse through rain or dew therefore secondary infection of spot blotch is mainly caused by airborne conidia (Duveiller et al. 2005).

B. sorokiniana is demarcated from other *Bipolaris* species, in terms of morphological features of conidiophores and conidia. In axenic culture, growth on PDA (potato dextrose agar) plates or test tubes shows maximum radial growth with loose cottony mass at initial phase of hyphal development, which later on turns to fluffy blackish colonies due to sporulation. The colonies can be white or light to dark gray depending on different isolates (Kumar et al. 2002; Aggarwal et al. 2009). Appressoria development at the initial stages can be clearly seen under light

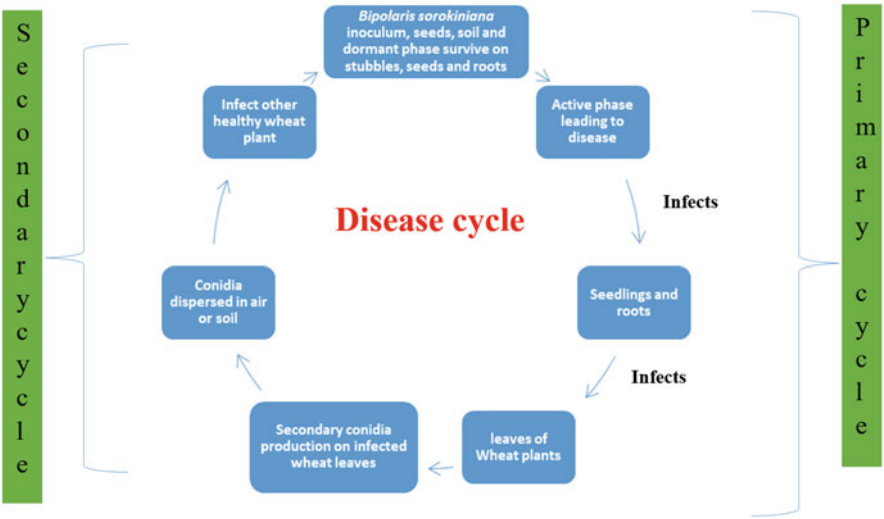

Fig. 1.1 Primary and secondary disease cycle of *Bipolaris sorokiniana*

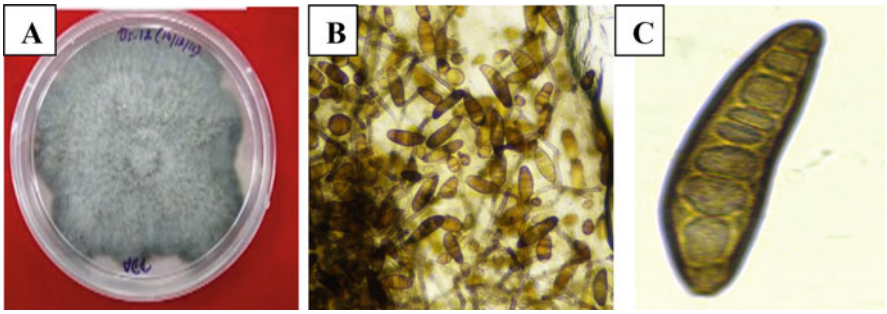

Fig. 1.2 Morphology of *Bipolaris sorokiniana*. (**a**) Fungal growth on PDA. (**b**) Conidia showing polar germination (10×). (**c**) Conidium showing 5–8 thick walled septa (40×)

microscope. Conidia appear black and shiny with 5–8 thick walled septa (Fig. 1.2) under light microscope with bipolar germination (Aggarwal et al. 2002).

1.7 Symptomatology

Symptoms of spot botch of wheat typically appears on leaf, sheath, nodes, and glumes; but in more severe conditions it affects spike and develops dark brown to black discoloration which is called as Black point. Lesion are developed on the leaf, coleoptiles, crowns, storms, and roots starting from few mm and extend up to 1–2 cm which appear as a dark brown spot (Chand et al. 2002). These spots progress and join with other spots forming large blotches that damage the entire part of the plant eventually killing it. Under humid conditions, conidia before coalescing develop on leaves and induce leaf tissue death. Due to copious production of these conidia under humid conditions, chlorotic streaks sometimes diffuse from the border of the lesions, which result in toxin production (Mercado vergnes et al. 2006; Bockus et al. 2010). Heavily infected plants may cause stunting and reduced tillering which ultimately lead to premature death.

1.8 Fungal Isolation and Its Diagnosis

Symptoms developed on various parts of the host plant are used for collection, isolation, diagnosis and to check the severity of the disease. For the isolation of this pathogen, leaves that are infected are surface sterilized and necrotic lesions are cut into smaller fragments. These fragments are washed with sodium hypochlorite ($NaOCl_2$) solution, followed by two times washing with water. To induce sporulation, these fragments are placed on PDA (potato dextrose agar) under a 12-h photoperiod at room temperature. Several single spores from each of the PDA plates are transferred to another PDA plate and are allowed to grow at 25 °C. Meanwhile, these spores are observed under microscope and fungus is identified based on

conidia and mycelial morphology. Conidiophores observed are unbranched, septate with conidia being brown to olivaceous brown in color.

Microscopic identification of the pathogen further needs verification, for which many molecular markers have been developed. RAPD (Random Amplified Polymorphism DNA) markers are very versatile, promising and informative tool to detect pathogen isolates, which are organized into different groups on the basis of their color and shape of colonies (Pandey et al. 2008). A SCAR marker (sequence characterized amplified region) based on diagnostic PCR assay was developed to detect this pathogen in wheat leaves and field soil, which detected the pathogen at very early stage even before the visual symptoms appear (Aggarwal et al. 2011b). More recently, the RPA (recombinase polymorphism amplification) assay was developed which is more rapid and effective method for detection of *B. sorokiniana*. This RPA depends on the sequences of calmodulin gene sequences (Zhao et al. 2021).

1.9 Disease Assessment

Aggressiveness or disease severity of the pathogen can be tested through a continuous scale using two available methods. Firstly, a single digit scoring method called as ADI (average disease index) calculated using 0–5 scale. In this disease severity (%) is calculated as per the scale; 0 = free of spots; 1 = up to 5% area of leaf enclosed with necrotic spots; 2 = 6–20% of the leaf area covered; 3 = 21–40% of the leaf area covered; 4 = 41–60% of the leaf area covered; 5 = spots inclusion more than 60% of the leaf area tangled.

$$\text{ADI} = ((\text{sum of rating of each leave})/(\text{total leaf} * 5)/100)$$

ADI is converted into disease responses *viz.* 0 = No infection; 0–10 = resistant response (R); 11–20 = moderately resistance (MR); 21–30 = moderately susceptible (MS); 31–50 = susceptible (S); and more than 50 = highly susceptible (HS) (Adlakha et al. 1984).

Secondly, a double digit scale (00–99), where the first digit (D1) signposts disease progress in the canopy height from ground level; the second digit (D2) denotes severity based on diseased leaf area. Both D1 and D2 are scored on a scale of 1–9. For each score, the percentage of disease severity is estimated based on the following formula:

$$\text{Severity } (\%) = (\text{D1}/9) \times (\text{D2}/9) \times 100$$

This disease evolves very rapidly around the affected portion, so it is necessary to record disease scores per plot at 3–7 days intervals over 3- to 4-week period between anthesis and the dough stage (Duveiller and Sharma 2009). The area under the disease progress curve (AUDPC) can be calculated using the formula for percentage severity:

$$\text{AUDPC} = \sum_{i=1}^{n-1} [(X_i + X_i + 1)/2](t_i + 1 - t_i)$$

where X_i severity on the ith date, t_i ith day, and n number of dates on which the disease recorded.

AUDPC calculation is categorized into disease responses as: (1) immune (00), (2) *R* resistant (<12), (3) MR moderately resistant (12–34), (4) MS moderately susceptible (56–68), (5) S susceptible (78–89), and (6) HS highly susceptible (89–99).

Recently, imaging technique has also been introduced to sense electromagnetic spectrum external to visible light, which allows enumerating disease symptoms that are not visible by eye (Mukta et al. 2015). More recently, a reliable and accurate disease detection is facilitated, i.e., sensor-based analysis such as RGB imaging, multi- and hyper-spectral sensors, chlorophyll florescence or thermography (Mahlein 2016).

1.10 Toxin Production and Host–Pathogen Interaction

Etiology of spot blotch disease majorly depends on toxins. Toxins are the secondary metabolites produced by a pathogen that penetrate the host and cause disease. Role of toxins in pathogenesis is reported in differentiating resistance and susceptible genotypes of wheat (Aggarwal et al. 2008). Phytopathogenic fungus *B. sorokiniana* produces a series of non-host selective toxins which are sesquiterpenoid toxins that are synthesized from farnesol and belong to eremophilane family like prehelminthosporol, helminthosporic acid, helminthosporol, sorokinianin, victoxinine, and bipolaroxin (Nakajima et al. 1994; Olbe et al. 1995; Apoga et al. 2002; Kumar et al. 2002; Jahani et al. 2006). The most abundant and active phytotoxin formed by the pathogen is Prehelminthosporol ($C_{15}H_{22}O_2$), a hydrophobic sesquiterpene with stumpy water solubility and little thermal stability (Carlson et al. 1991). It interrupts the integrity of cell organelles, plasma membrane and it stimulates enzyme callose synthase in host plants (Olbe et al. 1995; Apoga et al. 2002). Helminthosporol is synthesized from Prehelminthosporol, which on further transformation gives helminthosporic acid. Both helminthosporol and helminthosporic acid are plant growth regulators which inhibit seed germination and shoot growth (Qader et al. 2017). Sorokinianin, a novel phytotoxin is derived biogenetically from a farnesyl pyrophosphate having an inhibitory effect on germination of seeds (Nakajima et al. 1994). Victoxinine ($C_{17}H_{29}N_O$) is a toxic metabolite, a tricyclic base (sometimes called victorin) inhibits root growth (Pringle 1976). A bicyclical sesquiterpene belonging to family Eremophilane compound was identified and named as bipolaroxin. From culture filtrate of virulent isolate BS-75, bipolaroxin was purified using prep TLC, which is characterized further using NMR and GC-MS techniques. It produces necrotic lesions not only on wheat but also on barley, sorghum, maize, *Phalaris minor*, *Cynodon dactylon* and *Avena sativa* as studied using leaf infiltration bioassay (Jahani et al. 2006, 2014).

1.11 Population Differentiation and Molecular Characterization of *B. sorokiniana*

It is observed that like many other plant pathogens, specialization for virulence also arises in *B. sorokiniana* which is apparently more in barley as a host (Christensen 1922). Different isolates of *B. sorokiniana* do not show clear and unique differential virulence patterns on wheat genotypes like rust and it also has a range of aggressiveness in different strains with no specific host–pathogen interactions (Maraite et al. 1998; Duveiller and Garcia 2000; Aggarwal et al. 2019). In earlier study, it was noticed that only 1–2% of the variance is observed between 12 wheat differentials inoculated with 206 *B. sorokiniana* isolates (Hetzler et al. 1991). However, three, four or six pathotypes (0, 1, 2) or eight virulence groups (VIGs) were recognized in barley—*B. sorokiniana* pathosystem (Ghazvini and Tekauz 2007).

B. sorokiniana shows vast physiological and morphological variability with respect to multinucleate mycelium and conidia, with subsequent heterokaryosis (Mitra 1931; Day 1974). It was firstly reported that *B. sorokiniana* isolates varied significantly with respect to their virulence on wheat and barley (Christensen 1926). Many reports on barley—*B. sorokiniana* pathosystem are available, but scarce reports were available for wheat—*B. sorokiniana* pathosystem. *In vitro* morphological variability based on color and morphology of colonies observed on PDA (potato dextrose agar) plates has been reported in the population of wheat—*B. sorokiniana*, which ranges from black to white, and confirms more viability and aggressiveness in black colony (presence of melanin) as compared to white colony (Chand et al. 2002). Fifteen pathotypes of *B. Sorokiniana* monoconidial isolates were observed from diverse parts of the world (Hetzler et al. 1991). Earlier, pathological variability was testified, but it has not been correlated with the morphological or physiological variability. Morphological, molecular, and pathogenic variability has been studied in Brazilian (Oliveira et al. 1998) and Bangladesh isolates (Ahmed et al. 1997). A study was conducted to assess the cultural, pathogenic, and genetic variability in the Indian isolates of *B. sorokiniana* of wheat cultivars collected from different agro climatic zones, which confirmed the existence of five pathotypes in India (Aggarwal et al. 2009). The morphological variability correlated with pathological variability resulted in monitoring the populations of *B. sorokiniana*.

Heterokaryosis and parasexuality are the two major aspects that may be responsible for variability in the pathotypes of *B. sorokiniana*. Less than 10% of the total conidia form heterokaryotic conidium. Heterokaryosis (single hyphal cells comprising 3–6 nuclei) conditions may result from fusion between head-to-head hyphae belonging to the same or different individual mycelia through anastomosis (Glass et al. 2000). Heterokaryons can also be formed from nuclear migration amidst two genetically different hyphae (Tinline 1962). Heterokaryosis play important role in the variation of the pathogen. The isolation of recombinants constituted the genetic evidence of somatic heterozygous diploid and parasexuality in the fungus. Parasexual cycle is led by presence of heterokaryons recombination, during which nuclear and cytoplasmic material is swapped between two anastomosing hyphae (Burdon

and Silk 1997). In *B. sorokiniana*, the parasexual recombination is complex and very rare due to the presence of *Het* genes, which restrain the heterokaryon formation. Individuals differing at one or more of the loci of *Het* gene, the hyphae are aborted and it results in vegetative incompatibility (Glass et al. 2000).

Molecular characterization of the fungal isolates involves several approaches which includes: (1) molecular markers such as SCAR marker (sequence characterized amplified region) which is based on diagnostic PCR assay, RAPD (random amplified polymorphism DNA) markers, RPA (recombinase polymorphism amplification) assay, AFLP (amplified fragment length polymorphism), SSR (simple sequence repeat), RFLP (restriction fragment length polymorphism), etc.; (2) karyotyping using squashing and microscopic examination (Hrushovetz 1956; Huang and Tinline 1974), GTBM (germ tube burst method), and CHEF (contour-clamped homogeneous electric field method) (Zhong and Steffenson 2001; Zhong et al. 2002); and (3) whole genome sequencing using different platforms like Illumina Hiseq, Oxford nanopore sequencing, and ion-torrent platform technologies.

Molecular markers are an excellent tool for genetic analysis of a pathogen genome. PCR-based markers have been the most frequently used molecular markers for molecular characterization and variability study of *B. sorokiniana* isolates. For the study of diversity and variability among *B. sorokiniana* isolates, a number of universal rice primer (URP)-PCR-based markers were studied (Aggarwal et al. 2010). According to this study, from 12 URP markers 10 markers generated polymorphic fingerprint patterns in DNA of *B. sorokiniana* isolates obtained from diverse geographic regions. Ribosomal DNA polymorphism can also be used for characterization of isolates which involves sequence variation in the ITS (Aggarwal et al. 2014). As described earlier, a SCAR marker was developed to detect *B. sorokiniana* at pre-symptomatic stage in wheat tissue and field soil (Aggarwal et al. 2011b). Marker designated as SCRABS600 could clearly differentiate *B. sorokiniana* from other fungal plant pathogens. RAPD (random amplified polymorphism DNA) markers were used to detect *B. sorokiniana* isolates (Pandey et al. 2008). Markers from another species (*Pyrenophora teres* f.sp. *maculate*) were also developed and used for exploring the variation in *B. sorokiniana* isolates (Lu et al. 2010). RPA (recombinase polymorphism amplification) assay has also been developed which is more rapid and effective method for detection of *B sorokiniana* (Zhao et al. 2021). Molecular maps of *B. sorokiniana* whole genome covering all the 15 chromosomes, based on molecular markers (e.g., RFLP, SSR, AFLP, etc.) have also been successfully established (Mann et al. 2014).

1.11.1 Chromosome and Karyotypes of *B. sorokiniana*

Earlier, on the basis of microscopic examination, the haploid chromosome number of *B. sorokiniana* was $n = 7$ or 8. But later on, by GTBM and CHEF analysis it was found to be $n = 15$. To identify chromosome length polymorphisms the barley *B. sorokiniana* collected from varied regions of the world (USA, Japan, Canada,

Brazil, Poland, and Uruguay), were analyzed for the karyotypes using CHEF electrophoresis of 16 isolates comprising the three known pathotypes of *B. sorokiniana*, i.e., 0, 1, and 2. CHEF bands ranging from 8 to 13 were observed with a size range of 0.85–3.80 mega-bases (Mb). A unique banding pattern was observed in each of the 16 isolates, except for two isolates, i.e., ND90Pr and ND91-Bowman (North Dakota isolates). This signifies that large-scale structural changes took place in *B. sorokiniana* isolates at the chromosome level. This study has not been subjected to wheat isolates. However, study on karyotypes from wheat isolates can be rewarding to recognize the causes of variation and also to what extent structural changes can be a part of variation. Karyotypes results concluded that in the virulent isolate ND90Pr, at least five of the fifteen chromosomes were involved in translocations. In *B. sorokiniana* chromosomes, structural rearrangements are very common between the corresponding chromosomes responsible for length polymorphisms of two isolates of opposite mating types with complementary virulence (ND93-1 and ND90Pr). Hybridization through southern blots carrying CHEF-separated chromosomes can be used for single-copy DNA probes, to recognize highly polymorphic chromosomes among isolates. Such a comparison allowed identification of unequal chromosomal rearrangements among dissimilar isolates. For hybridizations with the CHEF blots, DNA markers linked to *VHv1* were also used as probes which suggests that the chromosome carrying *VHv1* in a specific isolate was longer than the corresponding isolates chromosome that lacked this gene. This recommends that the genomic region carrying *VHv1* is exclusive (Gupta et al. 2018).

1.11.2 Whole Genome Sequence Analysis

Using whole genome sequencing (WGS), host–pathogen interactions at the molecular level in several pathosystems, synthesis of secondary metabolites through genes encoding enzymes and other proteins which are involved in virulence can be identified. For wheat, till now whole genome sequences of seven *B. sorokiniana* isolates are available. Using next generation sequencing technology, whole genome sequence of Indian virulent isolate BS 112 of *B. sorokiniana* was generated (Aggarwal et al. 2019). A total sequence assembly size of 35.64 Mb was predicted with GC content of 50.2%, providing coverage of 97.6% on reference ND90Pr genome. A total of 235 scaffolds were obtained using pyScaf assembler with N_{50} of 1,654,800 bp. In addition, 152 transcription factors involved in various biological processes were identified and a total of 682 secretory proteins were predicted using secretome analysis. Total of 10,460 genes were analyzed with an average gene density of 250–300 genes/Mb. The average gene length predicted was 435–545 bp, the maximum gene length was 8506 bp, and the minimum gene length was 50 bp. Gene ontology (GO) annotations resulted in 10,460 annotated genes, which was further characterized into 1024 genes for biological processes, 493 genes for cellular components, and 1274 genes for molecular functions. Single-nucleotide polymorphisms (SNP) were identified as a result of which 93,122 variants

containing 88,672 SNPs and 4450 indels were identified. Further, simple sequence repeat (SSR) identification using the MISA tool version showed 5996 SSRs, and 146 of the 235 SSR-containing sequences were further examined (Aggarwal et al. 2019). Comparative secretomics analysis between available whole genome sequence of *B. sorokiniana* and *B. oryzae* led to the identification of 262 and 247 predicted small secreted proteins (SSPs), respectively, out of which 34 and 28 SSPs respectively were assigned gene ontology terms for putative function (Singh 2016). Functional analysis of several strain unique polyketide synthase and non-ribosomal peptide synthetase revealed a strong correlation with a role in virulence (Condon et al. 2013). Three Australian isolates of *B. sorokiniana* (BRIP27492, BRIP26775 and BRIP10943) were sequenced and screened for identification of the *ToxA* pathogenicity gene (Mcdonald et al. 2018). During Perrenoud (1990), *ToxA* was discovered in *Parastagonospora nodorum* causing Septoria nodorum blotch (SNB) which horizontally got transferred to *Pyrenophora tritici repentis* causing tan spot (Friesen et al. 2006; Stukenbrock and McDonald 2007). *ToxA* element is unique but not conserved in *Pyrenophora tritici repentis* genome (Moolhuijzen et al. 2018). More recently, Friesen et al. (2018) showed that *ToxA* gene is also present in the *B. sorokiniana* population in the winter wheat region of the United States. Necrosis was induced on wheat leaves of *ToxA*-sensitive wheat genotypes (possessing the *Tsn1* susceptibility gene) (Faris et al. 2010). A sensitivity gene *Tsn1* occurrence in wheat generally helps a *ToxA* positive pathogen to cause spot blotch disease. In general, the *ToxA-Tsn1* system is an illustration of an inverse gene-for-gene relationship (Navathe et al. 2019). *ToxA* gene was amplified in different isolates of *B. sorokiniana* collected from different regions of India, confirming the presence of this gene in Indian population (Anonymous 2019).

To understand genetic and molecular interactions of *B. sorokiniana* with its cereal hosts, it is significant to isolate and characterize genes for virulence or pathogenicity in the pathogen and resistance in the host based on genomic information. The gene responsible for infecting wheat was labeled as *VTa1* (Zhong et al. 2002). A dominant gene *Rbs7* conferring resistance to spot blotch was mapped in a genomic interval of 304 kb on barley chromosome 6H (Wang et al. 2019). A new spot blotch resistance gene designated as *Sb4* was identified and mapped in a genomic interval of 1.34 Mb on long arm of wheat chromosome 4B (Zhang et al. 2020). *VHv1*, a virulence gene was also identified in barley—*B. sorokiniana* pathosystem (Zhong et al. 2002). Recently in wheat—*B. sorokiniana* pathosystem, *ToxA* gene (virulence gene) has been mapped to wheat chromosome arm 5BL, which reveals inverse gene to gene relationship with a sensitivity gene called as *Tsn1* in wheat through which fungus invades the host and disease is caused (Liu et al. 2006; McDonald et al. 2018).

1.12 Disease Management

1.12.1 Use of Resistant Genotypes

Spot blotch disease resistance crosses involved moderately resistant cultivars like BH 1146 from Brazil. However, when similar tests were carried out at Poza Rica, Mexico (CIMMYT = International Maize and Wheat Improvement Centre), the level of resistance was insufficient. Resistance in wheat genotypes is mainly controlled by two to three genes, but in some of the Chinese lines such as Longmai 10 and Yangmai 6, polygenic resistance is observed (Sharma et al. 1997).

Spot blotch disease infection may occur at any growth stage of the crop and it is an important factor to decide the extent of losses in grain yield, therefore it is essential to assess resistance at important growth stage. A study was undertaken on character association analysis to assess the nature of magnitude of association between grain yield components and disease severity (Singh et al. 2008). A study conducted in different environmental conditions confirmed that a few genotypes had low disease severity while some showed higher disease severity in commercial cultivars of South Asia (Sharma et al. 2007). Another study reported high resistance in some wheat genotypes under different environmental conditions (Sharma et al. 2004; Kumar et al. 2015). A few genotypes (Chirya 7, Yangmai 6, and Chirya 1) with lower disease severity had relatively low grain yield and weight in South Asia. However, the genotype with the maximum grain yield and weight (Altar-84/*Ae. sq.* (224)//Yaco) also had less disease severity. This displays progress in merging high grain yield and spot blotch resistance, which was not possible earlier. High yielding commercial wheat cultivars in the region with lower resistance still shows 20% of yield loss due to spot blotch disease (Siddique et al. 2006). Wheat genotype K 8027 shows good level of resistance as studied earlier (Dubin et al. 1998), and reported that in early 1990s leading commercial wheat cultivars of South Asia had developed spot blotch disease severity than genotype K 8027. These findings demonstrate that improvement achieved in the eastern gangetic plains of South Asia is due to combined efforts at international level in improving spot blotch resistance of wheat cultivars. This confirms that disease resistant wheat genotypes are available for direct use in breeding programs to develop commercial cultivars.

During the late 1980s, the extensive crossing program at CIMMYT produced resistant sources, which contained *Thinopyrum curvifolium* for spot blotch resistance as an alien donor in their pedigree (Duveiller and Gilchrist 1994). Donors for resistance may include many species of *Aegilops* and *Triticum* species, such as *Aegilops triuncialis*, *A. speltoides*, *A. cylindrical*, *A. triaristata*, *T. dicoccoides*, *T. timopheevii*, *T. araraticum T. boeoticum*, *T. persicum*, *T. urartu*, and *T. sphaerococcum* (Singh and Dhaliwal 1993; Smurova and Mikhailova 2007). Further, extensive studies were conducted in different parts of the world through conventional breeding for selection of genotypes resistant for spot blotch disease in wheat. However, there is still need for further assessment of sources conferring resistance to spot blotch.

1.12.2 QTLs Identification

It is often challenging to achieve the anticipated level of resistance of host through conventional breeding. Genetics of resistance to spot blotch has been classified into two major categories. Mendelian approach as a first category involving crosses between resistant and susceptible genotypes, and quantitative genetics approach as a second category using molecular markers. Earlier, spot blotch resistance on genetic basis has been recognized as eight major QTLs, viz., QSb.bhu-2A, QSb.bhu-2B, QSb.bhu-2D, QSb.bhu-3B, QSb.bhu-5B, QSb.bhu-6D, QSb.bhu-7B, and QSb.bhu-7D (Kumar et al. 2009, 2010). Three microsatellite markers (Xgwm67, Xgwm469, and Xgwm570) allied with spot blotch resistance were reported by Sharma et al. (2007). Of these QTLs, four with major and stable effects have been designated as *Sb1* on chromosome 7DS (Lillemo et al. 2013), *Sb2* on 5BL (Kumar et al. 2015), *Sb3* on 3BS (Lu et al. 2016), and *Sb4* on 4BL (Zhang et al. 2020). Recently, four promising new QTLs on different chromosomes were identified, namely 1A (497.2 Mb), 1D (89.84 Mb), 2B (421.92 Mb), and 6D (6.84 Mb) linked with numerous protein families which show resistance to disease (Tomar et al. 2021). *Lr34* and *Lr46*, broadly described genes linked with leaf rust resistance, have also been reported in spot blotch resistance. *Lr34*, main locus for spot blotch resistance on chromosome 7D explaining up to 55% phenotypic variation across the six environments in the mean disease data, this locus was designated as gene *Sb*1 (Lillemo et al. 2013). Over the last few years, for spot blotch resistance, several QTLs and genetic markers have been recognized in wheat (Gurung et al. 2014; Zhu et al. 2014; Singh et al. 2018). Resistance genes effectiveness can be lost over time, due to evolutionary changes in pathogen populations. Therefore, breeding for disease resistance in wheat is the outcome of identification and mapping of novel resistance. This can be achieved through association mapping of spot blotch resistance QTLs.

1.12.3 Agronomic Practices

Manipulation of agronomic practices from different countries was suggested to manage spot blotch, like use of different mineral nutrients (Singh et al. 1998; Krupinsky and Tanaka 2000). Some reports also suggest the role of potash in reducing spot blotch disease severity (Regmi et al. 2002). Spot blotch disease severity can be reduced to certain level through good crop husbandry and optimum agronomy (Sharma et al. 2006). Potassium helps to reduce disease severity by hindering multiplication and survival of pathogen and it also controls the internal metabolism of the host plant and prevents the establishment and spread of the pathogen within the host species (Perrenoud 1990). It is reported that application of nitrogen can upsurge the severity of spot blotch (Singh et al. 2012). Crop rotation is an integrated management strategy which helps to manage spot blotch in the field. It promotes better plant nutrition and also favors beneficial soil organisms. Crop rotation is an appropriate way to break the cycle of disease by growing unrelated

crops. It is difficult to find out the suitable non-host crop for spot blotch pathogen because of wide host range. This pathogen can also survive on weeds, therefore, plowing the volunteer cereals, stubble, and grass weeds can reduce inoculum of the pathogen in the field (Diehl et al. 1982).

1.12.4 Fungicides, Bioagents, and Botanicals

For the management of spot botch several fungicides have been recommended for use. The fungicides like difenoconazole (Ishikawa et al. 2012), carbendazim (Yadav et al. 2013), propiconazole (Singh and Singh 2009; Gupta et al. 2018), and azoxistrobin (Navathe et al. 2019) were found efficient in managing spot blotch. At particular stage of host plant, i.e., between heading and grain filling stage, application of fungicides such as epoxiconazole, tebuconazole, cyproconazole, flutriafol, epoxiconazole, flusilazole, and metaconazole have been proved to be cost effective. The yield increase in fungicide-treated areas suffering from spot blotch diseases in comparison to untreated areas was 30% in Argentina (Castro et al. 2018) and 10% in Sweden (Djurle et al. 2018). Besides, it is reported that silver nanoparticles also act as fungicide against spot blotch (Mishra et al. 2014). Similarly, the use of silicon was also established to improve resistance of wheat against spot blotch (Domiciano et al. 2010). Seeds are the primary source of infection. Thus, seed treatment with a suitable fungicide can reduce inoculum potential. In Nepal, seed treatment with fungicide vitavax 200 B and bavistin reduced seedling infection and increased seed germination by 43% (Sharma et al. 2005). Seed treatment with vitavax 200B and carbendazim improved early plant establishment in wheat–rice rotation cropping soil areas. The seed treatment with fungicidal formulation vitavax 200 WS (carboxin + thiram 1:1) @ 2.0, 2.5, and 3.0 g/kg seed reduced incidence of foliar diseases and seedling mortality at different locations of India (Singh et al. 2007). Later on, seed treatment with vitavax power at 3 g/kg of seed followed by two sprays of propiconazole at 0.1% at the early disease infection stage, reduced disease intensity by 39.03% (Singh 2017).

The consequences of recommended dose of fungicides (carbendazim, propiconazole, and hexaconazole), bioagents, and botanicals on seed yield of wheat and severity of spot blotch disease have been documented (Yadav et al. 2015). Bio-control of spot blotch is usually restricted by environmental factors and growing conditions. Successful antagonists in suppressing *B. sorokiniana* identified are *Chaetomium* sp., *Gliocladium roseum*, and *Idriella bolleyi* (Knudsen et al. 1995) and recently, *Bacillus subtilis* TE3 strain verified to be effective against seed borne *B. sorokiniana* (Villa-Rodríguez et al. 2019). Furthermore, *Bacillus safensis* and *Ochrobactrum pseudogrignonense* have been described to stimulate spot blotch resistance (Sarkar et al. 2018). *Chaetomium globosum* Kunze is considered as a potential antagonist of numerous soil and seed borne plant pathogens (Vannacci and Harman 1987; Walther and Gindrat 1988). Bioefficacy of *C. globosum* under in vitro, in vivo, and mechanism of antagonism has been studied in detail (Aggarwal et al. 2004; Aggarwal 2015). Culture conditions for resourceful production of

extracellular xylanase from *C. globosum Cg 2* have been standardized and partially purified enzyme has been characterized. Antifungal activity was shown by both purified xylanase (100 μg ml^{-1} concentration caused 100% inhibition of conidia germination) and culture filtrate (inhibit germination up to 67.5%) against *B. sorokiniana* (Ahammed et al. 2008). Further, an extracellular β-1, 3-glucanase produced by *C. globosum* isolate Cg2, inhibited 93.5% conidial germination of spot blotch fungus *B. sorokiniana*, whereas the culture filtrate inhibited conidial germination up to 59.9% (Ahammed et al. 2012). Further, biology, bioefficacy, and mechanism of action of *C. globosum* against plant pathogens have been reviewed (Aggarwal 2015). *C. globosum Cg 2* also detoxifies the toxin Bipolaroxin produced by *B. sorokiniana* (Aggarwal et al. 2011c).

There are reports that application of ethanolic plant extracts, aqueous plant extracts in amalgamation with cow dung, and aqueous plant extracts in amalgamation with cow urine can inhibit conidial germination of *B. sorokiniana* causing spot blotch. Additionally, ethanolic extracts of *Adhatoda vasica* (leaf) and *Zingiber officinale* (rhizome) at 2.5% provided 100% inhibition of conidial germination (Akhter et al. 2006). The leaf extract of *Rauwolfia serpentina* at 10M concentrations inhibited the spore germination up to 93.7% and increased grain yield by 28.9% (McDonald et al. 2018). Garlic extract treatment of wheat seeds reduced the incidence of spot blotch and increased the seed germination (Khalaf et al. 2011). Extracts of garlic clove, eucalyptus leaf, neem cake, and neem leaf at tillering and boot leaf stage resulted in the higher yield of wheat (Yadav et al. 2015). Six plants extracts, namely garlic, clove, eucalyptus leaf, neem leaf, onion bulb, ginger rhizome, and black cumin, could significantly inhibit the growth of *B. sorokiniana* (Tiwari and Singh 2021).

1.13 Conclusions

It may be concluded that this disease will continue to be a major concern worldwide especially in warm and humid regions despite the substantial progress already made in understanding the biology of the pathogen. Therefore, there is a need of further attention from the researchers for the management of this disease, which can be a threat in near future due to climate change and changing agronomy. QTLs have been identified which will help to develop new resistant cultivars. However, resistance is very low in most of the cultivars. Improving resistance through hybrid program by exploring new resistance donors should be done continuously to keep disease at its lowest level, improving grain yield and to develop resistant cultivars for future use. Integrating conventional breeding, molecular approaches, application of fungicides and bioagents will offer eco-friendly and cost-effective control of spot blotch disease worldwide. The information generated through genomics will further aid in better understanding of pathogenesis leading to disease management.

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