Breeding for Spot Blotch Resistance in Wheat

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Abstract

Spot blotch is an important fungal disease caused by *Bipolaris sorokiniana* which affects wheat crop in South Asia and South America. This disease causes yield losses ranging from 15 to 25%. The disease also affects quality of harvested wheat grains. The chief symptoms of the disease include small, dark brown lesions ranging from 1 to 2 mm in length without chlorotic margin, and the lesions coalesce and induce the death of the leaf. Host resistance is recognized as an economical and eco-friendly approach of managing spot blotch, and the resistance is controlled by polygenes. A number of resistance sources have been identified and utilized in breeding varieties which were made available for cultivation. With the use of molecular marker technology and genome sequencing platforms some of the resistance genes have been identified and used in breeding using marker-assisted selection approach. This chapter focuses on the recent understanding of the genetics of resistance, identification, and mapping of new sources and genes/QTLs and breeding efforts to develop new improved genotypes with better resistance against spot blotch to ensure food security in the world.

Keywords

Spot blotch · *Bipolaris sorokiniana* · Host resistance · QTLs · GWAS

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13.1 Introduction

Wheat (*Triticum aestivum*) is one of the important staple food crops of the world occupying more cultivated land than any other crop (Maulana et al. 2018). For each degree rise in temperature, wheat yields are predicted to decline by 4.1–4% due to climate change (Liu et al. 2016). Wheat production faces several challenges due to increasing population pressure, future food security, changing climatic conditions, and increasing food demands, and there is a need to increase global grain yields by 2–3% annually. The Indo-Gangetic Plain (IGP) wheat-growing regions are experiencing extreme and unpredictable weather conditions due to erratic fluctuations in climate. There are a number of diseases affecting wheat crop, but from the last four decades, this disease has been a serious constraint in wheat influencing the production, not only in the Eastern Plains of northern India but also in Nepal, Bangladesh, Brazil, and other countries. The disease has become more important in certain growing regions having warm, humid climatic conditions across wheat growing areas. After green revolution, this disease gained importance due to the cultivation of semi-dwarf varieties covering most of the areas and susceptibility to this disease.

Spot blotch caused by a fungus pathogen *Bipolaris sorokiniana* mainly affects crops in areas experiencing warm and humid environments of Latin America. The similar conditions also occur in eastern regions of India having warm and humid climate and in the adjoining countries like Bangladesh and Nepal. The disease is also known to affect wheat crop in Thailand, the Philippines, Indonesia, and the high rainfall and the warmer wheat-growing area of China (Van Ginkel and Rajaram 1998). The wheat production is severely affected by relatively high temperature besides high spot blotch pressure in these areas.

The spot blotch infection severity increases when the crop is at late post-anthesis stage and coincides with a spell of higher relative humidity and temperature (Gupta et al. 2018). However, critical monitoring and survey of the disease in the Indian EGP along with collection of infected crop samples at different crop stages suggested that the pathogen is hemibiotroph *B. sorokiniana* (syn. *Drechslera prorokiniana* syn. *Helminthosporium sativum*, *Cochliobolus sativus*) which is also a causal agent of common root rot, seedling blight, head blight, and black point diseases of wheat and barley. Around 25 million hectares of area under wheat is affected globally by spot blotch (van Ginkel & Rajaram 1998), of which about 40% of the area is in India (Joshi et al. 2007a), where the crop losses due to spot blotch have been estimated to be in the range of 15–25% (Dubin and Van Ginkel 1991a, b). The yield loss in severely infected fields is sometimes much higher as this disease not only affects leaves but also affects post-harvest quality of wheat grains (Mehta 1998). Thus, even partial reduction in disease infection would have a considerable impact on the income of farmers. Being a hemibiotrophic pathogen, achieving complete resistance is not possible. Complete resistance approach is also not advisable and practical as this leads to breakdown of resistance as experienced in case of Southern corn blight and wheat stem and stripe rusts (Jindal et al. 2012).

In the Indian subcontinent, rice-wheat cropping system alone constitutes 9 mha of affected area of the total 10 mha infected land (Nagarajan and Kumar 1998). The rice-wheat cropping system offers conducive conditions for the survival and multiplication of foliar blight pathogens as rice acts as a host for the spot blotch fungus, and after harvest rice stubble serves as a substrate for the fungi (Saari 1998). Host resistance against this pathogen is low (Agarwal et al. 2004). However, several donors have been identified in the breeding program for the improvement for spot blotch resistance in wheat, namely, BH 114, Yangmai 6, Mon/Ald, Ning 8201, and Chirya 3. Moreover, the molecular markers linked with resistance genes/QTLs may further be useful for developing breeding strategies (Kumar et al. 2020).

13.2 Pathogen Distribution and Host Range

Spot blotch disease is important in wheat-growing regions having warm and humid climate. *B. sorokiniana* attacks a large number of species in the Gramineae family (Sprague 1950) and a few dicotyledonous species thereby having wide distribution (Spurr Jr and Kiesling 1961) and wide host range. Spot blotch is not only limited to India, but it occurs in other wheat growing regions of the world also, particularly in South East Asia and Latin America (Joshi et al. 2007b, Nagarajan and Kumar 1998). This disease is widespread in specific areas where it is most prevalent including African Asian, European, and South American countries. *Bipolaris sorokiniana* is a fungal pathogen infecting a wide range of hosts (Neupane et al. 2010) often infecting a large number of grasses including bread wheat, durum wheat, triticale, rye, maize, *Phalaris minor*, *Lacromani anum*, *Phleum pratense*, *Setaria italica*, barley, and wild grasses (Manamgoda et al. 2011). The pathogen may rarely attack dicotyledonous plants in the field. *Bipolaris sorokiniana* was isolated from leaf lesions in a field of Michelite beans (Spurr Jr and Kiesling 1961). In addition, Spurr Jr and Kiesling (1961) found that bean, cowpea, cucurbits, pea, sunflower, and tomato plants can be parasitized by *B. sorokiniana* in the greenhouse.

13.3 Pathogen Variability

Morphological and pathological variability was reported in the isolates of *Bipolaris sorokiniana* (Nelson and Kline 1961; Misra 1976; Maraite et al. 1998) while the evolution of pathogen toward more aggressiveness was confirmed by Maraite (1998). The virulence on wheat and barley varied with the differences in pathogen isolates (Christensen 1926). Morphologically, virulence is correlated with the groups and the most likely cause of large-scale epidemics (Chand et al. 2003; Asad et al. 2009). The morphological variation in the pathogen population could be utilized in monitoring the pathogen population if associated with pathological variability. The pathogen variability with respect to aggressiveness between different groups of spot blotch isolates was studied by Kumar et al. (2007). RAPD markers were also used to identify strains/races to analyze virulence variability (Malvic and Grau 2001).

Aggarwal et al. (2010) differentiated 40 different isolates of *Bipolaris sorokiniana* collected from different locations in India and divided them into three clusters where some isolates revealed <50% similarity. Intra-specific variability among *Bipolaris* populations was studied by Oliveira et al. (2002) to examine the host-pathogen relationship. Variation in pathogenicity level under different environmental conditions of the individual pathotype has also been recorded from Pakistan and Nepal (Mahto et al. 2002; Asad et al. 2009). Further, virulence level also depends on hyphal fusion, nuclear migration, and occurrence of a multinuclear state (Chand et al. 2003; Pandey et al. 2008).

13.4 Symptoms

The symptoms of *B. sorokiniana* infection vary with the wheat genotype and growth stage, the isolate of the pathogen, and the environmental conditions (Kiesling 1985). The spot blotch pathogen infects and produces symptoms on leaf, sheath, node, and glumes (Chand et al. 2003) at all stages of plant growth and development. When conidial spores germinate on leaf and form germ tube, the leaf lesions enlarge in size and form large necrotic spots (Acharya et al. 2011). Symptoms first appear as small brown spots on the leaves that enlarge into elliptical, uniformly dark brown blotches with distinct yellow halos but may later coalesce into irregular dark brown necrotic areas (Dickson 1956). The spots are usually restricted in width by leaf veins; however, in some cases, lesions may continue to enlarge to form blotches that cover larger areas of leaves (Mathre 1997). The infection generally initiates in the lower leaves and gradually moves upward. In most cases, the spikes are also affected and display black point on seeds (Kumar et al. 2002). The occurrence and spread of the disease are also influenced by prevailing environmental conditions and crop management practices (Joshi et al. 2007a, b, c). The most common characteristic symptom is the production of a dark brown color in the lesions (Kiesling 1985). Older spot blotch lesions often appear as olive black, due to sporulation of the fungus (Mathre 1997). Lesions closely resemble the spotted form of net blotch. Lesions may extend in length on the leaf blade, but they do not become long, narrow streaks as in net blotch (Bailey et al. 2003). Depending on host response (resistance or susceptibility), pathogen virulence, and environmental factors, lesion size may vary from minute to small necrotic lesions (0.3–0.7 mm in length and 0.3–0.5 mm in width) with no or slightly diffuse marginal chlorosis, indicative of low compatibility, to large necrotic lesions (4.0–8.0 mm in length and 1.4–3.2 mm in width) with specific chlorotic margins (ranging from 0.5 to 1.0 mm in width) indicative of high compatibility (Fetch and Steffenson 1999). Dark spots may also appear on the leaf sheaths, necks, and heads of the plants. Lesions on the stalk below the head, especially at the nodes, can result in "neck break" (Bailey et al. 2003). Early floral infections cause aborted embryos or severely shrivelled grains (Anderson and Banttar 1976). The grain blight phase of the disease is referred to as "black point" or "kernel blight" and may develop if inoculum is abundant following heading, and environmental conditions are conducive to infection (Mathre 1997). The dark brown areas that

develop on lemmas of infected grains are usually found at the basal end (Anderson and Banttar 1976). With the adoption of dwarf and semi-dwarf wheat varieties along with the changing climatic conditions and farm management practices, the incidence of spot blotch is becoming frequent in the main wheat-producing areas, particularly in South America and Asia (Singh et al. 2016; Gupta et al. 2018).

13.5 Disease Scoring/Phenotyping for Spot Blotch

The recording of spot blotch infection is done on a continuous scale using the methods described by Duveiller et al. (1998) and Bashyal et al. (2010). The single-digit scale with scores ranging from 0 (immune) to 9 (highly susceptible) is adopted for disease scoring as described by Saari and Prescott (1975), whereas the double-digit scale (00–99) is modified from Saari and Prescott's scale for assessing severity of foliar diseases of wheat. The first digit ($D1$) indicates advancement of disease in canopy height from the soil level while the second digit ($D2$) refers to the leaf area affected by the disease (Eyal et al. 1987). The double-digit scale of spot blotch evaluation has been widely adopted. Visual scoring is done for each entry/genotype using a double-digit scale (00–99) developed as a modification of Saari and Prescott's severity scale (Saari and Prescott 1975). Both $D1$ and $D2$ are recorded on a scale of 1–9. For each score, the percentage of disease severity is estimated based on the following formula:

$$\text{Disease severity } (\%) = (D_1/9) \times (D_2/9) \times 100$$

For efficient and effective evaluation of resistance, it is often necessary to record several observations per plot at 3–7 days interval over a period of 3–4 weeks from anthesis and the dough stage, depending upon the planting date (Duveiller and Sharma 2009). The area under the disease progress curve (AUDPC) is calculated using the percentage disease severity estimates corresponding to three to four recordings as shown below.

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

where X_i = disease severity on the ith date, t_i = ith day, and n = number of times on which the disease is recorded. AUDPC (%/day) measures the level of the disease as well as disease progress rate.

Singh and Kumar (2005) suggested on a new double-digit (0–9) scoring method based on percent leaf area covered due to blight in case of flag and penultimate leaf to flag leaf (F) at different growth stages (GS) on Zadoks scale (Zadoks et al. 1974). The first digit ($D1$) indicates the severity of blight on flag leaf (F), whereas the second digit ($D2$) represents the percent blighted area of flag-1 leaf (F-1). The disease evaluation is generally carried out from anthesis up to late dough (GS87)

stages. Based on disease score, the entries are classified as immune (00), resistant (01–23), moderately resistant (34–45), moderately susceptible (56–68), susceptible (78–89), and highly susceptible (>89). The clear distinction between resistant and susceptible genotypes can be made at late dough stage, and it is suggested that data at late dough stage should be used for ultimate classification of resistance. Multilocation data can be categorized by taking the average (by taking both digits separately) and highest score over locations/years.

13.6 Genetics of Spot Blotch Resistance

Spot blotch is a disease of warm and humid regions of the world causing considerable losses in yield (Gupta et al. 2018). The most economical and eco-friendly approach to contain this disease is the deployment of host resistance to develop improved resistant cultivars. A good understanding of the genetics of resistance is a must to improve the resistance in cultivars (Eshghi and Akhundova 2009; Zaazaa et al. 2012). The inheritance of this disease is governed both by major and minor genes. Earlier studies (Srivastava et al. 1971; Srivastava 1982; Adlakha et al. 1984) reported monogenic control but later on studies also indicated polygenic inheritance (Velazquez Cruz 1994; Joshi et al. 2004). Dubin and Van Ginkel (1991a, b), Duveiller and Gilchrist (1994), and Dubin and Rajaram (1996) suggested that spot blotch resistance is governed by several genes having additive affect. Velazquez Cruz (1994) identified segregation for >4 genes in moderately resistant to resistant lines (Gisuz, Cugap, Chirya1, and Sabuf). Dominant and major gene controlling resistance is reported by Neupane et al. (2007), whereas both dominant and recessive genes controlling resistance were reported by Duveiller and Sharma (2009). Similarly, Sharma and Bhatta (1999) characterized three dominant genes having epistatic effect, involved in the genetic control of disease. Few reports suggested partially dominant genes controlling the resistance, and resistance was quantitatively inherited (Sharma et al. 2006). In a field study in Mexico, Velazquez reported that spot blotch resistance was governed by two to three partially dominant genes. Additive gene controlling resistance to spot blotch in accession number 8226, Mon/Ald, Suzhoe8 was reported by Joshi et al. (2004). Likewise, Bhushan et al. (2002) reported recessive genes with additive effect controlling resistance in cultivars PBW343 and HS361 and three genes in RAJ3702. A single dominant gene *Sb3* controlling blight resistance in genotype 621–7-1 was reported by Lu et al. (2016). Similarly another gene *Sb2* conferring resistance to spot blotch was reported by Kumar et al. (2015) in the YS116 wheat line. Lillemo et al. (2013) mapped the Sb1 gene for resistance to spot blotch on chromosome 7DS in the wheat line "Saar." Several QTL mapping studies have reported QTLs for resistance to blotch disease on 7D and 5B (Kumar et al. 2005); 2A, 2B, 5B, and 6D (Kumar et al. 2009); and 2AS, 2BS, 5BS, and 7DS (Kumar et al. 2010). Collectively based on the genetics of resistance in all these studies, spot blotch resistance is quantitatively controlled which also got confirmed from molecular studies involving QTL and genome-wide association studies (Cheruiyot et al. 2014).

13.7 Spot Blotch Resistance in Wheat

Resistance against spot blotch exists within the primary cultivated gene pool and also in related wild species from within the tribe Triticeae constituting the secondary and tertiary gene pool.

13.7.1 Resistance in the Cultivated Germplasm

The earliest record on wheat varietal resistance to spot blotch was reported by Nima and Joshi (1973) who found "Sonora 64" and "NP884" more tolerant to spot blotch as compared to other genotypes. Srivastava et al. (1971) also reported wheat varieties resistant to spot blotch in India. However, the major effort on screening wheat for resistance to spot blotch happened in the 1980s when spot blotch attained the status of an important disease in warm and humid wheat-growing regions (Duveiller and Gilchrist 1994). At CIMMYT, Mexico, wheat genotypes Yangmai 6, M3 (W7976), Shanghai 4, and Chirya7 were developed, using germplasm from China which possessed good level of resistance (Ibeagha et al. 2005). To date, the best sources of resistance were discovered in the Brazilian and Zambian along with Chinese sources (Rajaram 1988; Dubin and Van Ginkel 1991a, b; Kohli et al. 1991). Duveiller and Sharma (2005) identified Milan/Shanghai #7 being the most resistant and good yielding genotype. Other studies have confirmed that Milan/Shanghai #7 and Chirya 3 are highly resistant to spot blotch (Duveiller et al. 2005; Joshi et al. 2004; Ragiba et al. 2004). Kumari et al. (2018) evaluated a large collection of wheat germplasm (1483) and identified seven genotypes (IC564121, IC529684, IC443669, IC443652, IC529962, IC548325, and EC178071-331) highly resistant to spot blotch. Choudhary et al. (2019) identified genotypes Chirya 7, Chirya 3, Ning 8139, Suzhou, Milan-3, HD 2888, HD 2967, and WR 95 as resistant at seedling stage, whereas genotypes Chirya 7, Chirya 3, Ning 8139, Suzhou, Milan-3, HD 2888, HD 2967, WR 95, and HW 3081 are resistant at adult plant stage. The identified sources along with their country of origin are presented in Table 13.1.

13.7.2 Alien Sources of Resistance

Wild species from the secondary gene pool have been utilized in breeding for spot blotch resistance during the late 1980s in CIMMYT. Initially *Thinopyrum curvifolium* was used for transferring resistance (Duveiller & Gilchrist 1994) along with some germplasm from China; resistant genotypes Mayoor and Chirya were developed. Apart from that, *Aegilops squarrosa* crosses were identified to be showing good resistance to spot blotch in Mexico. About 14,000 lines of wheat and related alien species, representing different genera and species assessed for spot blotch resistance at PAU, Ludhiana (Dhaliwal et al. 1993; Singh and Dhaliwal 1993), and resistant entries including *Ae. triuncialis*, *Ae. speltoides*, *Ae. squarrosa*, *Ae. triaristata*, *Triticum dicoccoides*, *Ae. cylindrica*, and *T. boeoticum* have been

Table 13.1 Sources of spot blotch resistance identified around the world

Genotypes	Country	References
BAW 969, BAW 1006, BAW 1008 Sharma et al. (2004a), Siddique et al. (2006)	Bangladesh	Sharma et al. (2004a); Siddique et al. (2006)
BH 1146, CEP 14, CNT 1, Ocepar 7, Trigo BR 8	Brazil	Mehta (1998); Sharma et al. (2004b, 2007b); Caierao et al. (2014)
Chuanmai 18, Fang 60, G162, Jinmai 4058, Longmai 10, Longmai 10370, Ning 8201, Ning 8319, Quangfeng, Shanghai #4, Shanghai #158, Suzhoe #1–58, Suzhoe #8, Suzhoe #128-OY, Yangmai 6	China	Sharma et al. (1997a, b, 2004b); van Ginkel and Rajaram (1998); Joshi et al. (2004a, 2007d); Ibeagha et al. (2005); Sharma and Duveiller (2007); Kumar et al. (2009, 2010)
Attila = NL781 = PBW343, BOW 'S', M3, Chirya 1, Chirya 3, Chirya 7, Chukui #1, Cigm 90.455, FFN/VEE #5, HLB25, Kauz/Vee/Muna, Milan/ Shanghai #7, SM-4-HSN24, Vayi #1, Afghan collection	CIMMYT, Mexico	Chaurasia et al. (1999); Sharma et al. (2004a, b, c); Ragiba et al. (2004); Duveiller et al. (2005); Ibeagha et al. (2005); Joshi et al. (2007a, b, c); Neupane et al. (2007); Sharma and Duveiller (2007); Kumar et al. (2009, 2010); Singh et al. (2015); Bainsla et al. (2020)
ACC 8226, BW 14999, CPAN 3003, CPAN 3048, CPAN 4006, CPAN 4007, CPAN 4011, CPAN 4042, CPAN 4065, CPAN 4070, HD 2662, HD 2819, HP 1729, HP 1808, HUW234, HUW206, HUW289, HUW302, HUW305, HUW323, HUW325, HW 2093, K 9107, M3109, PBW 343, PBW 486, RAJ 3702, Triveni, WH542, YS116 (Yangmai 6/Sonalika)	India	Chaurasia et al. (1999); Joshi and Chand (2002); Joshi and Chand (2002); Joshi et al. (2004a); Sharma et al. (2004a, b); Sharma and Duveiller (2007); Singh and Singh (2009); Khan and Chowdhury (2011); Kumar et al. (2015, 2016)
Achyut, Bhrikuti, BL1693, BL1724, BL1740, BL1813, BL1883, BL2069, BL2127, BL3704, BL4148, Gautam, Mayoor, NL835, NL868, NL872, WK 1204	Nepal	Sharma et al. (2004a); Sharma and Duveiller (2006); Joshi et al. (2007b); Mahto et al. (2011)
Abadgar 93, Anmal 91, Auqab 2000, Bahawalpur 2000, Bahkhar 2002, Bakhtawar 92, Darawar 97, Faisalabad 85, Inqilab 91, Iqbal 2000, Kaghan 93, Kirin 95, Kohistan 97, Kohsar 95, Magalla 99, Mexi Pak, Moomal 2002, Nowshera 96, Parwaz 94, Pasban 90, Pirsabak 2005, Punjab 96, Saleem 2000, Sariab 92, SH 2002, Shafaq 2006, Shaheen 94, Shahkar 95, Soughat 90, Wafaq 01, Watan 94	Pakistan	Iftikhar et al. (2012)
K 7, 30SAWSN5, and Coucal	Zambia	Sharma et al. (2004b); Batiseba et al. (2017)

identified. Alien sources include *Thynosporium curvifolium* and *Aegilops squarrosa*. Transfer of resistance from alien species (*Thinopyrum curvifolium*, *Elymus curvifolius*, and *T. tauschii*) to common bread wheat was also reported (Mujeeb-Kazi et al. 1996). Availability of resistance was reported in *T. timopheevii*, *T. araticum*, *T. boeoticum*, *T. persicum*, and *T. urartu* as well as in *T. sphaerococcum* (Smurova and Mikhailova 2007).

13.8 Breeding for Spot Blotch Resistance

Efforts have been made to effectively manage the disease, but no single effective control measure has been able to control the disease. Breeding for disease resistance is an eco-friendly and cost-effective means of managing spot blotch. However, it is important to understand the genetics of resistant genes and also to identify resistant genes responsible for SB resistance. The available literature suggests the trait is under the control of quantitative genes. The quantitative nature of resistance slows the progress in breeding for resistance because of low heritability.

Initially the efforts were made to identify new resistant germplasm involved in screening of wheat genotypes from Brazil, Zambia, and the Yangtze River Valley in China, and many lines were identified with satisfactory levels of resistance to spot blotch (Raemaekers 1991; Dubin and Rajaram 1996; Mehta 1998; van Ginkel and Rajaram 1998). These lines were widely used in CIMMYT's wheat breeding programs and were tested in international nurseries in many countries (Dubin et al. 1998). Mujeeb-Kazi et al. (1996) reported a number of lines from CIMMYT's wide crosses which were resistant to spot blotch. These initial sources of resistance were extensively tested in warm wheat-growing regions in international, regional, and national disease nurseries in the subsequent years. Based on data from regional trials, Dubin et al. (1998) recommended several wheat genotypes with good levels of spot blotch resistance.

Additional sources of resistance were reported in South Asia (Sharma et al. 2004a, b, c, Sharma and Duveiller 2007) and India (Singh et al. 1998; Joshi et al. 2004b). These resistance sources were used extensively, and resulting new varieties with higher levels of resistance than older varieties were selected (Sharma et al. 2004a, b, c; Siddique et al. 2006). Whereas international collaboration contributed to the development of wheat genotypes with improved spot blotch resistance, high grain yield, and acceptable agronomic traits (Sharma and Duveiller 2007), the sources with high level of resistance seem limited (Duveiller and Sharma 2009). From the comparison of older susceptible varieties to newly released relatively tolerant cultivars, it appears that a good deal of success has been achieved toward improving spot tolerance in South Asia (Duveiller and Sharma 2009). However, the level of resistance in the newly wheat cultivars represents only a partial success in improving resistance against spot blotch, and the disease remains a serious concern (Sharma and Duveiller 2006). As a result, many high yielding lines and spot blotch-resistant lines were identified and shared with centers across zones in India (Gyanendra et al. 2007). Besides, six new genetic stocks (LBRL 1, LBRL

4, LBRL 6, LBRL 11, LBRL 13, and DBW 46) possessing high level of leaf blight resistance in improved background have been developed and registered for use by the breeders across countries. In South Asia, moderate success in breeding for spot blotch and foliar blight resistance has been reported (Bhandari et al. 2003; Sharma et al. 2004c; Joshi et al. 2004a; Siddique et al. 2006; Gyanendra et al. 2007; Manoj 2013). In Zambia, germplasm exchange led to the release of resistant varieties in rainfed wheat production environments, e.g., PF7748 in Whydah and Hombill (= IAS64/Aldan). PF73339/Hahn, a CIMMYT material (Raemaekers 1987), has led to the increase of yield potential from 1.6–1.7 to 2.7 t ha^{-1} (Mukwavi 1995). Dubin and Rajaram (1996) suggested to practice selection in later generations to combine genes controlling minor resistance in segregating populations. Joshi and Chand (2002) suggested that genes for resistance must be combined with genes controlling erect leaf trait for better control of disease. The difficulty of improving resistance to spot blotch through conventional selection may be due to the limited effectiveness of the prevalent selection technique to identify multiple genes controlling resistance (Sharma and Bhatta 1999; Bhushan et al. 2002; Joshi et al. 2004a; Ragiba et al. 2004) under field conditions. Hence, the identification of molecular markers linked to spot blotch resistance could speed up breeding to improve resistance.

13.9 QTL Mapping

During the past few years, efforts have been made to identify the genes/QTLs involved with spot blotch resistance. Several QTLs responsible for SB resistance in wheat have been mapped (Table 13.2). With a cross from Chinese resistant cultivar Yangmai 6 and Sonalika (susceptible), four QTLs (*QSb.bhu-2A*, *QSb.bhu-2B*, *QSb.bhu-5B*, *QSb.bhu-6D*) have been identified for spot blotch resistance explained 8.04–41.10% of phenotypic variation, QTLs on chromosomes 2B and 5B with major effects (Kumar et al. 2009). Moreover, Kumar et al. (2010) further identified major QTLs on chromosome 2B and 7D in two mapping populations, viz., Ning 8201 × Sonalika and Chirya 3 × Sonalika, and validated the diagnostic markers for future breeding programs. Another report of Lillemo et al. (2013) determines the potential association of genes *Lr34* (7DS) and *Lr46* (1BL) with spot blotch resistance QTLs, *Lr34* gene explained up to 55% phenotypic variation for spot blotch disease resistance, and this locus was given the gene designation *Sb1*. In a CIMMYT synthetic wheat-derived line SYN1 mapping population, Zhu et al. (2014) reported three QTLs, namely, *QSb.cim-1B* (PVE-8.5%), *QSb.cim-3B* (PVE-17.6%), and *QSb.cim-5A* (PVE-12.3%), for spot blotch resistance. Furthermore, *QSb.bhu-5B*, which determines resistance to spot blotch, was mapped to an interval of 0.62 cM on chromosome arm 5BL; any of these SSR markers *Xgwm639* or *Xgwm1043* are linked closely to *Sb2* to be used as an indirect selection tool for spot blotch resistance (Kumar et al. 2015). Kumar et al. (2016) evaluated 19,460 wheat accessions for rust and spot blotch disease resistance and identified different combinations of genetic loci imparting resistance to rust and spot blotch using linked molecular markers. Addition to these, in two bi-parental mapping population, Singh

Table 13.2 QTL mapping studies for spot blotch resistance

Material	Markers	Chr	QTL designation	Flanking markers	PVE	Resistance allele	References
Sonalika × G162 (18 wheat genotypes and 16 F7 progeny)	SSR	5B		*Xgwm67*			Sharma et al. (2007a)
		6A		*Xgwm570*			
		6D		*Xgwm469*			
Yangmai 6 × Sonalika (139 RILs)	SSR	2AL	*QSb.bhu-2A*	*Xbarc353-Xgwm445*	14.8	Yangmai 6	Kumar et al. (2009)
		2BS	*QSb.bhu-2B,*	*Xgwm148-Xgwm374*	20.5	Yangmai 6	
		5BL	*QSb.bhu-5B*	*Xgwm067-Xgwm371*	38.62	Yangmai 6	
		6DL	*QSb.bhu-6D*	*Xbarc175-Xgwm732*	22.5	Yangmai 6	
Ning 8201 × Sonalika (103 RILs)	SSR	2AS	*QSb.bhu-2A*	*Xgwm425-Xbarc159*	15.2	Ning 8201	Kumar et al. (2010)
		2BS	*QSb.bhu-2B*	*Xgwm148-Xbarc91*	23.7	Ning 8201	
		5BL	*QSb.bhu-5B*	*Xgwm067-Xgwm213*	10.7	Ning 8201	
		7DS	*QSb.bhu-7D*	*Xgwm111-Xgwm1168*	39.2	Ning 8201	
Chirya 3 × Sonalika (128 RILs)	SSR	2BS	*QSb.bhu-2B*	*Xgwm148-Xgwm129*	13.1	Chirya 3	
		2DS	*QSb.bhu-2D*	*Xgwm455-Xgwm815*	10.7	Chirya 3	
		3BS	*QSb.bhu-3B*	*Xgwm533-Xgwm1037*	9.7	Chirya 3	
		7BS	*QSb.bhu-7B*	*Xgwm263-Xgwm255*	10.2	Chirya 3	

(continued)

Table 13.2 (continued)

Material	Markers	Chr	QTL designation	Flanking markers	PVE	Resistance allele	References
		7DS	*QSb.bhu-7D*	*Xgwm111-Xswm008*	11.9	Chirya 3	
Avocet (susceptible) × Saar (114 RILs)	CAPS and PCR marker	7DS	*Lr34*	*wPt-7654-gdm88*	37.1	Saar	Lillemo et al. (2013)
		1BL	*Lr46*	*hbe248-ncw1-V*	15.1	Saar	
		7DS		gwm1220-swm10	55.2	Saar	
SYN1 × Ocoroni (161 DH lines)	GBS and SSR	3B	*QSb.cim-3B*	*990,937|F|0–1,123,330|F|0*		SYN1	Zhu et al. (2014)
		5A	*QSb.cim-5A*	*1,086,218|F|0–982,608|F|0*		SYN1	
		1A	*QSb.cim-1A*	*1,026,215|F|0–1,088,769|F|0*		Ocoroni	
		1B	*QSb.cim-1B*	*Xwmc128 and Xgwm374*		SYN1	
YS116 × Sonalika (335 RILs)	SSR	5BL	*QSb.bhu-5B*	*Xgwm639*		YS116	Kumar et al. (2015)
		5BL	*QSb.bhu-5B,*	*Xgwm1043*		YS117	
BH 1146 × Sonalika (209 RILs)	SSR	7B	*QSb.iiwbr-7B*	*wmc758-wmc335*	11.4	BH 1146	Singh et al. (2016)
		7D	*QSb.iiwbr-7D*	*wmc653-barc121*	14.7	BH 1146	
BARTAI × CIANO T79 (232 RILs)	DArTSeq (GBS) platform	1B		*995,296–5,410,777*	6.5	CIANO T79	Singh et al. (2018)
		1B		*5,324,988–1,110,815*	8.9	BARTAI	
		1D		*100,142,243–1,037,975*	2.9	CIANO T79	

		3A		*1,109,808–990,692*	3.2	CIANO T79	
		4B		*1,247,021–1,702,494*	3.3	BARTAI	
		4D		*BS00036421_51–1,119,387*	–	BARTAI	
		5A		*Vrn-A1–3,064,415*	12.5	BARTAI	
		6D		*1,239,681–1,095,962*	–	BARTAI	
WUYA × CIANO T79 (232 RILs)	DArTSeq (GBS) platform	1B		*1,283,166–2,263,671*	8.3	WUYA	
		2D		*1,085,831–1,098,973*	9	WUYA	
		4B		*Rht-B1-1,241,652*	2.3	CIANO T79	
		4B		*1,217,659–997,944*	4.5	WUYA	
		5A		*1,135,154–2,260,918*	25.1	WUYA	

et al. (2018) identified the most outstanding minor quantitative trait locus (QTL) for spot blotch resistance with strong influence from *Vrn-A1* in both populations on chromosome 5AL.

13.10 Identification of Genomic Regions Controlling Spot Blotch Resistance Through GWAS

With the advent of new genomic technologies such as next-generation sequencing approaches, SNP chip, and genotyping by sequencing (GBS), more precise mapping methodologies like genome-wide association studies (GWAS) have gained importance for studying several complex traits such as spot blotch across a wide range of environment (Ayana et al. 2018). GWAS has been used to characterize disease resistance in wheat: SB resistance in wild barley (Roy et al. 2010), resistance to multiple leaf spot diseases of spring wheat (Gurung et al. 2014), resistance to bacterial leaf streak and SB in spring wheat (Adhikari et al. 2012), *Fusarium* head blight resistance in wheat (Arruda et al. 2016), tan spot resistance in European winter wheat (Kollers et al. 2014), and mapping for resistance to leaf and stripe rust in winter-habit hexaploid wheat landraces (Sun et al. 2015). Many studies, using methods of both bi-parental mapping and association mapping (AM), have reported several SB resistance QTLs on chromosomes 1A, 1B, 2A, 2B, 2D, 3B, 5A, 5B, 6B, 6D, 7A, 7B, and 7D (Neupane et al. 2007; Sharma et al. 2007a; Gonzalez-Hernandez et al. 2009; Kumar et al. 2009, 2010, 2015, 2016; Adhikari et al. 2012; Lillemo et al. 2013; Gurung et al. 2014; Zhu et al. 2014; Lu et al. 2016; Zhang et al. 2015; Gupta et al. 2018). Several association studies are available to discover putative QTLs to study the genetics of spot blotch resistance and discover SNP markers beneficial for MAS (Table 13.3). Using association mapping (AM) with 832 polymorphic Diversity Arrays Technology (DArT) markers, Adhikari et al. (2012) identified four genomic regions with wPt-1159 on 3B significantly associated with resistance to SB. Gurung et al. (2014) identified nine associated SNPs that were located on five chromosomes (1B, 5A, 5B, 6B, 7B) for SB resistance using genotypes from diverse geographic origin. Ayana et al. (2018) identified ten winter wheat genotypes resistant to SB and six genomic areas associated with SB resistance in conjunction with tightly linked SNPs. SB resistance locus on wheat chromosomes 2D, 3A, 5A, and 7B identified in this study is syntenic to the previously identified SB resistance locus on chromosomes 2H, 3H, 5H, and 7H in barley. Further in an association study comprising 301 Afghanistan genotype panel, 19 significant SNPs associated with resistance to SB were detected; the most significant SNP was on chromosome 5A (5411867) (Bainsla et al. 2020). Recently, researchers validated stable genomic region for spot blotch resistance on chromosomes 2B, 5B, and 7D in a 141 diverse wheat panel and identified a new genomic region on chromosome 3D associated with zinc finger protein that plays an important role in plant disease resistance (Tomar et al. 2020). In addition, they also conducted functional annotation with wheat genome assembly annotation (IWGSC Ref Seq v1.0) and identified NBS-LRR and 35 other plant defense-related protein families across multiple

Table 13.3 Genome-wide association study for spot blotch resistance in wheat

Material	Markers	Significant SNP	Chr	Position	R^2	References
528 diverse geographic origin	9 K wheat single nucleotide polymorphism	wsnp_Ex_c24700_33953160	1B	37.18 cM		Gurung et al. (2014)
		wsnp_JD_c8926_9848514	1B	37.2 cM		
		wsnp_Ex_c15342_23592740	5A	76.5 cM		
		wsnp_Ku_c17951_27138894	5A	76.5 cM		
		wsnp_Ex_rep_c70120_69069789	5B	109.52 cM		
		wsnp_ku_c50354_55979952	5B	146.88 cM		
		wsnp_ku_c20701_30355248	5B	147.03 cM		
		wsnp_Ex_c15785_24157360	6B	90.3 cM		
		wsnp_Ex_c52527_56097039	7B	56.8 cM		
294 accessions of hard winter wheat association mapping panel (HWWAMP)	Illumina iSelect 90 K	Kukri_c31121_1460	2D	80.2 cM	4.3	Ayana et al. (2018)
		Excalibur_c46082_440	3A	90.6 cM	4	
		IWA8475	4A	118.7 cM	5.5	
		Excalibur_rep_c79414_306	4B	36.8 cM	4.1	
		Kukri_rep_c104877_2166	5A	59.1 cM	6.2	
		TA005844-0160	7B	86.4 cM	6.3	
287 genotypes of WAMI panel	90 K wheat SNP array	BobWhite_c35961_80	6A	79 cM	8.3	Ahirwar et al. (2018)
		BobWhite_c3661_88	1B	64 cM	8.3	
		tplb0027f13_1493	5B	90 cM	10.0	
		wsnp_Ku_c40334_48581010	5B	90 cM	9.9	
		BS00009311_51	5B	62 cM	9.0	
		Excalibur_c96134_182	6B	5 cM	8.7	
301 Afghanistan genotype panel	DArTSeq	4,909,825	1B	557,997,411 bp	8.8	Bainsla et al. (2020)
		1,085,203	3A	595,935,042 bp	17.6	
		1,128,070	3A	502,404,423 bp	2.2	
		1,220,348	3A	598,916,422 bp	13.2	
		7,354,241	3B	12,103,697 bp	8.8	

(continued)

Table 13.3 (continued)

Material	Markers	Significant SNP	Chr	Position	R^2	References
		991,620	4A	658,343,324 bp	12.3	
		100,177,527	5A	3,319,047 bp	17.6	
		5,411,867	5A	586,600,348 bp	17.6	
		6,036,625	5B	407,623,507 bp	8.0	
141 Spring wheat lines	GBS-based 18,637 polymorphic SNP markers	S1B_646,895,451	1B	646,895,451 bp	10.8	Tomar et al. (2020)
		S2A_31,851,904	2A	31,851,904 bp	12	
		S2B_504,717	2B	504,717 bp	12.7	
		S2B_525,073	2B	525,073 bp	12.7	
		S2B_594,959	2B	594,959 np	10.4	
		S2B_6,253,562	2B	6,253,562 bp	10	
		S2B_8,311,062	2B	8,311,062 bp	10.1	
		S2B_90,662,917	2B	90,662,917 bp	10.5	
		S3B_763,230,831	3B	763,230,831 bp	11.5	
		S3B_763,236,179	3B	763,236,179 bp	11.5	
		S3B_763,267,753	3B	763,267,753 bp	11.5	
		S3B_764,192,662	3B	764,192,662 bp	9.7	
		S4A_710,830,493	4A	710,830,493 bp	11.6	
		S6B_719,904,092	6B	719,904,092 bp	9.2	
		S7D_181,974,079	7D	181,974,079 bp	6.7	

chromosome regions. The genomic prediction model for spot blotch disease resistance in wheat was tested and obtained moderate prediction accuracy.

13.11 Fine Mapping of Spot Blotch QTLs

To date, only four designated spot blotch (*Sb*) resistance genes (*Sb1–Sb4*) have been identified and fine mapped in wheat (Lillemo et al. 2013; Kumar et al. 2015; Lu et al. 2016; Zhang et al. 2020). *Sb1* was mapped on chromosome 7DS and also shown to be co-located with the cloned leaf rust resistance locus *Lr34* having pleiotropic effects on stripe rust, stem rust, powdery mildew, and leaf tip necrosis (Lillemo et al. 2013). The major QTL on chromosome 5BL reported by Kumar et al. (2015) was designated as *Sb2* harboring a 0.62-cM region between *Xgwm639* and *Xgwm1043* SSR markers. The third gene *Sb3* was located within a 0.15-cM interval spanning 602 kb region of Chinese Spring chromosome 3BS (Lu et al. 2016). Recently, Zhang et al. (2020) identified *Sb4*, a new spot blotch resistance gene mapped on chromosome 4BL in an interval of 1.19 cM corresponding to a 1.34 Mb physical genomic region containing 21 predicted genes. A resistance like gene *Tsn1* on wheat chromosome arm 5BL is required for virulence gene *ToxA* sensitivity, conferring disease susceptibility to fungal pathogens harboring *ToxA* (Friesen et al. 2018). The study of Navathe et al. (2020) suggests that the absence of *Tsn1* facilitated resistance against spot blotch of wheat. Therefore, the selection of wheat genotypes for the absence of the *Tsn1* allele can improve resistance to spot blotch. Recently, Wu et al. (2020) reported *ToxA* occurrence in *B. sorokiniana* populations of Mexico.

13.12 Marker-Assisted Introgression of Spot Blotch Resistance

It is imperative to identify robust diagnostic markers/genes and validate these tightly linked markers in diverse set/mapping populations before applying them for introgression. In case of spot blotch, marker-assisted backcross breeding was implemented successfully in wheat to improve spot blotch resistance. Singh et al. (2014) reported five diagnostic molecular markers (*Xgwm371*, *Xgwm425*, *Xgwm445*, *Xbarc59*, and *Xbarc232*) for spot blotch resistance. With the aim of marker-assisted selection (MAS), Vasistha et al. (2016) conducted two parallel backcross programs—one targeted the locus *QSb.bhu-2A*, and the second one targeted on the 2 loci *Qsb.bhu-2A* and *Qsb.bhu-5B* so as to transfer resistance to spot blotch within the susceptible cultivar HUW 234; hence, Chirya3 and Ning8201 were used as donor parent. The BC_3F_3 selection and those made in BC_3F_4 and BC_3F_5 showed enhanced resistance to spot blotch and also yielded better than the recipient parent in presence of the disease. Another study conducted by Vasistha et al. (2017) reported molecular introgression of leaf rust *Lr34* from CIMMYT breeding line Picaflor #1 into an Indian wheat cultivar HUW510 which validates enhanced effect on resistance to spot blotch and higher grain yield. These studies showed that stacking of known spot blotch QTLs/genes along with *Sb1* (*Lr34*),

Lr46, and *Vrn-A1* genes can be successfully introgressed into popular wheat cultivars leading to enhanced resistance to spot blotch disease. With the advent of new technologies such as high-throughput sequencing, phenomic technologies, and genome editing tools, the discovery of more number of robust QTLs/genes can be done and used to breed spot blotch-resistant cultivars.

13.13 Future Prospects

The resistance breeding targeting spot blotch, leaf rust, and wheat blast will gain attention of researchers to meet future targets of multiple disease resistance and also breeding for climate resilience. Utilizing information about known genes/QTLs/genomic region, markers for developing cassettes to introgress desirable traits/genes will be more commonly followed. Precision phenotyping platforms, use of AI tools, bioinformatics, and their synteny are likely to be futuristic approaches for resistance breeding in wheat to manage new and emerging threats amid changing climate.

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