Acta Scientiarum
http://www.periodicos.uem.br/ojs/
ISSN on-line: 1807-8621
Doi: 10.4025/actasciagron.v45i1.58054

Development of interspecific segregating populations of wheat and evaluation of agronomic characteristics and resistance to Fusarium head blight

Patrícia Frizon[1*], Sandra Patussi Brammer[2], Carolina Cardoso Deuner[1], Amanda Chequi[1], Maria Imaculada Pontes Moreira Lima[2] and Ricardo Lima de Castro[2]

[1]Programa de Pós-Graduação em Agronomia, Faculdade de Agronomia e Medicina Veterinária, Universidade de Passo Fundo, BR-285, 99052-900, Bairro São José, Passo Fundo, Rio Grande do Sul, Brazil. [2]Empresa Brasileira de Pesquisa Agropecuária, Embrapa Trigo, Passo Fundo, Rio Grande do Sul, Brazil. *Author for correspondence. E-mail: patriciafrizon@gmail.com

ABSTRACT. The objective of this study was to develop segregating wheat populations for resistance to gibberella through the introgression of synthetic wheat genes in traditional cultivars and to present the results using the backcross method. In addition, to evaluate these populations in F1RC2, agronomic aspects and type II and III resistance to Fusarium head blight (FHB). The cultivars BRS Guamirim and BRS 179 were used as male parents and three synthetic wheat cultivars (CIGM90.909, CIGM92.1666, and CIGM93.298) as female parents, both previously characterized for the reaction to FHB. Crossing and backcrossing methodological procedures were performed to provide adequate controlled conditions. The F_1 (winter 2016), RC_1 (summer 2016/2017), and RC_2 (winter 2017) populations were developed through emasculation and pollination procedures. In the winter of 2018, segregating populations were evaluated in the field using a randomized block design. In the three generations developed in a greenhouse, it was observed that the best crossbreeding combinations, verified by the number of grains, were obtained with the male parent BRS 179. Under field conditions, all segregating populations were considered moderately susceptible and susceptible to type II resistance. In type III resistance, all segregating populations, except for CIGM90.909/BRS 179 and BRS 179, showed values above 50%. Thus, it was evident that the crossings performed with the male parent BRS 179 generated better resistance to the disease.

Keywords: diseased grains; *Fusarium graminearum*; segregating populations; severity.

Received on March 3, 2021.
Accepted on July 17, 2021.

Introduction

Wheat is the second most cultivated cereal in the world after corn. In 2019, according to FAO data, approximately 1.15 billion tons of corn and 766 million tons of wheat were produced, with a market value of approximately US$ 114 million (Coêlho, 2021). Biotic stresses generate significant challenges to increasing productivity and have important implications for food security in many regions where wheat is the main food source, particularly in developing countries, where input costs are high (Jighly et al., 2016).

Fusarium head blight (FHB) (*Gibberella zeae* Schw.) is one of the main fungal diseases affecting wheat production. Of the 17 fusarium species that cause the disease worldwide, *F. graminearum* is the predominant cause of illness in many countries (Zhu et al., 2016). As a typical disease of warmer and humid climates (Brennan, Egan, Cooke, & Doohan, 2005), FHB reduces wheat yield and negatively affects milling, cooking, and pasta manufacturing (Bai & Shaner, 1994). In addition, the infected grains may contain mycotoxins such as deoxynivalenol (DON) that make grains unfit for human or animal consumption (Mcmullen, Jones, & Gallenberg, 1997). Genetic resistance is considered the most economical and environmentally friendly way to control the fungus and is one of the main objectives of many cereal breeding programs (Osman et al., 2015). However, the disease is highly complex because it is genetically quantitative and is controlled by many genes with a smaller effect. Mesterházy (1995) described five types of resistance: type I (resistance to initial infection), type II (resistance to spread on the spike), type III (grain resistance), type IV (resistance to mycotoxin accumulation), and type V (resistance expressed by yield). Resistances I, II, and III have been the most studied compared to IV and V.

Commercial cultivars are not good sources of resistance. Thus, breeding programs look for wild, closely related species as alternative sources of resistance to FHB. Hexaploid synthetic wheat has been reported as a repository of wide genetic diversity for the improvement of wheat (Ogbonnaya et al., 2013). This material is a sterile hybrid that originated from the hybridization of two species, the tetraploid *T. turgidum* (genomes AABB) and the wild diploid *Aegilops tauschii* (genome DD). Its fertility is restored in the laboratory with the use of colchicine for chromosome duplication (Van Ginkel & Ogbonnaya, 2007). Genetic diversity for resistance has been identified in synthetic wheat for a wide range of biotic stresses including FHB (Buerstmayr, Steiner, & Buerstmayr, 2020).

The selection of parents and segregating populations is the first step in the breeding process. This is one of the most important decisions to be taken by the breeder (Tekle et al., 2018) as the overall efficiency of the program depends on it. The backcrossing method has been used in genetic breeding programs aimed at the incorporation of resistance genes from wild species in varieties with commercial characteristics (Faleiro, Junqueira, & Braga, 2005).

Because of the high relevance of FHB on damage to productivity and quality of wheat, the present study had the following objectives: i) introgress genes from synthetic wheat in wheat cultivars to introduce FHB resistance in the backcrossing program and ii) to evaluate the segregating F_1RC_2 wheat populations originating from the crossing between synthetic wheat and commercial cultivars, focusing on the agronomic aspects and type II and type III resistance to FHB.

Material and methods

Experiment 1

Parents used

The material used comprised two commercial wheat cultivars male parents adapted to the conditions of Rio Grande do Sul, Brazil; they were chosen based on the divergence for several characteristics: grain production, days for maturation, and plant height. As female parents and sources of exotic resistance, three accessions of synthetic hexaploid wheat were obtained from CYMMIT (International Corn and Wheat Improvement Center, Mexico) and previously characterized as moderately resistant to FHB (Table 1).

Table 1. Genotypes used in the crossings aiming resistance to FHB.

Acession	Genealogy	Resistance
CIGM90.909	GAN/ *Ae. Squarrosa*	MR[1]
CIGM92.1666	RASCON// *Ae. Squarrosa*	MR
CIGM93.298	CIGM93.298	MR
BRS Guamirim	Embrapa 27/Buck Nandu//PF93159	MR
BRS 179	BR 35/PF 8596/3/PF 772003*2/PF 813//PF 83899	MR

[1]Moderately resistant.

Characterization of the experimental area

The experiment was performed in 2016 and 2017, in a greenhouse at Embrapa Trigo, in the city of Passo Fundo, Rio Grande do Sul State, Brazil, located at an altitude of 687 m, the latitude of 28°15' S, and longitude of 52°24' W.

Obtaining the F_1 wheat hybrids

The experiment was performed in a controlled conditions greenhouse. The parental sowing occurred at three different times within a week of each other, starting on June 26, 2016, to coincide with flowering, making it possible to accomplish the crossings and also as a precaution against any issue that may occur that could hinder the program execution. In each season, 12 buckets (dimensions of approximately 0.20 m in height and 0.20 m in diameter) were used for each parent, totaling 144, with five seeds per bucket sown. The buckets were arranged in rows spaced 0.40 m apart, filled with soil corrected for pH, and fertilized properly. The following operations were performed to accomplish the crossings:

i) Emasculation: The anthers were removed from the flowers, keeping only the female part. It occurred at the end of the boot phase, that is, at the 10.1 stage of the Feeks-Large scale (Large, 1954).

ii) Pollination: This was performed three to five days after emasculation. The bag that surrounded the emasculated spike was cut at the top, and then the spike with the mature anthers was introduced, stirring so that the pollen grains were released, pollinating the stigmas. The paper bags were then closed and stapled, placing a label indicating the crossing and its date. In this procedure, several spikes were used to obtain the seeds required for the work.

iii) Harvesting hybrid seeds in generation F_1: At the time of maturation, the spikes that were bagged and properly identified from the three crossings were harvested and dried under environmental conditions, and then threshed manually. The grains were stored under adequate environmental conditions for the breeding program.

Obtaining the F_1RC_1 and F_1RC_2 backcrossings

The seeds of the wheat parents and part of the seeds of the hybrids in the F_1 generation were sown in December 2016 in plastic buckets in the same greenhouse. The parents were sown in three seasons, separated from each other ten days, and the hybrids in the F1 generation were also sown in three seasons to obtain the RC_1 backcrossing. This procedure was performed for the reasons mentioned above. In each season, the three parents were sown in 12 bucket each, totaling 36 buckets per season, with five seeds in each bucket. Using the seeds of the F_1 generation hybrids, 12 buckets were sown per hybrid, with five seeds each, totaling 36 buckets. The buckets and crossings were prepared as previously described. Thus, the seeds of the RC_1 backcrossing were obtained in the summer of 2017 and RC_2 in the winter of 2017. The seeds of the F_2 and F_3 generations were obtained by natural self-fertilization of the F_1 and F_2 hybrids, respectively.

After each harvest of the emasculated and pollinated spikes, they were tracked, and the grains were quantified and stored for determining the reaction to FHB in the field in the winter of 2018.

Experiment 2

The study was performed in the experimental field at Embrapa Trigo, in the city of Passo Fundo, Rio Grande do Sul State, Brazil, located at an altitude of 687 m, the latitude of 28°15' S, and longitude of 52°24' W, in the winter of 2018; six segregating populations of wheat were used in F_2RC_2: CIGM90.909/BRS Guamirim; CIGM90.909/BRS 179; CIGM92.1666/BRS Guamirim; CIGM92.1666/BRS 179; CIGM93.298/BRS Guamirim, and CIGM93.298/BRS 179. These populations came from crossings between synthetic wheat and elite wheat cultivars, developed in a greenhouse in 2016 and 2017.

The experimental design was completely randomized blocks with eight treatments and four replicates consisting of six combinations of crossings and two male parents. Each material was sown manually on July 16, 2018, in plots with a 5 m row, with 50 seeds spaced at 10 cm. The spacing between plots in the sowing row was 1.00 m and laterally 0.40 m. For the blocks, the spacing was 0.80 m to allow the placement of hoses on the ground to wet the spikes. The trial was conducted following the Technical Indications for Wheat and Triticale Culture. Leaf diseases were controlled until stage 10 (boot) of the Large (1954) scale. It was determined the date when each crossing and the male parent reached 50% of the spiking and the date of collection of the dry spikes of the main stem. At the beginning of the spike formation, wheat grains with mature perithecium of G. zea, produced according to the protocol routinely used at Embrapa Trigo, were distributed in each irrigation row (Lima, 2007). Afterwards, the experimental area was subjected to wetting of spikes with fog formation, on days without rainfall (Lima, Fernandes, & Picinini, 2002).

In the spiking phase, the main stem of each plant was marked with labels and the number of spikelets per spike. In stage 11.2 (grain in soft mass) of Large (1954) scale, the number of spikelets with FHB symptoms was quantified by spike without detaching the plant to determine the real disease severity. The classification of genetic resistance was based on the Kohli (1989) scale: 0, immune; 1, resistant (1 - 5% severity); 2, moderately resistant (5 - 25% severity); 3, moderately susceptible (25 - 50% severity); 4, susceptible (50 - 75% severity); and 5, very susceptible (> 75% severity). This method was used to determine type II resistance.

At the time of wheat maturation, all rows of each plot were manually harvested and the following characteristics were evaluated.

Height of plants (HP): the length of the main stem tiller was measured in centimeters from the ground level to the top of the spike, without the edges; - Number of grains per spike of the main stem of each plant in the row (NGS): the number of grains per spike was evaluated on all spikes of each row harvested from the main tiller, threshed manually, counting the total number of grains obtained per spike. Measurements and counting were performed in the laboratory.

Number of healthy grains (NHG) and the number of diseased grains per spike (NDG) of the main stem of each plant in the row: The grains threshed manually were separated into normal and diseased grains in the laboratory to assess type III resistance.

Grain yield per plant (GYP): The remaining tillers of each plant in each row were harvested and tracked separately in a stationary electric harvester. The air intake was reduced for total grain retention. Afterward, the samples were passed through the blower to eliminate excess straw and then counted to determine the grain yield per plant.

The morphological data of shape and density of the spike were obtained in the maturation phase, after harvesting on the spike of the main tiller, and before manual threshing, at the National Service for the Protection of Cultivars, according to the morphological descriptors elaborated by Brasil (1998).

The evaluated characteristics were subjected to analysis of variance using the F test at a 5% error probability level to detect the effects of genotypes and replicates. For the percentage of the combination of resistant (R) and moderately resistant (MR) plants, the average resistances II and III were used. For the Tukey test at the 5% level, the means of the genotypes in each location were compared. Correlation data were obtained using Pearson's method.

Results and discussion

Development of interspecific segregating populations

The development of the F_1 segregating population of wheat occurred in the winter of 2016. The number of emasculated/pollinated spikes was different for each parental combination because not enough pollen was always obtained to pollinate all the emasculated spikes. The largest number of grains was observed in CIGM93.298/BRS 179 (950 grains) with 50 pollinated spikes and the smallest in CIGM90.909/BRS Guamirim (131 grains) with 39 pollinated spikes. A combination of CIGM90.909/BRS 179 produced 400 grains with 48 pollinated spikes, CIGM92.1666/BRS Guamirim produced 392 grains with 39 pollinated ears, CIGM92.1666/BRS 179 produced 439 grains with 31 pollinated spikes, and CIGM93.298/BRS Guamirim produced 500 grains with 37 pollinated spikes. Combining ability between the parents represents a more accurate measure for the selection of superior populations since the genetic parameters are estimated based on the progenies and parents (Pimentel et al., 2013).

The production of the RC_1 population occurred in the summer of 2017. Because wheat is not a summer crop, the number of grains in the combinations at the crossings was drastically reduced, except for the CIGM90.909/BRS Guamirim combination that, with fewer pollinated spikes than in F_1, produced more grains (180 grains) with 30 pollinated ears. The CIGM90.909/BRS Guamirim combination with 25 pollinated spikes produced 170 grains, CIGM92.1666/BRS Guamirim with 30 pollinated spikes produced 150 grains, CIGM92.1666/BRS 179 with 30 pollinated spikes produced 160 grains, CIGM93.298/BRS Guamirim with 30 pollinated spikes produced 70 grains, and CIGM93.298/BRS 179 with 30 pollinated spikes produced 100 grains.

Some studies have shown that crossings with synthetic parental generated greater production of grains under environmental heat stress. There was an increase of 10% and 15% in the number of grains, indicating that synthetic wheat can also be a potential source of adverse environmental conditions (Sharma et al., 2014; Jafarzadeh et al., 2016), possibly because *Ae. tauschii* grows widely in hostile environments and therefore may have useful genes for tolerating biotic and abiotic stresses (Elbashir, Gorafi, Tahir, Kim, & Tsujimoto, 2017). However, this was only verified in one crossing, and it is worth noting that the generation was still in RC_1. Thermal stress negatively affects the morphological, phenological, and physiological characteristics of all phases; in particular, it decreases the chlorophyll content and photosynthetic capacity of the leaves (Prasad, Pisipati, Mutava, & Tuinstra, 2008).

The ideal temperature for the growth and yield of wheat is 18 - 24°C (Kuhnem, Rosa, Wagner, & Rosa, 2020). The temperature in the summer of 2017 was high (28°C) impairing the yield, although this species shows some plasticity related to this variable that allows the development of segregating populations even in that period, a strategy adopted by numerous genetic breeding programs. However, according to Stone and Nicolas (1994), exposure to 28 - 32°C for five to six days reduced wheat yield by up to 20%. In addition, when the culture is exposed to high temperatures before anthesis, the number of grains is reduced by a decrease in the number of spikes and the number of grains per spike (Barnabás, Jäger, & Fehér, 2008). This is because high temperatures affect the mobilization of starch within the anther, interrupting the development of the pollen grain and intensifying its mortality (Zhang et al., 2013).

The last generation of segregating populations produced in this study was developed in the winter of 2017. All breeding combinations had the same number of pollinated and emasculated spikes (40 spikes). In addition, it is worth noting that the work was conducted in a greenhouse under equal and well-controlled conditions for all plants. In this generation, all genotypes of synthetic wheat crossed with the BRS 179 cultivar presented a higher number of grains. In contrast, all genotypes crossed with the BRS Guamirim had a lower number of grains.

The selection of crossings that represent a good combination between parents maximizes the grain yield per plant, the number of grains per spike, and the plant height (Bornhofen et al., 2013). The main characteristics desired for the new wheat cultivars are grain yield, industrial quality, tolerance to spike germination, disease resistance, drought tolerance, toxic aluminum in the soil, natural threshing and lodging, tillering capacity, good response to fertilization, and wide adaptability and phenotypic stability (Marchioro et al., 2009).

Major differences in important agronomic characters together with the estimates of the degrees of dominance/heterosis as well as the heritability and correlation studies obtained will allow to properly conduct segregating populations, aiming at obtaining lines that can eventually be transformed into new cultivars of interest for wheat cultivation in the state of Rio Grande do Sul.

Agronomic and morphological assessments of segregating populations

Analysis of variance identified significant differences in the parameters of evaluated segregating populations of wheat and the male parents, except for the number of plants (Table 2) and spike shape (Table 3).

Table 2. Summary of the analysis of variance of wheat segregating populations and male parents evaluated in the field in 2018.

		Medium Square			
FV	DF	NP	NTS	NDG	PDG
Block	3	9.83^{ns}	3.56*	1.35*	15.69*
Treatment	7	11.5	18.84	12.26	776.63
Residue	21	17.5	0.38	1.29	44.58
CV (%)		10.24	3.62	15.50	15.20

*significant ($p < 0.05$); ns = not significant ($p > 0.05$); FV: factor of variation; DF: degrees of freedom; NP: number of plants; NTS: number of total spikelets; NDG: number of diseased grains; PDG: percentage of diseased grains; CV: coefficient of variation.

Table 3. Summary of the analysis of variance of wheat segregating populations and male parents evaluated in the field in 2018.

		Medium Square							
FV	DF	NTG*	NDG*	PDG*	NHG*	GYP*	HP*	SSns	SD*	R+MR*
Block	3	46.53	9.69	178.24	25.79	9057.98	99.12	1.16	3.36	4.88
Treatment	7	122.83	32.88	161.69	44.96	1706.90	493.15	1.45	20.17	4.12
Residue	21	3.85	3.50	41.88	3.17	275.39	18.06	0.79	1.08	1.03
CV (%)		8.18	13.48	10.45	17.68	10.57	5.54	35.08	2.32	20.79

*significant ($p < 0.05$); ns = not significant ($p > 0.05$). FV: factor of variation; DF: degrees of freedom; NTG: number of total grains; NDG: number of diseased grains; PDG: percentage of diseased grains; NHG: number of healthy grains; GYP: grain yield per plant; HP: plant height; SS: spike shape; SD: spike density; R + MR: resistant + moderately resistant.

For the variable number of spikelets, four distinct groups were formed (Table 4). The BRS 179 cultivar and the crossing CIGM93.298/BRS 179 remained in the same group and did not differ statistically. This group had the highest number of spikelets. All segregating populations crossed with BRS Guamirim, including this male parent, remained in the same statistical group, with fewer spikelets. Gadimaliyeva et al. (2018) analyzed agronomic traits in three groups of synthetic wheat and found an average of 20 spikelets per spike of wheat. Similar values were found in the present study at crossings with BRS 179.

For the variables spikelet with FHB and its percentage, two statistically distinct groups were formed (Table 4): CIGM93.298/BRS Guamirim (76.72%) crossing that presented the most affected spikelets. The other crossings and male parents remained in the same group, with percentages of less than 50% of diseased spikelets. According to the Kohli (1989) scale, all segregating populations plus male parents were considered moderately susceptible and very susceptible to type II resistance. Similar studies by Hartel, Berzonsky, Kianian, and Ali (2004), carried out in two seasons, spring and summer, observed that the combinations of synthetic wheat 01NDSWG-5 from *T. turgidum* L. var. *dicoccoides* with *T. tauschii* exhibited FHB severity rates of 36 and 32%, respectively. Cultivar Alsen, spring wheat resistant to FHB, showed severity of 9% and 30%, respectively. The susceptible cultivar used in this study had a severity of 70 and 96%, respectively. Phenotypic evaluations were conducted in a greenhouse located on the campus of the University of North Dakota.

Mendes, Ponte, Feltrin, Badiale-Furlong, and Oliveira (2018), looking for FHB resistance in Brazilian wheat cultivars, observed high severity (> 70%) in the spikes of all wheat genotypes at 5 days after the 2-day incubation, especially in plants inoculated with *F. graminearum*.

Table 4. Number of spikelets per plant (NSP), number of diseased spikelets (NDS), and percentage of diseased spikelets (PDS) in F_1RC_2 populations and male parents produced in 2018.

Crossings/Male parents	NSP	NDS	PDS
CIGM90.909/BRS Guamirim	15.00 c[1]	7.16 b	47.72 b
CIGM90.909/BRS 179	17.68 b	7.77 b	42.69 b
CIGM92.1666/ BRS Guamirim	15.14 c	6.00 b	39.30 b
CIGM92.1666/BRS 179	18.55 ab	6.73 b	36.00 b
CIGM93.298/BRS Guamirim	14.82 c	11.39 a	76.72 a
CIGM93.298/BRS 179	19.29 a	6.83 b	35.26 b
BRS Guamirim	14.86 c	5.83 b	39.20 b
BRS 179	19.85 a	6.95 b	34.65 b

[1]Averages followed by the same letter in the column did not differ statistically by the Tukey test at 5% of error probability.

For the variable number of grains (Table 5), two groups were formed. In group (a), the crossings performed with the male parent BRS 179 presented a higher number of grains, whereas group (b) composed of the male parent BRS Guamirim and all its crossings produced fewer grains. There were four statistically distinct groups for the number of diseased grains, with the crossing CIGM93.298/BRS 179 presenting the greatest number of diseased grains and the CIGM93.298/BRS Guamirim the least. For the percentage of diseased grains, three groups were formed, with the majority of crossings except for CIGM90.909/BRS 179 and male parents having values above 50% of diseased grains. In a study of resistance to gibberellas, Martin et al. (2017) found that all analyzed wheat samples inoculated with *F. graminearum* contained grains affected by the disease at different intensities regardless of the variety and the environment.

For the number of healthy grains, the best results were observed in the male parent BRS 179 and in the crossing CIGM90.909/BRS 179 that did not differ statistically. The parent BRS Guamirim and its crossings CIGM92.1666/BRS Guamirim and CIGM93.296/BRS Guamirim showed lower values for healthy grains.

For the number of grains per plant, four statistically distinct groups were formed: the greatest yield was found for the crossing CIGM93.298/BRS 179 and the lowest for CIGM90.909/BRS 179 (Table 5). Grain yield in wheat is determined by several components: number of spikes per plant, number of spikelets per spike, number of grains per spike and spikelet, and average grain weight, depending directly on genetic and environmental factors (Grafius, 1956; Cruz & Carneiro, 2006). In 2003, Chuanmai 42, a cultivar obtained from a cross between a CIMMYT synthetic wheat and a variety of wheat, was launched in China, producing 22.7% more than the commercial control, the Chuanmai 107 cultivar (Yang et al., 2009). Ogbonnaya et al. (2007) found that lines resulting from synthetic wheat yield 8 - 30% more than elite wheat cultivars in different environments. This reinforces previous research performed at CIMMYT in which lines from synthetic wheat significantly improved grain yield in different environments (Ortiz et al., 2008).

Table 5. Number of grains per spike (NGS), number of diseased grains (NDG), percentage of diseased grains (PDG), number of healthy grains per plant (NHG), and number of grains per plant (NGP) in the F_1RC_2 and male parents in 2018.

Crossings/Male parents	NGS	NDG	PDG (%)	NHG	NGP
CIGM90.909/BRS Guamirim	20.62 b[1]	11.75 bc	60.52 ab	8.83 bc	154.78 bc
CIGM90.909/BRS 179	27.67 a	12.83 bc	49.58 b	14.73 a	134.05 c
CIGM92.1666/ BRS Guamirim	18.45 b	11.75 bc	67.55 a	6.71 c	137.70 bc
CIGM92.1666/BRS 179	31.60 a	16.18 ab	60.69 ab	11.80 ab	150.75 bc
CIGM93.298/BRS Guamirim	18.60 b	11.64 c	66.76 a	6.84 c	153.05 bc
CIGM93.298/BRS 179	31.60 a	19.48 a	64.43 ab	12.14 ab	198.11 a
BRS Guamirim	18.26 b	12.09 bc	68.65 a	6.12 c	152.80 bc
BRS 179	28.84 a	15.48 abc	57.38 ab	13.37 a	174.90 ab

[1]Averages followed by the same letter in the column did not differ statistically by the Tukey test at 5% of error probability.

For the variable plant height (HP), two statistical groups were formed, the first being all crossings made with the parent BRS 179 with larger plants and the second with all crossings made with BRS Guamirim with smaller plants (Table 6). Plant height has been reported as a passive mechanism of resistance to FHB in numerous studies, showing that taller plants tend to have fewer diseases (Buerstmayr & Buerstmayr, 2015). A study by He, Singh, Duveiller, Dreisigacker, and Singh (2013) also revealed this trend reporting that among

12 accessions that presented very little infection, eight were taller than 120 cm; there may be new genes or resistance alleles in these tall accessions. The lack of symptoms of FHB is generally associated with tall stature and low yield; the breeding experience in China has shown that it is difficult to use these materials in breeding (Bai & Shaner, 2004). Gadimaliyeva et al. (2018) studied agronomic traits in lines from synthetic wheat and found average height for all synthetic wheat and its derivatives ranging from 85 to 126 cm, with no line being considered semi-dwarf. Szabo-Hever et al. (2018) indicated that smaller synthetic wheat plants had higher disease severity. The relationship between resistance and plant height was confirmed by a meta-analysis of several QTLs. Smaller plants differently affect the severity of FHB; the mechanisms of these associations are complicated (Mao et al., 2010). However, it has been suggested that taller plants can dry out more quickly and might be less infected by the disease (Cheng et al., 2018).

Resistance to FHB has often been found to be associated with morphological characteristics and plant development, especially height, spike architecture, anther extrusion, and flowering date, acting mainly as passive resistance factors (Mesterházy, 1995).

For spike shape, no material statistically differed (Table 6). The spike density showed the formation of four distinct groups, with the highest density found for the crossing CIGM93.298/BRS Guamirim and the lowest density in crossings with the male parent BRS 179 (Table 6). Some morphological characteristics are associated with the increased disease. In genotypes with dense spikes, the tendency is for the pathogen to spread rapidly, in contrast to the lengthened spike genotypes. In addition, small genotypes are more severely infected than tall genotypes (Alves, Nora, Franco, Costa, & Stangarlin, 2013).

Table 6. Plant height (HP), spike shape (SS), and spike density (SD), in F_1RC_2 populations and male parents produced in 2018.

Crossings/Male parents	HP (cm)	SS	SD (mm)
CIGM90.909/BRS Guamirim	69.87 b[1]	2.07 a	46.07 bc
CIGM90.909/BRS 179	84.69 a	3.41 a	42.33 d
CIGM92.1666/ BRS Guamirim	63.86 b	2.29 a	44.79 bcd
CIGM92.1666/BRS 179	82.61 a	3.30 a	42.74 d
CIGM93.298/BRS Guamirim	71.71 b	2.03 a	48.82 a
CIGM93.298/BRS 179	90.14 a	2.29 a	44.39 cd
BRS Guamirim	62.47 b	1.88 a	47.12 cd
BRS 179	88.73 a	2.93 a	43.42 d

[1] Averages followed by the same letter in the column did not differ statistically by the Tukey test at 5% of error probability.

The correlation analysis of the data (Table 7) showed that the greater the number of diseased spikelets, the greater the percentage of diseased spikelets. In turn, the greater the number of diseased grains, the greater the percentage of diseased grains. The other variables analyzed had a weak correlation; Dancey and Reidy (2006) showed that the factors are related as follows: r = 0.10 to 0.30 (weak); r = 0.40 to 0.6 (moderate); r = 0.70 to 1 = strong).

Table 7. Pearson's correlation of all analyzed variables.

Variables	NDS	PDS	NDG	PDG	NHG	GYP	HP	SS	SD
NDS	1								
PDS	0.96	1							
NDG	0.11	-	1						
PDG	0.36	0.34	0.56	1					
NHG	-0.32	-0.36	-0.30	-0.85	1				
GYP	0.17	0.09	0.23	0.03	0.08	1			
HP	-	-0.12	0.32	-0.15	0.38	0.28	1		
SS	-	-	0.06	-0.18	0.27	0.15	0.28	1	
SD	0.32	0.38	-	0.17	-0.20	0.20	-0.17	-0.48	1

NDS = Number of diseased spikelets; PDS = Percentage of diseased spikelets; NDG = Number of diseased grains; PDG = Percentage of diseased grains; NHG = Number of healthy grains; GYP = Grain yield per plant; HP = Plant height; SS = Spike shape; SD = Spike density; ** ($p < 0.01$); * ($p < 0.05$) by the t test, where $0 < rxy < 0.3$ (weak correlation); $0.3 \leqslant rxy < 0.6$ (moderate correlation); $rxy \geqslant 0.6$ (strong correlation) (Callegari-Jacques, 2003).

Based on the results of the analyzed variables, in tests to verify type II and III resistance in crossings and male parents, it was identified that the best resistance to FHB was present in the crossing with the synthetic accession CIGM90.909 with the cultivar BRS 179 (Table 8). More than 60% of this material showed resistance, in addition to having a medium cycle, being in the group of taller and less dense plants, corroborating with the reports that taller plants start flowering later and possess better resistance to FHB. Wheat cultivars differ in their response to FHB; some are more resistant, others are highly susceptible, but no genotype is immune (Gunupuru, Perochon, & Doohan, 2017).

Although the F_1RC_2 CIGM90.909/BRS 179 population was the most resistant to types II and III, it had the lowest yield. It is often difficult to combine the full set of desired characteristics in an initial crossing between primary synthetic wheat and elite wheat or commercial cultivar. There is a low probability of finding lines with yield, disease resistance, and agronomic performance equal to or greater than that of elite wheat that simultaneously has good grain quality. Therefore, lines developed in this way can be a useful source of genetic diversity, but they need to present competitive agronomic characteristics or be superior to commercial varieties of wheat in all aspects (Dunckel, Crossa, Wu, Bonnett, & Poland, 2017).

Table 8. Percentage of the combination of resistant (R) and moderately resistant (MR) plants in F_1RC_2 populations and male parents produced in 2018.

Crossings/ Male parents	Combination of plants R + MR (Type II e III %)
CIGM90.909/BRS 179	6.9 a[1]
BRS 179	5.5 ab
CIGM90.909/BRS Guamirim	5.3 ab
CIGM92.1666/BRS 179	4.9 ab
CIGM93.298/BRS Guamirim	4.4 b
CIGM92.1666/BRS Guamirim	4.4 b
GIGM93.298/BRS 179	4.1 b
BRS Guamirim	3.7 b

[1] Averages followed by the same letter in the column did not differ statistically by the Tukey test at 5% of error probability.

The results of the present study were less satisfactory than those reported by Zhu et al. (2016) that evaluated CIMMYT's synthetic wheat lines (SYN1- Mayoor // Tksn1081 / *Ae. Squarrosa*-222), classified as highly resistant to FHB, with an average rate of 5.7% over two years. Jin et al. (2013) characterized the resistance of 363 synthetic lines from the USA and found that 75% were susceptible and moderately susceptible (43 and 32%, respectively), and only 25 (7.0%) were classified as resistant. Szabo-Hever et al. (2018) analyzed 149 lines of synthetic wheat and their wheat tetraploid progenitors (*T. turgidum* L.) for the analysis of genetic diversity and evaluation of resistance to FHB. These synthetic lines were developed by crossing 10 accessions of *Ae. tauschii* with 74 tetraploid wheat accessions belonging to durum wheat and five other subspecies of tetraploid wheat (*T. carthlicum*, *T. dicoccum*, *T. polonicum*, *T. turgidum*, and *T. turanicum*); they found 13 synthetic lines showing a high level of resistance to FHB, offering on average reduction of 18.3% in FHB compared to tetraploid parents. In our study, the CIGM90.909/BRS 179 segregating population also proved to be more resistant than the parents, perhaps indicating that the D genome may play an important role in reducing the severity of the disease in synthetic wheat (Szabo-Hever et al., 2018).

Overall, our observations suggest that all types of resistance to FHB interact and can be interdependent (Mesterházy, Bartók, Mirocha, & Komoroczy, 1999); resistance to this disease is believed to be a combination of its response to the incidence and severity (Giancaspro, Giove, Zito, Blanco, & Gadaleta, 2016).

Conclusion

In the three generations (F_1, RC_1, and RC_2), the best crossbreeding combinations as verified by the number of grains were with the male parent BRS 179; in two consecutive generations RC_1 and RC_2, the highest number of grains was in CIGM90.909/BRS 179. For field evaluations performed in 2018, the F_1RC_2 CIGM90.909/BRS 179 population showed better results for resistance to FHB but lower grain yield per plant. Therefore, it is evident that the crossings conducted with the male parent BRS 179 had better resistance to the disease. It is recommended to continue backcrossing with the same male parents, developing segregating populations.

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