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Genetic diversity of resistance genes controlling fusarium head blight with simple sequence repeat markers in thirty-six wheat accessions from east asian origin

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Summary

Fusarium head blight (FHB) is a serious disease of wheat worldwide that may cause substantial yield and quality losses. Breeding for FHB-resistant cultivars is the most cost-effective approach to control FHB. The objective of the present study was to determine the relationship of resistance between new resistant sources and Sumai 3 using five simple sequence repeat (SSR) markers closely linked to the major QTL for FHB resistance on chromosome arms 3BS and 6BS. All five SSR markers were highly polymorphic between Sumai 3 (and its derivatives) and susceptible Canadian wheat lines. Most of the Sumai 3-derived Chinese wheat accessions and three Canadian FHB-resistant lines had all the Sumai 3 SSR marker alleles on chromosome arms 3BS and 6BS. The Chinese landrace Wangshuibai and two Japanese accessions Nobeokabozu and Nyu Bai had the same banding patterns as Sumai 3 for all five SSR marker alleles, and another Chinese landrace Fangshanmai had three of the five SSR markers in common with Sumai 3, and therefore most likely carries the same QTL as Sumai 3 on 3BS and 6BS. The Brazilian cultivar Frontana had no alleles in common with Sumai 3 on either QTL, and the Chinese landrace Hongheshang had only one of the five SSR markers in common with Sumai 3, therefore likely carrying resistance genes different from Sumai 3. The Italian cultivar Funo is not the donor of either the 3BS QTL or 6BS QTL. All five SSR seem to be effective candidates for marker-assisted selection to increase the level of resistance to FHB in wheat breeding programs.

Introduction

Fusarium head blight (FHB), caused mainly by *Fusarium graminearum* Schwabe (teleomorph *Gibberella zeae* (Schwein.) Petch), is one of the most severe and notorious diseases of wheat worldwide (Bai & Shaner, 1994; Yang, 1994; Parry et al., 1995). FHB causes substantial losses in grain yield, quality and performance of saved seed. Furthermore, the most serious threat of FHB is the problem of grain contamination caused by associated mycotoxins such as deoxynivalenol (DON), zearalenone and moniliformin. As FHB epidemics have become more frequent and severe in Canada, contamination with mycotoxins may become a major problem for animal production and human health, and may impact both domestic and export food and feed markets. In western Canada, the Manitoba FHB epidemic of 1993 caused losses in wheat and barley of $100 million, and since then losses have averaged $50 million annually (Gilbert & Tekauz, 2000; Tekauz et al., 2000). Further spread of FHB could threaten Saskatchewan, and possibly Alberta. In eastern Canada, epidemics of FHB in wheat have occurred regularly in Ontario, Quebec and Prince Edward Island. Canada's reputation as a reliable supplier of sound, high quality grain is at stake, particularly if FHB becomes more widespread.

The development of genetically FHB-resistant cultivars is the most cost-effective method to control the disease (Yang, 1994; Parry et al., 1995).

Sources of FHB resistance from China, including Sumai 3 and its derivatives, primarily have been used in wheat breeding programs around the globe, including most major Canadian spring wheat breeding programs (Humphreys et al., 2001). The molecular characterization of the genetic basis of FHB resistance in elite wheat germplasm is key to providing early generation screening for breeders' lines, allowing for more rapid advances in breeding FHB-resistant cultivars using marker-assisted selection, and utilizing additional resistance sources for increasing the level of FHB resistance through stacking of resistance genes. Molecular mapping studies have identified and mapped quantitative trait loci (QTL) for Type II resistance to fungal spread within the spike (Schroeder & Christensen, 1963) on chromosomes 2BL (Zhou et al., 2002; Gervais et al., 2003), 2AS (Gupta et al., 2000; Zhou et al., 2002), 2DL(Somers et al., 2003), 3AL (Anderson et al., 2001), 3BS (Bai et al., 1999; Waldron et al., 1999; Anderson et al., 2001; Buerstmayr et al., 2002; Yang et al., 2002, 2003; Shen et al., 2003), 4BL (Waldron et al., 1999), 5A (Buerstmayr et al., 2002; Yang et al., 2002), 6B (Anderson et al., 2001; Yang et al., 2002, 2003; Shen et al., 2003), and 6AS (Anderson et al., 2001). The most prominent and consistent QTL segregating for Sumai 3-derived resistance genes were identified on chromosome arms 3BS and 6BS. The 3BS QTL was detected in different mapping populations segregating for Sumai 3-derived resistance genes, explaining 10%–60% of the phenotypic variation for resistance to spread within a wheat spike in different replicates or test environments (Bai et al., 1999; Waldron et al., 1999; Anderson et al., 2001; Buerstmayr et al., 2002; Yang et al., 2002, 2003; Shen et al., 2003). The best SSR markers linked to the 6BS QTL explained 4%–22% of the phenotypic variation for FHB resistance (Anderson et al., 2001; Yang et al., 2002, 2003; Shen et al., 2003).

The predictive value of a molecular marker depends on linkage to the resistance gene and the phenotypic expression of the gene in other genetic backgrounds and environments. Bai et al. (2003) and Liu and Anderson (2003) characterized wheat accessions with markers tightly linked to the major 3BS QTL. McCartney et al. (2004) haplotyped a diverse set of wheat germplasm with SSR markers linked to QTL on chromosome arms 2DL, 3BS, 3BSc, 4B, 5AS and 6BS. These studies provided an effective approach to rapidly differentiate germplasm with different resistance genes. The objective of this study was to evaluate the effectiveness of SSR markers on 3BS and 6BS QTL identified in Sumai 3 and its derivatives as a marker-assisted selection (MAS) tool for screening putative new FHB-resistance resources.

Materials and methods

Plant materials

Thirty-six accessions of spring wheat from China, Japan, Brazil and Canada were used to evaluate the polymorphism of five SSR markers linked to QTL on chromosome arms 3BS and 6BS found in Sumai 3 and its derivatives (Table 1). The majority of these wheat accessions were reported to show various levels of FHB resistance based on field and greenhouse inoculations, including the well-known Chinese FHB-resistant accessions Wangshuibai, Sumai 3, Ning 7840, and a set of advanced lines from Chinese wheat breeding programs. Three Canadian wheat cultivars, AC Foremost, Glenlea and HY368, were used as the susceptible controls.

DNA isolation and genotyping

Leaf tissue of each wheat accession was collected at the 2–3 leaf stage, submerged in liquid nitrogen, lyophilized and stored dry at $-20\,^{\circ}C$ for DNA isolation. Genomic DNA was isolated with the DNeasy 96 plant kit (Qiagen, Mississauga, Ontario) and was quantified by fluorometry using Hoechst 33258 stain.

Five SSR markers tightly linked to the two major QTL for FHB resistance, three on chromosome arm 3BS (Xgwm389, Xgwm533 and Xgwm493) and two on chromosome arm 6BS (Xgwm644 and Xgwm88) (Yang et al., 2002, 2003), were used to screen putative new sources of FHB resistance in 36 wheat accessions. SSR markers were amplified according to the method of Röder et al. (1998) with some modifications and separated on 3% (w/v) Metaphor agarose gels. Amplified DNA products were electrophoresed at 120 V for 4 h, and visualized by staining with ethidium bromide and photographed under UV light for scoring alleles. Presence and absence of a band with the same molecular size was assumed to be two alleles at a locus and variation in band presence was recorded as a polymorphism.

Inoculum, inoculation and FHB phenotyping

The inoculum of *Fusarium graminearum* used for inoculation in the present study was a mixture of

Table 1. Origin and pedigrees of 36 wheat accessions

Accession	Origin	Pedigree
Sumai 3(1)	Jiangsu, China	Funo/Taiwanxiaomei
Sumai 3(2)	China	Funo/Taiwanxiaomei, from Canada Gene Bank
Ning 7840	Jiangsu, China	Aurora/Anhui 11//Sumai 3
Fu 5114	Jiangsu, China	Longxi 18/(Aurora/Anhui 11//Sumai 3)
Fu 5125	Jiangsu, China	Ning 7840/Fufan 904
Ning 8331	Jiangsu, China	Yangmei 4/(Avrora/Anhui 11//Sumai 3)
Ning 8026	Jiangsu, China	Avrora/Sumai 3//Yangmai 2
N991126	Jiangsu, China	Sumai 3/Wannian 2//90P25
Wangshuibai	Jiangsu, China	Landrace from Jiangsu Province
Hongheshang	Jiangsu, China	Landrace from Zhejiang Province
Fangshanmai	Fujian, China	Landrace from Fujian Province
N991130	Jiangsu, China	Yangmai 158/Wangshuibai
N894037	Jiangsu, China	Somatic mutant from Yangmai 3
AT2	Jiangsu, China	Somatic mutant from Ningmai 3
N894013	Jiangsu, China	Somatic mutant from Ningmai 3
N962424	Jiangsu, China	Somatic mutant from Yangmai 158
N991069	Jiangsu, China	87-158/89-92//Ning 894013
N983222	Jiangsu, China	Derivative of wheat/Elymus
N991119	Jiangsu, China	Ning 895004/Mexico 354
Shenmai 2	Shanghai, China	Unknown, from recurrent selection
Lunhui 201	Shanghai, China	Unknown, from recurrent selection
SH19089	Shanghai, China	Unknown, from recurrent selection
Yangmai158	Jiangsu, China	Yangmai 4/St 1472/506
Yangmai 5	Jiangsu, China	(Nanda 2419/Triumph//Funo)/St 1472/506
Yangmai 4	Jiangsu, China	Nanda 2419/Triumph//Funo
Nanda 2419	Jiangsu, China	Reselected line from Mentana
Funo	Italy	Duecentodieci/Demiano
Nobeokabozu	Japan	Landrace
Nyu Bai	Japan	Unkown
Frontana	Brazil	Fronteira/Mentana
92FHB 21	CRC, Canada	Ning 8331/HY368
93FHB 37	CRC, Canada	HY611/Ning 8331
DH181	ECORC[1], Canada	Sumai 3/HY368
AC Foremost	CRC[2], Canada	HY368*5 /BW533// HY368*6 /7424.B
HY 368	CRC, Canada	Tobari 66 / Romany
Glenlea	Canada	Pembina*2/Bage//CB100

[1]ECORC: Eastern Cereal and Oilseed Research Centre, Agriculture and Agri-Food Canada; [2]CRC: Cereal Research Centre, Agriculture and Agri-Food Canada.

four relatively aggressive isolates (Yang et al., 2003). Actively growing cultures of *F. graminearum* on potato dextrose agar (Becton, Dickinson and Company, Sparks, MD, USA) were blended, added to liquid carboxymethyl cellulose sodium salt (Omega Chemical Company Inc, Quebec, Canada) and incubated under aeration for 5–7 d at room temperature. Concentration was standardized to 50,000 macroconidia/ml using a hemacytometer.

Thirty-six wheat accessions were evaluated for FHB resistance in both greenhouse and field trials. In the greenhouse trials, each accession was planted in five pots and randomly arranged on greenhouse benches in each experiment. In each pot, the first two spikes that flowered at the same time were inoculated using single-floret inoculation. The central florets on 10 spikes were inoculated with a micropipette at early anthesis with 10 μl of a macroconidial suspension of *F. graminearum*. Following inoculation, plants were incubated in a humidity chamber at 100% relative humidity for 24 h and then returned to the greenhouse bench. Temperatures were maintained at 20–25 °C and

lights were set to give 16 h daylight. The greenhouse temperature averaged 22 °C during the day with a range of 18 to 25 °C and 18 °C at night with a range of 17 to 21 °C. The greenhouse relative humidity averaged 75% with a range of 50% to100%. Disease symptoms were rated 21 d after inoculation and recorded as percentage of infected spikelets per spike.

In the field trials, the 36 accessions were planted during a two-year period (2001 and 2002) in the black soil zone at the Glenlea Experimental Station, Cereal Research Centre, Manitoba, Canada. The experimental layout was a randomized complete block design with two replicates. Plots consisted of a single row with 17 cm row spacing and 1.5 m length. Sowing density was approximately 60 seeds per row. In both years the heads of the entire row were spray-inoculated with a macroconidial suspension of 50,000 macronidia/ml using a CO_2-powered backpack sprayer (40 ml/row) at 50% anthesis. The spray-inoculation was repeated two or three days later. Nurseries were mist-irrigated with a sprinkler system for 30 min after each inoculation to favor development of the disease. The whole row was harvested at 21 d and frozen for recording disease symptoms. The number of infected spikelets and total spikelets per spike were counted for at least 30 random spikes from each plot to provide a measure of field FHB resistance expressed as percentage infected spikelets per spike.

Statistical analysis

The FHB data were subjected to analysis of variance (ANOVA) (SAS Institute, Raleigh, NC) as a randomized complete block design.

Results and discussion

ANOVA indicated significant variation for FHB resistance among the 36 wheat accessions and the accession × experiment interaction in both controlled environment and field experiments (Data not shown). The wheat accessions showed significantly different reactions to *F. graminearum* infection (expressed as percentage of symptomatic spikelets) (Table 2). In the greenhouse trials, mean FHB severities of wheat accessions ranged from 4.5% (Hongheshang) to 76.9% (AC Foremost). Except for FHB-susceptible control cultivars, 21 accessions had percentage infected spikelets per spike less than 20%, and ten had percentage infected spikelets per spike less than 40%, indicating that most wheat accessions evaluated in the present study were resistant or moderately resistant to FHB.

In the field trials, percentage infected spikelets per spike ranged from 6.2% (Sumai 3) to 89.1% (AC Foremost). Most of the wheat accessions showed higher FHB infection levels in the field than in the greenhouse (Table 2). Twenty accessions had percentage infected spikelets per spike less than 20%, and eleven had percentage infected spikelets per spike less than 40%. Six accessions Ning 8026, Nobeokabozu, Frontana, Fangshanmai, N991119 and N991130 showed higher head blight levels in the greenhouse than in the field trials. The correlation coefficient between field and greenhouse data was significant ($r = 0.96$).

Table 2 shows polymorphisms of the five SSR markers linked to the two major QTL on chromosome arms 3BS and 6BS among the 36 accessions. A total of seven Sumai 3-derived resistant accessions, Ning 7840, Fu 5114, Ning 8331 and N991126 from China, and 92FHB 21, 93FHB 37 and DH181 from Canada, had all five Sumai 3 SSR alleles, and therefore most likely carry both 3BS and 6BS QTL. One Sumai 3-derived resistant accession Fu 5124 exhibited three SSR marker alleles similar to Sumai 3 and for Xgwm389 a marker allele similar to AC Foremost while that of Xgwm644 was missing. Ning 8026 had only the same banding pattern as Sumai 3 for SSR marker loci on the 6BS QTL. None of the three Canadian FHB-susceptible cultivars used in the present study had banding patterns in common with Sumai 3. Identification of putative new FHB-resistant germplasm using SSR markers tightly linked to the major 3BS QTL and 6BS QTL is important when selecting parents in wheat breeding programs aimed at improving FHB resistance, since the two QTL explain a large portion of the genetic variation for FHB resistance (Yang et al., 2002, 2003). Chinese FHB-resistant wheat germplasm is one of the main sources of FHB resistance (Snijders, 1990; Yang, 1994; Wan et al., 1997; Gilchrist et al., 2000), being used in most major Canadian spring wheat breeding programs (Humphreys et al., 2001). All five SSR markers, three on the 3BS QTL (Xgwm389, Xgwm533 and Xgwm493) and two on the 6BS QTL (Xgwm644 and Xgwm88) were highly polymorphic between FHB-resistant accessions (most from China) and FHB-susceptible cultivars from Agriculture and Agri-Food Canada. Therefore, these SSR are effective candidate markers for marker-assisted selection of the two major FHB-resistant QTL in Canadian wheat breeding programs. Most Sumai 3-derived accessions

Table 2. SSR markers linked to the 3BS QTL and 6BS QTL and percentage of infected spikelets per spike (PIS,%) for 36 accessions based on the greenhouse and field trials

Accession	Xgwm389*	Xgwm533	Xgwm493	Xgwm644	Xgwm88	PIS(Greenhouse)	PIS (field)
Sumai 3 (1)	+	+	+	+	+	5.6	6.2
Sumai 3 (2)	+	+	+	+	+	6.0	6.7
Ning 7840	+	+	+	+	+	7.9	9.1
FU5114	+	+	+	+	+	6.2	9.8
FU5125	−	+	+	/	+	6.6	11.1
Ning 8331	+	+	+	+	+	7.9	23.8
Ning 8026	−	−	−	+	+	18.4	10.9
N991126	+	+	+	+	+	22.3	34.7
Wangshuibai	+	+	+	+	+	4.6	9.8
Hongheshang	−	+	−	/	−	4.5	8.8
Fangshanmai	+	−	+	−	+	15.6	8.2
N991130	+	+	+	+	+	20.4	14.2
N894037	+	+	+	+	+	6.6	10.6
AT2	−	+	−	/	−	18.2	17.8
N894013	−	−	−	−	−	17.4	9.4
N962424	−	−	−	−	−	20.3	31.3
N991069	/	−	−	−	−	8.6	13.0
N983222	−	/	+	+	+	20.9	25.5
N991119	−	−	−	−	−	29.4	17.4
Shenmai 2	+	+	+	+	+	15.5	24.5
Lunhui 201	−	+	+	−	−	21.2	29.4
SH19089	+	+	+	+	+	5.2	17.6
Yangmai158	−	−	−	−	−	25.4	22.8
Yangmai 5	−	−	−	−	−	26.7	31.6
Yangmai 4	−	−	−	−	−	23.7	27.1
Nanda2419	−	−	−	−	−	65.2	68.7
Funo	−	−	−	−	−	70.5	76.3
Nobeokabozu	+	+	+	+	+	16.3	28.8
Nyu Bai	+	+	+	+	+	9.2	15.4
Frontana	−	−	−	−	−	25.6	14.4
92FHB 21	+	+	+	+	+	14.5	15.2
93FHB 37	+	+	+	+	+	17.1	21.6
DH181	+	+	+	+	+	7.5	7.1
AC Foremost	−	−	−	−	−	76.9	89.1
HY 368	−	−	−	−	−	71.4	82.7
Glenlea	−	−	−	−	−	74.5	85.7
LSD0.05						26.3	29.7

* +: Resistant allele in common with Sumai 3, − : susceptible allele in common with AC Foremost, and /: missing data.

had the Sumai 3 SSR alleles of both 3BS QTL and 6BS QTL and exhibited FHB-resistance levels equal to Sumai 3 in the greenhouse and field (Table 2). These FHB-resistant accessions will be valuable in marker-assisted selection breeding schemes to develop FHB-resistant germplasm and cultivars.

Four lines derived from tissue culture with the goal of developing somaclonal variants for FHB resistance evaluated in the present study were resistant or moderately resistant to FHB (Table 2). One of them, N894037, was shorter and earlier maturing than Sumai 3 (Lu et al., 2000), and had the same banding pattern as Sumai 3 for SSR marker loci on both 3BS QTL and 6BS QTL (Table 2), in agreement with the result of Shen et al. (2003). Recently, Shen et al. (2003) reported that N894037 carried one major FHB resistance QTL on 3BS and one minor QTL on 6BS. N894037 is a highly FHB-resistant somaclonal variant developed by *in vitro* screening using Yangmai 3 as the source of cells (Lu et al., 2000). The latter was reselected for earliness from the Italian cultivar Funo that is one of the parents of Sumai 3 (Yang, 1994), but is not the donor of

either the 3BS QTL (Bai et al., 2003; Shen et al., 2003; McCartney et al., 2004) or the 6BS QTL (McCartney et al., 2004). It is most unlikely that all five SSR loci at 3BS and 6BS of N894037 were mutated at the same time, although several FHB-resistant wheat variants were obtained by *in vitro* screening (Yang et al., 1998; Lu et al., 2000). Therefore, the FHB resistance QTL of N894037 might come from alternative mechanisms such as outcrossing (Shen et al., 2003). The somatic line AT2 had only one SSR marker allele at Xgwm533 in common with Sumai 3. The other two somatic variants N894013 and N962424 had no Sumai 3-type allele for any of the five SSR markers on either 3BS or 6BS QTL, which is consistent with the results of genetic diversity studies (Lin & Anderson, 2003; McCartney et al., 2004). Recently, Liu and Anderson (2003) reported that N894013 had no Sumai 3-type allele for any of the five SSR markers on the 3BS QTL. McCartney et al. (2004) further confirmed that N894013 and N962424 had no SSR banding patterns in common with Sumai 3 on either 3BS QTL or 6BS QTL. Therefore, these lines most likely carry different resistance genes from Sumai 3.

SH19089 and Shenmai 2 had all the Sumai 3 marker alleles of both 3BS and 6BS QTL, and Lunhui201 had two Sumai 3 marker alleles at the 3BS QTL. These cultivars with resistance or moderate-resistance to FHB (Table 2) were developed by recurrent selection from an initial population pooled with 15 FHB-resistant parents from different regions (Yang et al., 2000). The occurrence of Sumai 3 SSR alleles at Xgwm644 and Xgwm88 in the wheat-Elmyus derivative N983222 suggested the presence of the 6BS QTL.

The Chinese landrace Wangshuibai had the same banding pattern of SSR markers as Sumai 3 on both 3BS QTL and 6BS QTL, which is consistent with the results of recent molecular mapping of FHB-resistance genes in Wangshuibai (Gonzalez-Hernandez et al., 2002, 2003, Zhou et al., 2003, Lin et al., 2004). Gonzalez-Hernandez et al., (2002, 2003) and Lin et al., (2004) reported that Wangshuibai had the major FHB resistance QTL on chromosome arms 3BS and 6BS. Zhou et al., (2003) also found that Wangshuibai had the same major 3BS QTL as Sumai 3. Wangshuibai is an excellent source of both field resistance (combined resistance to initial infection and spread within spike) and resistance to spread within the spike (Table 2). Besides FHB resistance genes on the 3BS and 6BS, Wangshuibai may have FHB resistance genes different from Sumai 3 (Lin et al., 2004). N991130 had Wangshuibai in its pedigree and carried all the SSR alleles from Wangshuibai. Another Chinese landrace Fangshanmai from Fujiang province had three of the five SSR markers in common with Sumai 3 on the 3BS and 6BS QTL. Also, the two Japanese cultivars Nobeokabozu and Nyu Bai showed the same genotypes as Sumai 3 for all five SSR markers on both 3BS and 6BS QTL. However, genetic diversity studies showed that Wangshuibai, Fangshanmai, Nobeokabozu and Nyu Bai had no SSR allele in common with Sumai 3 on either 3BS QTL (Bai et al., 2003; Liu & Anderson, 2003, McCartney et al., 2004) or 6BS QTL (McCartney et al., 2004). Bai et al. (2003) did not find the SSR marker alleles from Sumai 3 in Fangshanmai and Wangshuibai at the 3BS QTL, but recently Zhou et al.(2003) reported that Wangshuibai had the same major QTL as Sumai 3 on 3BS. This ambiguous genotyping of these accessions at SSR loci linked to the 3BS and 6BS QTL may be due to the drawbacks of the dinucleotide nature of repeats of the SSR tested (Liu & Anderson, 2003; McCartney et al., 2004). The regular Metaphor agarose gels used in the present study and even regular sequencing gels (Liu & Anderson, 2003) are unable to effectively distinguish the size difference of two or three nucleotides produced by these dinucleotide repeats.

The Brazilian cultivar Frontana showed no banding pattern in common with Sumai 3 for SSR loci on either 3BS QTL or 6BS QTL. The Chinese landrace Hongheshang from Zhejiang province had only one of five SSR markers in common with Sumai 3 (Table 2). These two wheat accessions, therefore, mostly seem to have FHB resistance genes different from Sumai 3. Previous studies indicated that Frontana had resistance genes different from Sumai 3 (van Ginkel et al., 1996), and no allele in common with Sumai 3 on 3BS (Liu & Anderson, 2003, McCartney et al., 2004) and 6BS (McCartney et al., 2004). Incorporation of FHB-resistant resources from Frontana into adapted Canadian germplasm would diversify the FHB resistance gene pool for improvement of FHB resistance and benefit Canadian wheat breeding programs.

Nanda 2419 and Funo had no Sumai 3 allele for any SSR markers of either 3BS or 6BS QTL (Table 2). Nanda 2419 was reselected from the Italian wheat cultivar Mentana, a relative of Funo (He et al., 2001), which is one of the crossing parents of the Brazilian FHB-resistant germplasm Frontana (Yang, 1994). Nanda 2419 and Funo were highly susceptible to head blight (Table 2). Chinese wheat breeders believe that the FHB resistance in the cultivar Sumai 3 was from the Italian cultivar Funo (He et al., 2001), because they reselected

several major cultivars with improved FHB resistance from either Nanda 2419 or Funo in the mid-lower Yangtze Valley in the late 1970s, including Yangmai 1, Wumai 1 and Wannian 2. Three derivatives of Nanda 2419 and Funo, Yangmai 4, Yangmai 5 and Yangmai 158, in addition to a somatic variant N962424 from Yangmai 158, had no allele in common with Sumai 3 for any of the five SSR markers on either 3BS QTL or 6BS QTL (Table 2). The Yangmai system was moderately resistant to FHB and might carry different FHB resistance genes from Sumai 3. FHB resistance genes in the Yangmai lines might be derived from Nanda 2419 because the latter had three FHB resistance QTL (Lin et al., 2004). Also, Bai et al. (2003) found that Funo-derived cultivars Yangmai 1 and Wannian 2 had no banding pattern in common with Sumai 3 on the 3BS QTL. Molecular evidence in the present study further supported the postulation that the major 3BS QTL of Sumai 3 was most likely derived from Taiwanmai (Bai et al., 2003; Shen et al., 2003). Meanwhile, based on the results of this study, Funo and its derivatives have no Sumai 3 allele for any of two SSR markers linked to the 6BS QTL found in Sumai 3 and its derivatives. Therefore, it can be postulated that the Italian cultivar Funo is not one of the donors that contributed the FHB resistance genes associated with the 6BS QTL. The 6BS QTL of Sumai 3 seems to be most likely derived from Taiwanmai. Further study is worthwhile to determine the possible donor of the 6BS QTL for FHB resistance in the cultivar Sumai 3.

Sumai 3 and its derivatives have been extensively employed as FHB resistance sources in the last decade (Humphreys et al., 2001), therefore, it is necessary to broaden the genetic diversity and search for alternative sources of FHB resistance from Japan and Brazil to reduce the dependency of Canadian wheat breeding on Sumai 3-derived resistance sources. The result of the present study indicated that FHB-resistant resources from Japan and China seem to be very close. Bai et al. (2003) and McCartney et al. (2004) clustered the Chinese landraces including Wangshuibai, Fangshanmai and Hongheshang and the Japanese FHB resistant resources including Nobeokabozu and Nyu Bai into one group. In other words, the genetic diversity among these Asian FHB-resistant resources is very narrow. The reason may be due to the fact that the Japanese and Chinese landraces were probably selected and grown in similar environments under favorable FHB-epidemic conditions. Nine FHB-resistant accessions with resistance or moderate resistance to FHB including Frontana showed all five SSR markers different from Sumai 3 and might carry novel FHB resistance genes. Lin et al. (1992) reported that Yangmai 4 had one FHB resistance gene different from Sumai 3. Even the FHB susceptible cultivar Nanda 2419 had FHB resistance QTL (Lin et al., 2004). Therefore, these accessions could be used to make crosses with Canadian wheats as alternative sources of FHB resistance to broaden the genetic diversity in Canadian wheat breeding programs aimed at improving FHB resistance.

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