

## Research Paper

# Mapping a QTL conferring resistance to Fusarium head blight on chromosome 1B in winter wheat (*Triticum aestivum* L.)

Zenta Nishio<sup>\*1,2</sup>, Chihiro Onoe<sup>3</sup>, Miwako Ito<sup>2</sup>, Tadashi Tabiki<sup>2</sup>, Koichi Nagasawa<sup>2</sup> and Hideho Miura<sup>3</sup>

<sup>1</sup> Tokyo University of Agriculture, 1737, Funako, Atsugi, Kanagawa 243-0034, Japan

<sup>2</sup> NARO Hokkaido Agricultural Research Center, Memuro Research Station, 9-4 Shinsei-minami, Memuro, Kasai, Hokkaido 082-0081, Japan

<sup>3</sup> Obihiro University of Agriculture and Veterinary Medicine, Nishi 2-11, Inada, Obihiro, Hokkaido 080-8555, Japan

Fusarium head blight (FHB) is one of the most devastating diseases of wheat (*Triticum aestivum* L.), and the development of cultivars with FHB resistance is the most effective way to control the disease. Yumechikara is a Japanese hard red winter wheat cultivar that shows moderate resistance to FHB with superior bread-making quality. To identify quantitative trait loci (QTLs) for FHB resistance in Yumechikara, we evaluated doubled haploid lines derived from a cross between Yumechikara and a moderate susceptible cultivar, Kitahonami, for FHB resistance in a 5-year field trial, and we analyzed polymorphic molecular markers between the parents. Our analysis of these markers identified two FHB-resistance QTLs, one from Yumechikara and one from Kitahonami. The QTL from Yumechikara, which explained 36.4% of the phenotypic variation, was mapped on the distal region of chromosome 1BS, which is closely linked to the low-molecular-weight glutenin subunit gene *Glu-B3* and the glume color gene *Rg-B1*. The other QTL (from Kitahonami) was mapped on chromosome 3BS, which explained 11.2% of the phenotypic variation. The close linkage between the FHB-resistance QTL on 1BS, *Glu-B3* and *Rg-B1* brings an additional value of simultaneous screening for both quality and FHB resistance using the glume color.

**Key Words:** Fusarium head blight, wheat, resistance, quantitative trait loci.

## Introduction

Fusarium head blight (FHB; also known as scab disease) is caused mainly by *Microdochium nivale* and *Fusarium graminearum* in Tokachi area, Hokkaido. FHB is one of the most devastating fungal diseases of wheat (*Triticum aestivum* L.) (McMullen *et al.* 1997). Besides causing losses in grain yield and quality, *F. graminearum* leads to severe and harmful contamination of the grain with fungal toxins. FHB infection is favored by warm and humid climates during the flowering and maturing stages, and upward trends during growing-season temperatures are the cause of some concern in wheat-producing regions.

Host plant resistance is the most effective way to control FHB, and breeding for improved FHB resistance has thus become one of the most important targets of wheat breeding programs. Even though a number of studies have shown that the inheritance of FHB resistance is quantitative in nature,

the goal for wheat breeders is to create cultivars that are regionally adapted for high yield and quality with FHB resistance. Fortunately, large genetic variations for FHB resistance in the world wheat gene pool have been reported, and several quantitative trait loci (QTL) for FHB resistance were mapped on all wheat chromosomes (Buerstmayr *et al.* 2013).

Currently, one of the most repeatable QTLs for FHB resistance is reported on chromosome 3BS (*Fhb1*, *Qfhs.ndsu-3BS*) (Anderson *et al.* 2001, Bai *et al.* 1999, Buerstmayr *et al.* 2002, Cuthbert *et al.* 2006, Pumphrey *et al.* 2007, Waldron *et al.* 1999), in addition, a nearly perfect DNA marker *Umn 10* for *Fhb1* (Liu *et al.* 2008) has been developed, and significant progress regarding FHB resistance has also been accomplished in a hard red spring wheat cultivar ‘Alsen’ (Bakhsha *et al.* 2013, Frohberg *et al.* 2006) that possesses *Fhb1*.

Other QTLs are mapped on chromosomes 5AS (*Fhb5*, *Qfhs.ifa-5A*, *Qfhi.nau-5A*) (Buerstmayr *et al.* 2003, Lin *et al.* 2006, Xue *et al.* 2011), 6BS (*Fhb2*) (Anderson *et al.* 2001, Cuthbert *et al.* 2007) and 4BL (*Fhb4*, *Qfhi.nau-4B*) (Somers *et al.* 2003, Xue *et al.* 2010) from Asian spring wheat sources, and 1BL (*Qfhs.lfl-1BL*) (Häberle *et al.*

Communicated by Hisashi Tsujimoto

Received June 6, 2016. Accepted July 4, 2016.

First Published Online in J-STAGE on August 3, 2016.

\*Corresponding author (e-mail: zn206185@nodai.ac.jp)

2009), 6AL (*Qfhs.lfl-6AL*) and 7BS (*Qfhs.lfl-7BS*) (Häberle *et al.* 2007, Schmolke *et al.* 2005) from European winter wheat cultivars.

It was demonstrated that the pyramiding of minor to moderate FHB resistance QTLs also provides enhanced resistance to FHB (Miedaner *et al.* 2006). A higher number of QTLs with minor to moderate effects have been detected in several European and North American winter wheat populations, and they were mapped on several chromosomes (Buerstmayr *et al.* 2009, 2013). Most of those QTLs showed minor to moderate effects on FHB resistance, depending on their genetic backgrounds. Nevertheless, they would be useful for breeding due to their improved agricultural traits compared to exotic FHB resistant sources. FHB-resistant elite wheat cultivars like ‘Glenn’ and ‘Parshall’ were reported not to possess *Fhb1*, which indicates that the use of new sources of FHB resistance is still meaningful for practical breeding (EIDoliefy 2015, Rosyara *et al.* 2009). Thus, the exploration of new genetic sources is still necessary to enhance the genetic diversity of FHB resistance.

‘Yumechikara’ is a hard red winter cultivar with wheat yellow mosaic resistance, winter hardiness and favorable bread-making quality that was developed in Hokkaido, Japan (Ito *et al.* 2015, Kojima *et al.* 2015, Tabiki *et al.* 2011, Terasawa *et al.* 2016). Repeated evaluations of the FHB resistance in Yumechikara proved its stable resistance to FHB throughout a 5-year nursery trial. Hence, an exploration of the FHB resistance in Yumechikara may provide additional sources of FHB resistance for wheat breeding. The objective of the present study was to clarify novel QTLs for FHB resistance in the advanced winter wheat bread-making cultivar Yumechikara.

## Materials and Methods

### Plant materials

We developed F<sub>1</sub>-derived doubled haploid (DH) lines from the cross of ‘Yumechikara/Kitahonami’. Yumechikara (Satsukei 159/KS 831957//Kitanokaori) and Kitahonami (Kitamoe/Kitakei 1660), which is a soft red winter wheat cultivar (Yanagisawa *et al.* 2007). The cross was made in 2003, and DH lines were developed in 2004 from field-grown F<sub>1</sub> plants using the maize pollination procedure (Inagaki and Tahir 1990).

### Evaluation of FHB resistance

We evaluated their FHB resistance of 94 DH lines in a 5-year field trial (2008–2012) at the NARO Hokkaido Agricultural Research Center (Memuro, Japan) (42°53′ N, 143°05′ E). The soil type was volcanic ash soil. All DH lines and the parents were sown in mid-September. In each field trial, the experimental units contained 20 plants in a single 0.5-m row, spaced 0.72 m from the adjacent row with two replications. The plots received roughly 40 kg N ha<sup>-1</sup> at the seeding stage in the fall and an additional 60 kg N ha<sup>-1</sup> after the April snow melt. Inoculum was prepared using

*F. graminearum* isolated in Hokkaido (Nishio *et al.* 2008), and colonized oat grain was incubated for 2 weeks under room temperature. Approx. 20 L/100 m<sup>2</sup> of oat grain was spread before the flowering of the DH population. A gentle mist was applied with overhead sprinklers every hour for 10 mins (from 6:00 am to 6:00 pm) to maintain high humidity from the day of the first flowering of the materials until all of the materials were scored for FHB severity. The amount of precipitation by simulated rainfall was estimated to be approx. 10 mm/day.

At 2 weeks after flowering, we scored the plants’ FHB severity on a scale of 0 to 9 per 20 plants as follows: 0 = no damage; 2 = 1–3 spikelets diseased (20%); 4 = 4–5 spikelets diseased (40%); 6 = 6–7 spikelets diseased (60%); 8 = 8–9 spikelets diseased (80%); 9 = almost all or all spikelets diseased (90% or more) based on the reported index (Ban and Suenaga 2000), and then mean scores were calculated. All tests were conducted with two replicates of each line for each year, but in 2008 the test was conducted without replication.

### Molecular marker analysis

Leaf tissue (2 g) was ground in a mortar with liquid nitrogen until powdered, and then 10 ml of extraction buffer (1.5% cetyltrimethylammonium bromide [CTAB], 75 mM Tris, pH = 8.0, 15 mM Na<sub>2</sub>EDTA, 1.05 M NaCl) was added. The resultant slurry was incubated at 65°C for 30 min (Murray and Thompson 1980). DNA was purified in 24:1 chloroform/isoamyl alcohol, precipitated from the aqueous phase by the addition of isopropanol and then washed in 70% ethanol. DNA samples were dissolved in distilled water, and the DNA concentration was determined by measuring the optical density at 260 nm.

We screened 1,224 microsatellite (SSR) markers based on the maps of Gupta *et al.* (2002), Guyomarc’h *et al.* (2002), Pestsova *et al.* (2000), Röder *et al.* (1998), Somers *et al.* (2004), Song *et al.* (2005), Sourdille *et al.* (2001) and Torada *et al.* (2006) for polymorphism between the parental cultivars Yumechikara and Kitahonami. A total of 265 primer pairs amplified one or more polymorphic bands between the parents; of these, 224 markers were used to assemble linkage groups using the mapping population.

We conducted a polymerase chain reaction (PCR) in 10- $\mu$ l volumes using a GeneAmp PCR System 9700 thermal cycler (Applied Biosystems, Foster City, CA, USA). The amplification program began with 1 cycle of primary denaturation at 94°C for 5 min, followed by 42 cycles of 94°C for 1 min of denaturing, 51°C, 55°C or 61°C for annealing and 72°C for extension. One additional cycle was performed at 72°C for 5 min for the final elongation of the PCR product. Each 10- $\mu$ l PCR reaction contained 10 ng of template DNA, 1  $\times$  PCR mix, 1.7 mM MgCl<sub>2</sub>, 200 nM of each primer, 200  $\mu$ M of each dNTPs and 0.2 U *Taq* polymerase (Promega, Madison, WI). The PCR products were separated on a Sub-Cell<sup>®</sup> Model 96/192 (Bio-Rad, Hercules, CA) electrophoresis apparatus with 4% agarose gels. The gels

were run in  $1 \times$  TBE at a constant 100 V for 2 hr and then visualized after staining with ethidium bromide for 30 min.

### Mapping and QTL analysis

We used MAPMAKER/Exp v3.0b (Lander *et al.* 1987) to construct the wheat microsatellite map. The Kosambi function was applied to convert recombination fractions into map distances (Kosambi 1944). Logarithm of odds (LOD) thresholds of 3 and a distance threshold of 35 cM were used in the ordering and grouping of markers. We analyzed the QTLs for FHB resistance by performing composite interval mapping (CIM) using Windows QTL Cartographer ver. 2.5 with model 6 of the Zmapqtl procedure (Basten *et al.* 1996, Wang *et al.* 2006). For each analysis, genome-wide thresholds of LOD scores for significant ( $P < 0.05$ ) QTLs were determined by 1,000 permutations, and the significance of the detected QTLs was confirmed.

## Results

### Phenotypic evaluation of FHB response

The flowering date of parents and the mean temperatures during FHB development (from the first flowering of the DH lines to 2 weeks after the last flowering of the DH lines) in the 5-year field experiments are shown in **Table 1**. The mean values of the FHB disease index of the DH lines and their parental cultivars for the 5-year tests are shown in **Fig. 1**. The resistant parent Yumehikara showed a mean FHB severity of 3.6, ranging from 3.3 to 4.4 across the tested years, whereas the susceptible parent Kitahonami showed a mean FHB severity of 5.4, ranging from 4.8 to 6.3. The mean FHB severity of all DH lines was 5.1 and ranged from 4.5 to 5.5 across the tested years, and the results indicated a continuous variation of FHB severity with a single peak (**Fig. 1**).

Yumehikara showed the most extreme disease-resistance index among the progeny lines except for 2010, whereas transgressive segregation was observed in the susceptible lines, suggesting that the susceptible parent might contribute to resistance QTLs in the population. The ANOVA results indicated highly significant variations in genotypes, tested years, and genotype by tested year interactions (**Table 1**).

**Table 1.** Flowering dates of parents, and mean temperatures during FHB development in the 5-year field experiments

Year	Flowering date		Mean temperatures during FHB development <sup>1)</sup> (°C)
	Yumehikara	Kitahonami	
2008	17-Jun	18-Jun	15.7
2009	15-Jun	15-Jun	15.6
2010	21-Jun	21-Jun	20.4
2011	16-Jun	17-Jun	17.2
2012	18-Jun	19-Jun	16.6

<sup>1)</sup> From the first flowering of the DH lines to 2 weeks after the last flowering of the DH lines. The starting date of flowering for the DH lines was the same as Yumehikara, and the finishing date of flowering for the DH lines was about 2 days after the flowering of Kitahonami.

The correlation coefficients of the FHB disease index between the DH lines were all significant during the tested years, as was the combined mean (**Table 2**).

### Linkage map construction of the DH population

We used 224 markers to assemble the linkage groups in the mapping population of DH lines. The obtained linkage map contained 32 linkage groups covering a total genetic distance of 2254.3 cM in the DH population. The chromosome identity for the linkage groups was based on anchor microsatellite markers (Röder *et al.* 1998) and the wheat consensus map (Somers *et al.* 2004).

### Effects of QTL on FHB resistance

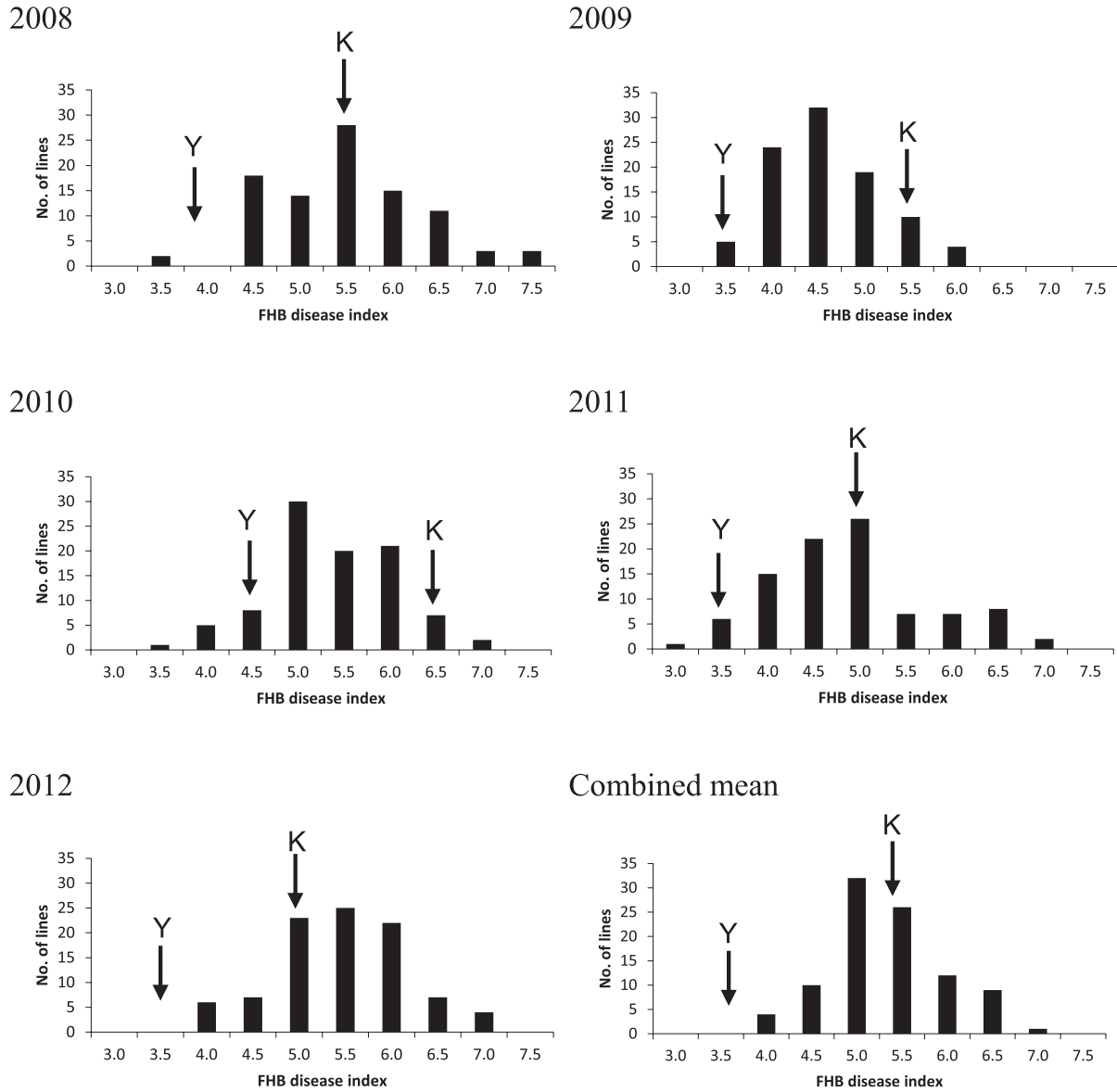
Our QTL analysis revealed that a distal region of the short arm of chromosome 1B of Yumehikara was significantly associated with FHB resistance through all five tested years (**Fig. 2**, **Table 3**). The FHB-resistance QTL peak of Yumehikara was mapped between the low-molecular weight glutenin subunit gene *Glu-B3* and the SSR marker *Xbarc32*, and the QTL region of approx. 10 cM included the wheat glume color gene *Rg-B1* (Khlestkina *et al.* 2006) (**Fig. 2**). The phenotypic variations of the FHB disease index explained by the QTL ranged from 23.9% to 31.8%, and that for the combined mean was 36.4% over the 5-year trial (**Table 3**).

Another QTL for FHB resistance was detected on chromosome 3B of the susceptible parent cultivar ‘Kitahonami’. The putative resistance QTL for FHB in Kitahonami was located between the SSR markers *Xgwm389* and *Xwmc754*, and the explained phenotypic variation ranged from 2.9% to 12.7%, and that for the combined mean was 11.2%; the effects of this QTL varied among the years as the LOD score was not significant in 2008, 2010 or 2011 (**Table 3**). The effect of the QTL on chromosome 3BS was smaller than that of the QTL on chromosome 1BS through all five tested years.

## Discussion

For practical breeding, the identification and utilization of FHB resistance in advanced wheat lines are important to achieve improved FHB management. Several QTLs for FHB resistance have been reported from Asian FHB-resistant spring wheat gene pools (Anderson *et al.* 2001, Bai *et al.* 1999, Buerstmayr *et al.* 2002, Waldron *et al.* 1999). Nevertheless, the use of those FHB-resistant sources sometimes corrupts important traits like winter hardiness and bread-making quality. Thus, the exploration, validation and use of new FHB-resistant sources from elite cultivars are valuable for practical breeding.

In this study, Yumehikara showed stable resistance compared to Kitahonami by serial 5-year field trials, and a significant FHB-resistant QTL was identified on the distal region of chromosome 1BS from the resistant parent Yumehikara. Our results indicated that the expression of



**Fig. 1.** The frequency distribution of the mean FHB disease index values in the doubled haploid lines derived from ‘Yumechikara/Kitahonami’ and their parents in the 5-year field experiments. Y; Yumechikara, K; Kitahonami.

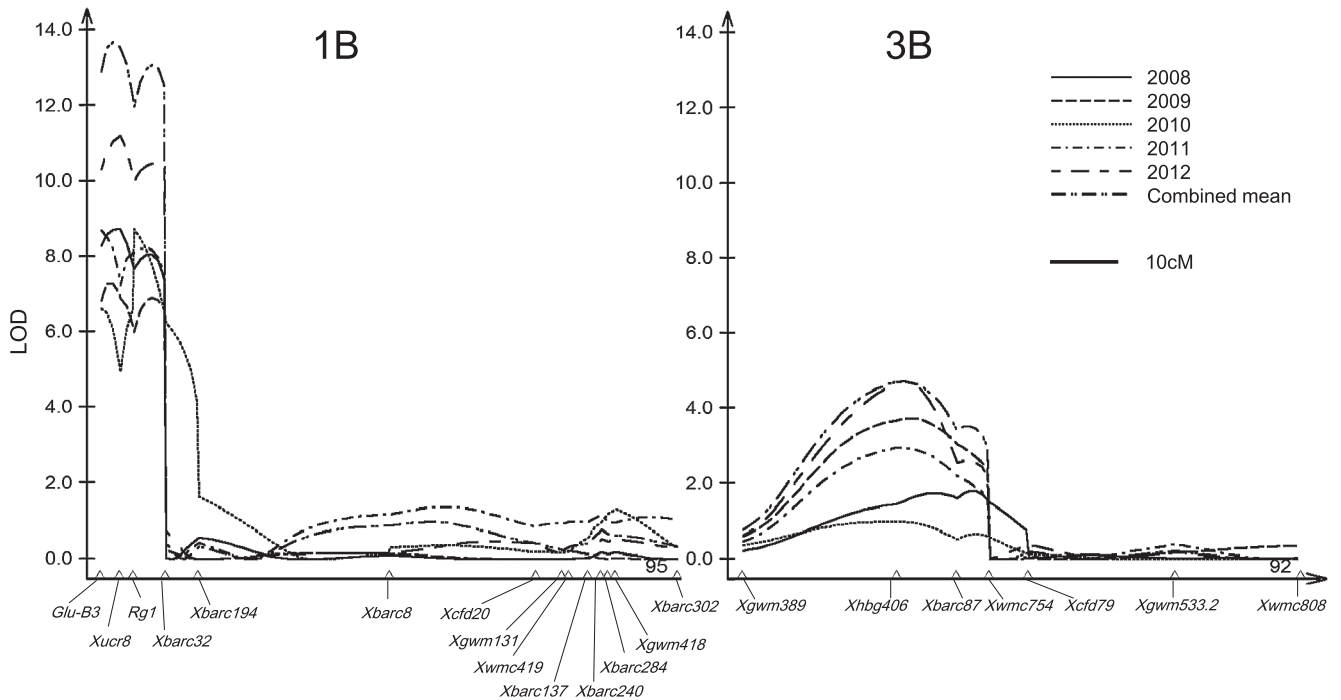
**Table 2.** Correlation coefficients between the FHB disease index of the 5-year field experiments and the combined mean of all tested years

Year	2008	2009	2010	2011	2012
2009	0.477***	–			
2010	0.719***	0.478***	–		
2011	0.631***	0.426***	0.646***	–	
2012	0.708***	0.536***	0.715***	0.695***	–
Combined mean	0.863***	0.683***	0.858***	0.843***	0.883***

\*\*\* P < 0.001.

the FHB-resistance QTL is rather stable under varied climate conditions. Out of 52 studies reporting a QTL for FHB resistance in wheat, nine studies reported QTLs on chromosome 1B (Buerstmayr *et al.* 2009), including T1BL.1RS translocation derived from the rye chromosome.

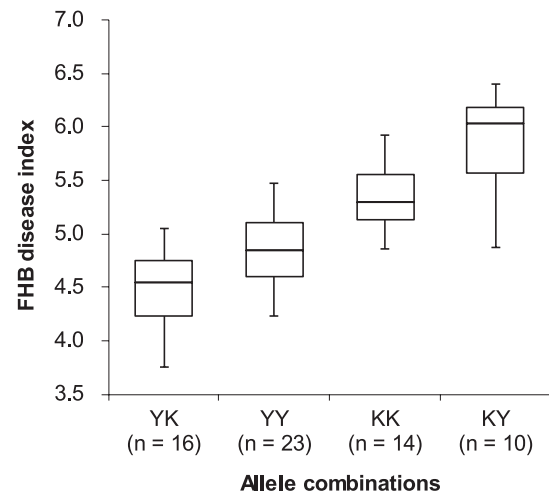
The FHB-resistance QTL has been mapped on the long arm of wheat chromosome 1B (*Qfhs.lfl-1BL*) in European winter wheat Cansas (Häberle *et al.* 2009), the CIMMYT breeding line CM-82036 (Buerstmayr *et al.* 2002), Chinese land race Wangshuibai (Zhou *et al.* 2004), the CIMMYT cultivar Seri 28 (Mardi *et al.* 2006), and the Swiss cultivar Arina (Semagn *et al.* 2007), whereas the rest of the QTLs are mapped on the rye chromosome of T1BL.1RS translocation (Ittu *et al.* 2000, Schmolke *et al.* 2005, Shen *et al.* 2003, Zhang *et al.* 2004). Since both parents Yumechikara and Kitahonami do not possess T1BL.1RS translocation (data not shown), we propose that the QTL of Yumechikara is a novel FHB-resistance QTL in winter wheat. From the consensus map of wheat (Somers *et al.* 2004), it is apparent that the *Glu-B3* gene is located 5 cM from the distal end of the



**Fig. 2.** Maps of quantitative trait loci (QTL) for FHB resistance constructed from doubled haploid lines derived from ‘Yumechikara/Kitahonami’ based on the 5-year field experiments.

**Table 3.** Flanking markers, logarithm of the odds (LOD), coefficients of determination ( $R^2$ ), and additive effect of the significant QTL regions detected by the composite interval mapping based on the 5-year field experiments

Years	Resistance donor Locus Flanking markers	Yumechikara 1BS <i>Glu-B3-Xbarc32</i>	Kitahonami 3BS <i>Xgwm389-Xwmc754</i>	Threshold of LOD scores
2008	LOD	8.71	1.76	3.61
	$R^2$ (%)	28.0	5.3	–
	Additive effect	–0.454	0.197	–
2009	LOD	7.27	3.72	3.73
	$R^2$ (%)	23.9	12.4	–
	Additive effect	–0.327	0.228	–
2010	LOD	8.73	0.95	3.03
	$R^2$ (%)	28.6	3.0	–
	Additive effect	–0.386	0.122	–
2011	LOD	8.69	2.95	3.39
	$R^2$ (%)	26.0	8.9	–
	Additive effect	–0.480	0.279	–
2012	LOD	11.15	4.69	2.7
	$R^2$ (%)	31.8	12.7	–
	Additive effect	–0.429	0.271	–
Combined mean	LOD	13.67	4.69	3.79
	$R^2$ (%)	36.4	11.2	–
	Additive effect	–0.396	0.215	–



**Fig. 3.** Effects of the four different combinations of QTLs for the FHB disease index in the doubled haploid lines derived from ‘Yumechikara/Kitahonami’ based on the 5-year field experiments. YK carries the resistance alleles at QTLs on 1BS and 3BS. YY carries a resistance allele at QTL-1BS. KK carries a resistance allele at QTL-3BS. KY carries none of the resistance QTLs. The lines in which all genotypes are the same between flanking markers (*GluB3-barc32* for 1BS and *gwm384-wmc754* for 3BS) were selected.

chromosome 1BS, and there are no previous reports of an FHB-resistance QTL on such a distal end region of wheat chromosome 1BS.

Wheat glume color is controlled by the three homoeologous major genes *Rg-A1*, *Rg-B1*, and *Rg-D1* located on

chromosomes 1A, 1B, and 1D, respectively (Khlestkina *et al.* 2006). Yumechikara has the *Rg-B1b* allele on chromosome 1B that shows red glume color, whereas Kitahonami has *Rg-B1a* with white glume color. The peak of the FHB resistance QTL from Yumechikara included the flanking

region of the glume color gene *Rg-B1* (Fig. 2), indicating that the glume color should be a useful phenotypic marker to screen FHB-resistant lines. In the present study, the FHB resistance was scored after two weeks of flowering when the glume coloration is still in the beginning stage, and thus the FHB disease index scoring was not influenced by the glume color.

On the distal region of the short arm of chromosome 1B, the low-molecular-weight glutenin subunit gene *Glu-B3* was also located close to the peak of the FHB-resistance QTL. Based on our analysis of the *Glu-B3* alleles for the pedigree lines of Yumehikara (KS 831957/Satsukei 159//Kitanokaori), we speculate that the source of the FHB-resistance QTL is the former major cultivar Takunekomugi, a parental line of Satsukei 159, which is derived from the native winter wheat gene pools, because only Takunekomugi and its descendants including Yumehikara possess the *Glu-B3ab* (*b\**) allele.

It has been reported that cosegregation of the wheat glume color gene *Rg-B1* and the low-molecular-weight glutenin subunit gene *Glu-B3* can be applied as a useful ‘field-marker’ for desired gluten strength (Fujii *et al.* 2011). Since Yumehikara is characterized as extra-strong-quality wheat that possesses a combination of the high-molecular-weight glutenin subunit allele *Glu-D1d* and low-molecular-weight glutenin subunit allele *Glu-B3ab* (*b\**) (Ito *et al.* 2015), this close linkage between the FHB-resistance QTL, *Glu-B3* and *Rg-B1* brings an additional value of simultaneous screening for both quality and FHB resistance improvement.

The FHB-resistance QTL from the susceptible parent Kitahonami was mapped on the short arm of chromosome 3B, the nearby *Fhb1* region between *Xgwm389* and *Xwmc754*. The effect of combinations of the two FHB-resistance QTLs from both Yumehikara and Kitahonami were validated among the DH lines in which all genotypes are the same between flanking markers (*GluB3-barc32* for 1BS and *gwm384-wmc754* for 3BS) (Fig. 3). The results indicated an obvious additive effect by the combination of two FHB-resistance QTLs derived from both parental cultivars Yumehikara and Kitahonami (YK type) with the lowest FHB disease index, whereas the DH lines with two FHB-susceptible QTLs from both parents (the KY type) showed the most severe FHB disease index (Fig. 3). The DH lines with each parental-type QTLs (YY and KK types) showed FHB disease index values that were intermediate and similar to their respective parents. In this study, most of the DH lines showed higher FHB severity compare to Yumehikara (Fig. 1), and minor QTLs that did not show significant LOD scores in all field trials were detected on chromosomes 2A, 6D and 7D from Yumehikara. We speculate that the reason for the higher FHB severity in DH lines compare to resistant parent Yumehikara is the absence of those minor QTLs. However, the number of markers in the minor QTLs is still small and further analysis is necessary to elucidate the location and interactions between them.

Among the five tested years, the effect of FHB-resistance

QTLs was the least in 2010, when the mean temperature between June and August in Hokkaido was the highest since the start of recorded observations in 1946, and the mean number of days from heading to maturation was shortest at only 37 days, which brought about a historically low yield (Nishio *et al.* 2013). In 2010, both parents showed a higher FHB disease index than the other years, which might be one of the reasons for the lower effects of the FHB resistance QTL (Fig. 1). The correlation coefficient between the mean temperature from heading to maturing and the mean FHB disease index was not significant (data not shown); nevertheless, the unusual high temperature during the FHB disease occurrence in 2010 might have contributed to the lower effect of the FHB-resistance QTLs.

In this study, we clarified a novel FHB-resistance QTL on chromosome 1BS that is closely linked to both the glume color gene *Rg-B1* and the low-molecular-weight glutenin subunit gene *Glu-B3*, and a minor to moderate FHB-resistance QTL on chromosome 3BS. To our knowledge, there have been no reports on the relationship between FHB resistance and glume color in wheat, and the QTL could be a potentially new FHB-resistance source. Further study is necessary to confirm whether the glume color may have a certain effect on *Fusarium* infection into the spike.

### Acknowledgements

We thank A. Hayata, S. Takahashi, T. Kobayashi, K. Odajima, and M. Oizumi for their technical assistance.

### Literature Cited

- Anderson, J.A., R.W. Stack, S. Liu, B.L. Waldron, A.D. Fjeld, C. Coyne, B. Moreno-Sevilla, J. Mitchell Fetch, Q.J. Song, P.B. Cregan *et al.* (2001) DNA markers for *Fusarium* head blight resistance QTLs in two wheat populations. *Theor. Appl. Genet.* 102: 1164–1168.
- Bai, G.H., F.L. Kolb, G.E. Shaner and L.L. Domier (1999) Amplified fragment length polymorphism markers linked to a major quantitative trait locus controlling scab resistance in wheat. *Phytopathology* 89: 343–348.
- Bakhsh, A., N. Mengistu, P.S. Baenziger, I. Dweikat, S.N. Wegulo, D.J. Rose, G. Bai and K.M. Eskridge (2013) Effect of *Fusarium* head blight resistance gene *Fhb1* on agronomic and end-use quality traits of hard red winter wheat. *Crop Sci.* 53: 793–801.
- Ban, T. and K. Suenaga (2000) Genetic analysis of resistance to *Fusarium* head blight caused by *Fusarium graminearum* in Chinese wheat cultivar Sumai 3 and the Japanese cultivar Saikai 165. *Euphytica* 113: 87–99.
- Basten, C.J., B.S. Weir and Z.-B. Zeng (1996) QTL Cartographer: A reference manual and tutorial for QTL mapping. Department of Statistics, North Carolina State University, Raleigh, NC.
- Buerstmayr, H., M. Lemmens, L. Hartl, L. Doldi, B. Steiner, M. Stierschneider and P. Ruckebauer (2002) Molecular mapping of QTLs for *Fusarium* head blight resistance in spring wheat. I. Resistance to fungal spread (type II resistance). *Theor. Appl. Genet.* 104: 84–91.
- Buerstmayr, H., B. Steiner, L. Hartl, M. Grisser, N. Angerer, D. Lengauer, T. Miedaner, B. Schneider and M. Lemmens (2003) Molecular

- mapping of QTLs for Fusarium head blight resistance in spring wheat. II. Resistance to fungal penetration and spread. *Theor. Appl. Genet.* 107: 503–508.
- Buerstmayr, H., T. Ban and J.A. Anderson (2009) QTL mapping and marker-assisted selection for *Fusarium* head blight resistance in wheat: a review. *Plant Breed.* 128: 1–26.
- Buerstmayr, H., M. Buerstmayr, W. Schweiger and B. Steiner (2013) Genomics-assisted breeding for Fusarium head blight resistance in wheat. *In: Varshney, R.K. and R. Tuberosa (eds.) Translational Genomics for Crop Breeding: Biotic Stress, Volume 1*, Wiley, London, pp. 45–61.
- Cuthbert, P.A., D.J. Somers, J. Thomas, S. Cloutier and A. Brule-Babel (2006) Fine mapping *Fhb1*, a major gene controlling Fusarium head blight resistance in bread wheat (*Triticum aestivum* L.). *Theor. Appl. Genet.* 112: 1465–1472.
- Cuthbert, P.A., D.J. Somers and A. Brule-Babel (2007) Mapping of *Fhb2* on chromosome 6BS: a gene controlling Fusarium head blight field resistance in bread wheat (*Triticum aestivum* L.). *Theor. Appl. Genet.* 114: 429–437.
- EiDoliefy, A.E.A. (2015) Molecular mapping of Fusarium head blight resistance in two adapted spring wheat cultivars. North Dakota State University Ph. D. dissertation. pp. 1–92.
- Frohberg, R.C., R.W. Stack, T. Olson, J.D. Miller and M. Mergoum (2006) Registration of ‘Alsen’ wheat. *Crop Sci.* 46: 2311–2312.
- Fujii, K., T. Tsuji, T. Yoshida, W. Maruyama-Funatsuki and T. Ikeda (2011) Applicability of ‘glume color’ controlled by *Rg-B1* locus as a field-marker to detect *Glu-B3* alleles encoding low-molecular-weight glutenin subunits in common wheat. *Breed. Sci.* 61: 11–16.
- Gupta, P., H. Balyan, K. Edwards, P. Isaac, V. Korzun, M. Röder, M.-F. Gautier, P. Joudrier, A. Schlatter, J. Dubcovsky *et al.* (2002) Genetic mapping of 66 new microsatellite (SSR) loci in bread wheat. *Theor. Appl. Genet.* 105: 413–422.
- Guyomarc’h, H., P. Sourdille, G. Charmet, K.J. Edwards and M. Bernard (2002) Characterisation of polymorphic microsatellite markers from *Aegilops tauschii* and transferability to the D-genome of bread wheat. *Theor. Appl. Genet.* 104: 1164–1172.
- Häberle, J., M. Schmolke, G. Schweizer, V. Korzun, E. Ebmeyer, G. Zimmermann and L. Hartl (2007) Effects of two major *Fusarium* head blight resistance QTL verified in a winter wheat backcross population. *Crop Sci.* 47: 1823–1831.
- Häberle, J., J. Holzapfel, G. Schweizer and L. Hartl (2009) A major QTL for resistance against Fusarium head blight in European winter wheat. *Theor. Appl. Genet.* 119: 325–332.
- Inagaki, M.N. and M. Tahir (1990) Comparison of haploid production frequencies in wheat varieties crossed with *Hordeum bulbosum* L. and maize. *Japan. J. Breed.* 40: 209–216.
- Ito, M., W. Maruyama-Funatsuki, T.M. Ikeda, Z. Nishio, K. Nagasawa and T. Tabiki (2015) Dough properties and bread-making quality-related characteristics of Yumehikara near-isogenic wheat lines carrying different *Glu-B3* alleles. *Breed. Sci.* 65: 241–248.
- Ittu, M., N.N. Saulescu, I. Hagima, G. Ittu and P. Mustatea (2000) Association of Fusarium head blight resistance with gliadin loci in a winter wheat cross. *Crop Sci.* 40: 62–67.
- Khlestkina, E.K., T.A. Pshenichnikova, M.S. Röder, E.A. Salina, V.S. Arbutova and A. Börner (2006) Comparative mapping of genes for glume colouration and pubescence in hexaploid wheat (*Triticum aestivum* L.). *Theor. Appl. Genet.* 113: 801–807.
- Kojima, H., Z. Nishio, F. Kobayashi, T. Sasaya, C. Kiribuchi-Otobe, M. Seki, S. Oda, M. Saito and T. Nakamura (2015) Identification and validation of a quantitative trait locus associated with wheat yellow mosaic virus pathotype I resistance in a Japanese wheat variety. *Plant Breed.* 134: 373–378.
- Kosambi, D.D. (1944) The estimation of map distance from recombination values. *Ann. Eugen.* 12: 172–175.
- Lander, E.S., P. Green, J. Abrahamson, A. Barlow, M.J. Daly, S.E. Lincoln and L. Newburg (1987) MAPMAKER: an interactive computer package for constructing primary genetic linkage maps of experimental and natural populations. *Genomics* 1: 174–181.
- Lin, F., S.L. Xue, Z.Z. Zhang, C.Q. Zhang, Z.X. Kong, G.Q. Yao, D.G. Tian, H.L. Zhu, C.J. Li, Y. Cao *et al.* (2006) Mapping QTL associated with resistance to Fusarium head blight in the Nanda2419 × Wangshuibai population. II: Type I resistance. *Theor. Appl. Genet.* 112: 528–535.
- Liu, S., M.O. Pumphrey, B.S. Gill, H.N. Trick, J.X. Zhang, J. Dolezel, B. Chalhouh and J.A. Anderson (2008) Toward positional cloning of *Fhb1*, a major QTL for Fusarium head blight resistance in wheat. *Cereal Res. Commun.* 36: 195–201.
- Mardi, M., L. Pazouki, H. Delavar, M.B. Kazemi, B. Ghareyazie, B. Steiner, R. Nolz, M. Lemmens and H. Buerstmayr (2006) QTL analysis of resistance to Fusarium head blight in wheat using a ‘Frontana’-derived population. *Plant Breed.* 125: 313–317.
- McMullen, M.P., R. Jones and D. Gallenberg (1997) Scab of wheat and barley: A re-emerging disease of devastating impact. *Plant Dis.* 81: 1340–1348.
- Miedaner, T., F. Wilde, B. Steiner, H. Buerstmayr, V. Korzun and E. Ebmeyer (2006) Stacking quantitative trait loci (QTL) for Fusarium head blight resistance from non-adapted sources in an European elite spring wheat background and assessing their effects on deoxynivalenol (DON) content and disease severity. *Theor. Appl. Genet.* 112: 562–569.
- Murray, M.G. and W.F. Thompson (1980) Rapid isolation of high molecular weight plant DNA. *Nucleic Acids Res.* 8: 4321–4325.
- Nishio, Z., M. Ito, M. Tanio, T. Tabiki and H. Yamauchi (2008) Microsatellite marker-assisted selection of Fusarium head blight resistance in backcrossed Japanese winter wheat lines. *Breed. Sci.* 58: 183–185.
- Nishio, Z., M. Ito, T. Tabiki, K. Nagasawa, H. Yamauchi and T. Hirota (2013) Influence of higher growing-season temperatures on yield components of winter wheat (*Triticum aestivum* L.). *Crop Sci.* 53: 621–628.
- Pestsova, E., M.W. Ganal and M.S. Röder (2000) Isolation and mapping of microsatellite markers specific for the D genome of bread wheat. *Genome* 43: 689–697.
- Pumphrey, M.O., R. Bernardo and J.A. Anderson (2007) Validating the *Fhb1* QTL for Fusarium head blight resistance in near-isogenic wheat lines developed from breeding populations. *Crop Sci.* 47: 200–206.
- Röder, M.S., V. Korzun, K. Wendehake, J. Plaschke, M.H. Tixier, P. Leroy and M.W. Ganal (1998) A microsatellite map of wheat. *Genetics* 149: 2007–2023.
- Rosyara, U.R., J.L. Gonzalez-Hernandez, K.D. Glover, K.R. Gedye and J.M. Stein (2009) Family-based mapping of quantitative trait loci in plant breeding populations with resistance to Fusarium head blight in wheat as an illustration. *Theor. Appl. Genet.* 118: 1617–1631.
- Schmolke, M., G. Zimmermann, H. Buerstmayr, G. Schweizer, T. Miedaner, V. Korzun, E. Ebmeyer and L. Hartl (2005) Molecular mapping of Fusarium head blight resistance in the winter wheat population Dream/Lynx. *Theor. Appl. Genet.* 111: 747–756.
- Semagn, K., H. Skinnnes, Å. Bjørnstad, A.G. Marøy and Y. Tarkegne (2007) Quantitative trait loci controlling Fusarium head blight resistance and low deoxynivalenol content in hexaploid wheat population from ‘Arina’ and NK93604. *Crop Sci.* 47: 294–303.

- Shen, X.R., M. Ittu and H.W. Ohm (2003) Quantitative trait loci conditioning resistance to Fusarium head blight in wheat line F201R. *Crop Sci.* 43: 850–857.
- Somers, D.J., G. Fedak and M. Savard (2003) Molecular mapping of novel genes controlling Fusarium head blight resistance and deoxynivalenol accumulation in spring wheat. *Genome* 46: 555–564.
- Somers, D.J., P. Isaac and K. Edwards (2004) A high-density microsatellite consensus map for bread wheat (*Triticum aestivum* L.). *Theor. Appl. Genet.* 109: 1105–1114.
- Song, Q.J., J.R. Shi, S. Singh, E.W. Fickus, J.M. Costa, J. Lewis, B.S. Gill, R. Ward and P.B. Cregan (2005) Development and mapping of microsatellite (SSR) markers in wheat. *Theor. Appl. Genet.* 110: 550–560.
- Sourdille, P., H. Guyomarc'h, C. Baron, B. Gandon, V. Chiquet, F. Artiguenave, K. Edwards, N. Foisset and P. Dufour (2001) Improvement of the genetic maps of wheat using new microsatellite markers. *Plant & Animal Genome IX, final abstracts guide*. Applied Biosystem Press, Foster City, CA, USA, p. 167.
- Tabiki, T., Z. Nishio, M. Ito, H. Yamauchi, K. Takata, T. Kuwabara, N. Iriki, M. Tanio, T. Ikeda and W. Funatsuki (2011) A new extra-strong hard red winter wheat variety : 'Yumechikara'. *Res. Bull. NARO Hokkaido Agric. Res. Cent.* 195: 1–12.
- Terasawa, Y., M. Ito, T. Tabiki, K. Nagasawa, K. Hatta and Z. Nishio (2016) Mapping of a major QTL associated with protein content on chromosome 2B in hard red winter wheat (*Triticum aestivum* L.). *Breed. Sci.* 66: 471–480.
- Torada, A., M. Koike, K. Mochida and Y. Ogihara (2006) SSR-based linkage map with new markers using an intraspecific population of common wheat. *Theor. Appl. Genet.* 112: 1042–1051.
- Waldron, B.L., B. Moreno-Sevila, J.A. Anderson, R.W. Stack and R.C. Frohberg (1999) RFLP mapping of QTL for Fusarium head blight resistance in wheat. *Crop Sci.* 39: 805–811.
- Wang, S., C.J. Basten and Z.B. Zeng (2006) Windows QTL cartographer 2.5. Department of Statistics, North Carolina State University, Raleigh, NC, USA.
- Xue, S., G. Li, H. Jia, F. Xu, F. Lin, M. Tang, Y. Wang, X. An, H. Xu, L. Zhang *et al.* (2010) Fine mapping *Fhb4*, a major QTL conditioning resistance to Fusarium infection in bread wheat (*Triticum aestivum* L.). *Theor. Appl. Genet.* 121: 147–156.
- Xue, S., F. Xu, M. Tang, Y. Zhou, G. Li, X. An, F. Lin, H. Xu, H. Jia, L. Zhang *et al.* (2011) Precise mapping *Fhb5*, a major QTL conditioning resistance to Fusarium infection in bread wheat (*Triticum aestivum* L.). *Theor. Appl. Genet.* 123: 1055–1063.
- Yanagisawa, A., Y. Yoshimura, Y. Amano, S. Kobayashi, T. Nishimura, K. Nakamichi, K. Araki, K. Tanifuji, T. Tabiki, K. Mikami *et al.* (2007) A new winter wheat variety "Kitahonami". *Bull. Hokkaido Pref. Agric. Exp. Sta.* 91: 1–13.
- Zhang, X., M.P. Zhou, L.J. Ren, G. Bai, H.X. Ma, O.E. Scholten, P.G. Guo and W.Z. Lu (2004) Molecular characterization of *Fusarium* head blight resistance from wheat variety Wangshuibai. *Euphytica* 139: 59–64.
- Zhou, W., F.L. Kolb, J. Yu, G. Bai, L.K. Boze and L.L. Domier (2004) Molecular characterization of Fusarium head blight resistance in Wangshuibai with simple sequence repeat and amplified fragment length polymorphism markers. *Genome* 47: 1137–1143.