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ORIGINAL PAPER

Fine mapping *TaFLW1*, a major QTL controlling flag leaf width in bread wheat (*Triticum aestivum* L.)

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Abstract
Introduction Flag leaf width (FLW) is directly related to photosynthetic capacity and yield potential in wheat. In a previous study, *Qflw.nau-5A* controlling FLW was detected on chromosome 5A in the interval possessing *Fhb5* for type I Fusarium head blight (FHB) resistance using a recombinant inbred line population derived from Nanda2419 × Wangshuibai.
Materials and methods *Qflw.nau-5A* near-isogenic line (NIL) with the background of Mianyang 99-323 and PH691 was developed and evaluated. FLW inheritance was investigated using two F_2 populations developed from crossing the *Qflw.nau-5A* NILs with their recurrent parents. One hundred ten and 28 recombinants, which included 10 and 5 types of recombinants, were identified from 2816 F_2 plants with Mianyang 99-323 background and 1277 F_2 plants with PH691 background, respectively, and phenotyped in field trials for FLW and type I FHB resistance. Deletion bin mapping was applied to physically map *Qflw.nau-5A*.
Results and conclusions The introduction of Wangshuibai *Qflw.nau-5A* allele reduced the FLW up to 3 mm. In the F_2 populations, *Qflw.nau-5A* was inherited like a semi-dominant gene, and was therefore designated as *TaFLW1*. The FLW of the recombinant lines displayed a distinct two-peak distribution. Recombinants with wider leaves commonly have Mianyang 99-323 or PH691 chromatin in the 0.2 cM *Xwmc492-Xwmc752* interval that resided in the 5AL12-0.35–0.57 deletion bin, and recombinants with narrow leaves were Wangshuibai genotype in this interval. Phenotypic recombination between FLW and type I FHB resistance was identified, implying *TaFLW1* was in close linkage with *Fhb5*. These results should aid wheat breeders to break the linkage drag through marker-assisted selection and assist in the map-based cloning of *TaFLW1*.

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Introduction

Bread wheat (*Triticum aestivum* L.) is one of the most important crops and is a staple food for about 40 % of the world population. Therefore, improvement of wheat yield is vital to relieve food shortages. Grain yield is a complex trait and is not only directly related to kernel weight, kernel number per spike and spike number per unit area, but also affected by other factors, such as accumulation and transport of photosynthetic products (Cui et al. 2003). The flag leaf contributes about 45–58 % of the total photosynthetic activity (Xu and Zhao 1995) and about 41–43 % of the carbohydrates for grain filling (Sharma et al. 2003). A number of studies showed that flag leaf size was positively associated with 1,000-grain weight, kernel number per spike, grain yield per plant or other yield-related traits in cereals such as rice and wheat (Cui et al. 2003; Mei et al. 2003; Khaliq et al. 2008; Wang et al. 2011). Optimizing

flag leaf morphology is expected to improve photosynthetic efficiency and enhance grain yield in cereal crops.

The morphological traits of flag leaves, including length, width and area are inherited quantitatively and influenced greatly by the environments (Coleman et al. 2001; Yue et al. 2006; Xue et al. 2008a). With the availability of molecular markers and genetic maps, a number of quantitative trait loci (QTL) for flag leaf morphology have been detected in rice and barley (Mei et al. 2003; Yue et al. 2006; Xue et al. 2008a; Farooq et al. 2010). In rice, *qFL1* associated with length and *qFLW4* associated with width of flag leaves have been fine mapped (Wang et al. 2011; Chen et al. 2012), and *Nal1* and *Nal7*, both associated with flag leaf width (FLW), have been cloned (Fujino et al. 2008; Qi et al. 2008). Considerable progress has already been made in genetic research on wheat yield and the various yield components (Kato et al. 2000; Groos et al. 2003; Quarrie et al. 2005; Zhang et al. 2010); however, reports on mapping QTLs associated with flag leaf morphology in wheat are still lacking.

In our previous study, QTL *Qflw.nau-5A* controlling FLW was detected in the *Xbarc303–Xbarc100* interval on chromosome 5A, which consistently explained over 25 % of the phenotypic variation in the Nanda2419 × Wangshuibai recombinant inbred line population (Ma et al. 2008; Jia et al. 2013). The Wangshibai allele of this QTL contributed to narrow flag leaves. This QTL interval in Wangshuibai also carries a major QTL, i.e., *Fhb5*, conditioning type I resistance to *Fusarium* head blight (FHB) (Lin et al. 2006; Xue et al. 2011). Development and evaluation of the *Fhb5* near-isogenic lines (NIL) in the background of Mianyang 99-323 and PH691 demonstrated that introduction of *Fhb5* significantly improved type I resistance and reduced the FLW up to 3 mm compared with the recurrent parents (Xue et al. 2010), suggesting linkage drag between FHB resistance and FLW.

In this study, we reported the fine mapping of *Qflw.nau-5A* using secondary F_2 populations created with its NILs in Mianyang 99-323 and PH691 backgrounds, and showed that *Qflw.nau-5A* was a semi-dominant gene located on the long arm of chromosome 5A tightly linked to *Fhb5*.

Materials and methods

Plant materials

Two NILs with the *Xbarc303–Xbarc100* interval of Wangshuibai covering *Fhb5* and *Qflw.nau-5A* were developed by marker-assisted backcross using susceptible wide-leaf cultivars Mianyang 99-323 and PH691 as the recurrent parents. Both NILs, no matter with the Mianyang 99-323 background (NIL/MY) or with the PH691 background (NIL/PH), were resistant to *Fusarium* infection (Xue et al. 2010) and had significantly narrower flag leaves compared with their recurrent parents (Fig. 1). The secondary F_2 populations were developed by crossing the NILs with the respective recurrent parents.

Fig. 1 Flag leaves of *Qflw.nau-5A* NILs and their recurrent parents Mianyang 99-323 and PH691. *Scale bar* 10 mm

Wheat cultivar 'Chinese Spring' (CS), its ditelosomic lines, as well as a set of deletion lines of chromosome 5A (Endo and Gill 1996) were used for chromosomal arm and bin assignments of molecular markers.

Genotyping, mapping and recombinant screening

Genomic DNA was extracted from young leaves according to Ma and Sorrells (1995). Polymerase chain reaction (PCR) was performed following the procedure of Ma et al. (1996). The PCR products were separated in 8 % non-denaturing polyacrylamide gels (acrylamide:bis-acrylamide = 19:1) at room temperature with 1 × TBE buffer and visualized by silver staining (Bassam et al. 1991).

All DNA markers that mapped to the *Qflw.nau-5A* interval flanked by *Xbarc303* and *Xbarc100* on chromosome 5A (Röder et al. 1998; Somers et al. 2004; Song et al. 2005; Lin et al. 2006; Xue et al. 2008b) were employed in genotyping for map construction. A linkage map was constructed with MAPMAKER/EXP Version 3.0 (Lincoln et al. 1992) using the Kosambi mapping function (Kosambi 1944) and drawn with MapDraw Version 2.1 (Liu and Meng 2003).

Plants with recombination occurring within the *Qflw.nau-5A* interval were identified using flanking markers *Xbarc303* and *Xbarc180* in the NIL/MY-derived secondary F_2 population and flanking markers *Xbarc56* and *Xbarc180* in the NIL/PH-derived secondary F_2 population. The selfed progenies of the identified plants were then genotyped for screening homozygous recombinants with

all the markers mapped in the QTL interval and polymorphic in the respective populations. The identified recombinants were self-pollinated to form recombinant lines.

FLW and FHB resistance evaluation

Evaluation of the parental NILs, recurrent parents, 132 F_2 plants derived from NIL/MY, 125 F_2 plants derived from NIL/PH and recombinants with the PH691 background was carried out in the 2012 season in a field of Nanjing Agricultural University (NAU) Jiangpu (JP) experimental station. The row spacing was 50 cm and 15 seeds were planted per row. At the early grain filling stage, the FLW of three to six tillers per plant was measured. The FLW mean of each plant was calculated to represent the phenotype value.

FLW evaluation of recombinants with the Mianyang 99-323 background, the parental NIL and recurrent parent was performed in the 2011–2012 growing season in a field house of Pailou (PL) and a field of JP experimental station of NAU in Jiangsu and in a field at Fengyang (FY) County of Anhui, using a randomized complete block design with two replicates. Seeds in the FY trial were sowed 20 days later than in the PL and JP trials. Each plot included one 1.4-m row in the PL trial and two 1.5-m rows in the JP and FY trials. The row spacing was 25 cm and 20 seeds were planted per row. Conventional field management was applied during the growth period. At the early grain filling stage, the main tiller FLW of 10 plants randomly chosen from each plot (border plants excluded) was measured. The FLW mean of each plot was calculated to represent the phenotype value.

The FLW measurements (mm) were taken at the widest part of the flag leaf in all these trials.

To evaluate the FHB resistance of NIL/MY-derived recombinants, inoculation was carried out 2 weeks before anthesis through scattering the scabby corn grains fully infected with four locally isolated highly virulent strains F4, F15, F34 and F7136 on the soil surface in the JP and FY trials. Fifteen days after the anthesis, the total number of spikelets and the number of spikelets with visible disease symptom in ten randomly chosen spikes from each recombinant line were counted and the percentage of diseased spikelets (PDS) was calculated.

Statistical analysis

Statistical analysis was performed using software Data Desk v. 5.0 (Data Description, Inc., Ithaca, NY, USA). Dunnett's test or Student's *t* test was used for recombinant-versus-control comparisons and Tukey's HSD test was applied for multiple mean comparisons. Analysis of variance (ANOVA) across the environments was carried out using the mixed linear model function of the software package QTModel (http://ibi.zju.edu.cn/software/qtmodel), in which genotypes were specified as fixed effects, whereas environments, blocks within environments and genotype × environment interactions were specified as random effects. The line-based broad sense heritability across environments was estimated using the formula:

$$h^2 = \sigma_g^2 / \left(\sigma_g^2 + \sigma_{g\times e}^2 / n + \sigma_e^2 / nr \right),$$

where σ_g^2 is the genetic variance, $\sigma_{g\times e}^2$ is the variance from genotype × environment interaction, σ_e^2 is the experimental error, n is the number of environments and r is the number of replicates in each environment.

Results

Inheritance of *Qflw.nau-5A*

To find out the inheritance mode of *Qflw.nau-5A*, 132 NIL/MY-derived F_2 plants and 125 NIL/PH-derived F_2 plants were evaluated for FLW and genotyped with *Xgwm415* that falls in the *Qflw.nau-5A* interval. The FLW displayed a continuous distribution in both populations (Fig. 2). Based on the marker genotype at *Xgwm415*, the F_2 plants in each population were classified into three groups, including Wangshuibai type (WW), Mianyang 99-323 type (MM) or PH691 genotype type (PP), and heterozygous type WM or WP. As shown in Fig. 2, the FLW distribution of plants in the WW group did not overlap with the distribution of plants in the MM and PP group, but the FLW distribution of the plants in the heterozygous groups overlapped with both parental types. The FLW means of the WM and WP groups were significantly larger than the WW group, but significantly smaller than the MM or PP group (Table 1). These results suggested that *Qflw.nau-5A* was semi-dominant in controlling the width of flag leaves. *Qflw.nau-5A* had a significantly larger additive effect and negligible dominant effect on FLW (Table 1).

Marker saturation of the *Qflw.nau-5A* interval

A fine map of the *Qflw.nau-5A* interval in the NIL/MY-derived F_2 population of 491 plants has been reported by Xue et al. (2011). In this study, SSR markers WMC492 and WMC752 were found to be polymorphic between the NIL/MY and Mianyang 99-323 and were added to the genetic map using the same F_2 plants (Fig. 3a). Between NIL/PH and PH691, 16 markers that mapped to the QTL interval were polymorphic. Using 487 NIL/PH-derived F_2 plants, these markers were mapped and an 8.9 cM marker map was constructed (Fig. 3b). This map bordered by *Xbarc303* and *Xbarc100* had a marker density of 1.8 markers/cM.

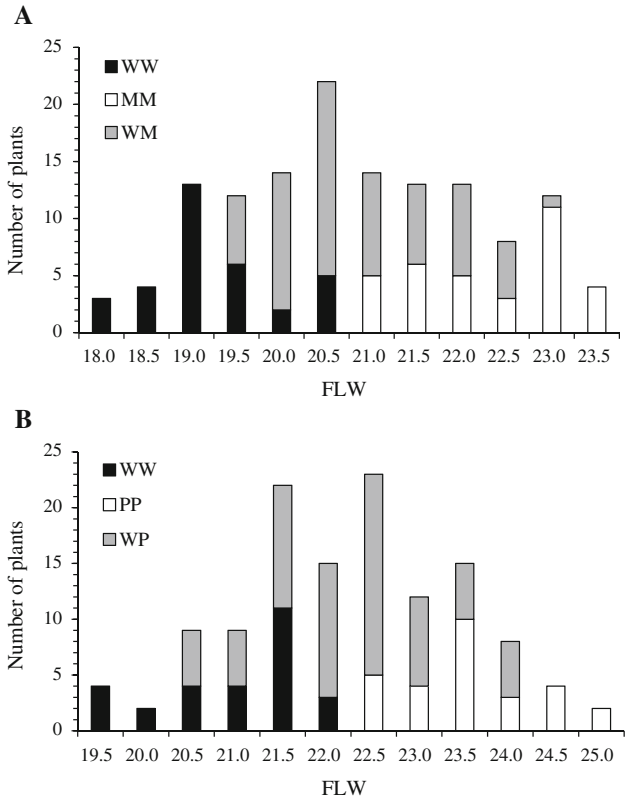

Fig. 2 FLW (mm) frequency distribution in the NIL/MY-derived F_2 population (**a**) and NIL/PH-derived F_2 population (**b**). The F_2 plants in each population were classified into three groups based on the marker genotype at *Xgwm415*: *WW*, *MM* and *PP* Wangshuibai, Mianyang 99-323 and PH691 genotypes, *WM* and *WP* heterozygous genotypes of Wangshuibai with Mianyang 99-323 or PH691

Recombinant identification of the *Qflw.nau-5A* interval

Genotyping 2816 NIL/MY-derived F_2 plants at *Xbarc303* and *Xbarc180* led to identification of 50 heterozygous recombinant plants. One hundred and ten homozygous recombinants were obtained by surveying 10–15 F_3 progenies from each of these recombinants with the ten markers mapped in the interval. These recombinants represented ten kinds of genotypes at the *Qflw.nau-5A* interval (Fig. 4), but recombination between the co-segregating markers shown in the genetic map was not found.

Eleven heterozygous recombinants were identified in the NIL/PH-derived F_2 population by genotyping 1,277 plants at *Xbarc56* and *Xbarc180*. Twenty-eight homozygous recombinants representing five kinds of genotypes at the *Qfhi.nau-5A* interval were obtained from the F_3 progenies based on the genotypes at the 12 marker loci shown in Fig. 3b (Fig. 5). In this population, recombination between *Xbarc56* and *Xgwm304* was not found. The marker orders determined using the identified recombinants were consistent with the genetic map.

Fine mapping of *Qflw.nau-5A*

Across the three field trials in different locations, the FLW of NIL/MY-derived recombinant lines showed significant variation (Table 2). Even though the variations between trials and the block variations within environments were also significant, the genotype × environment effect was not. The mean squares for genotypes were much larger than those for genotype × environment. The between-environment correlation coefficients of the genotype variable ranged from 0.95 to 0.98, all significant at $P \leq 0.0001$. The line-based broad sense heritability across environments was estimated to be 98.6 %. The FLW data displayed a discontinuous bimodal distribution in all three environments (Fig. 6a–c), with the two areas of the distribution spaced by 1.7–2.6 mm, which were significant at $P \leq 0.0001$. This allowed us to classify the recombinant lines into wide-leaf group (W) and narrow-leaf group (N) in each trial. The FLW of NIL/PH-derived recombinants were evaluated in only one trial. It also displayed a discontinuous distribution (Fig. 6d). In this trial, the FLW of the W and N groups

Table 1 FLW mean of plants with the common marker genotypes at *Xgwm415* in the *Qflw.nau-5A* NIL-derived F_2 populations and the genetic effects of *Qflw.nau-5A*

Population	Genotype group	Number of plants	FLW mean ± SE (mm)	Tukey test[a]	A[b]	D[c]	D/A[d]
(NIL/MY × Mianyang 99-323) F_2	WW	33	19.0 ± 0.1	a			
	WM	65	20.6 ± 0.1	b	1.5	0.09	0.06
	MM	34	22.0 ± 0.2	c			
(NIL/PH × PH691) F_2	WW	28	20.7 ± 0.2	a			
	WP	69	21.9 ± 0.1	b	1.3	0.01	0.01
	PP	28	23.2 ± 0.1	c			

[a] Groups with different letters were significantly different at $P = 0.01$

[b] Additive effect

[c] Dominance effect

[d] Dominant degree

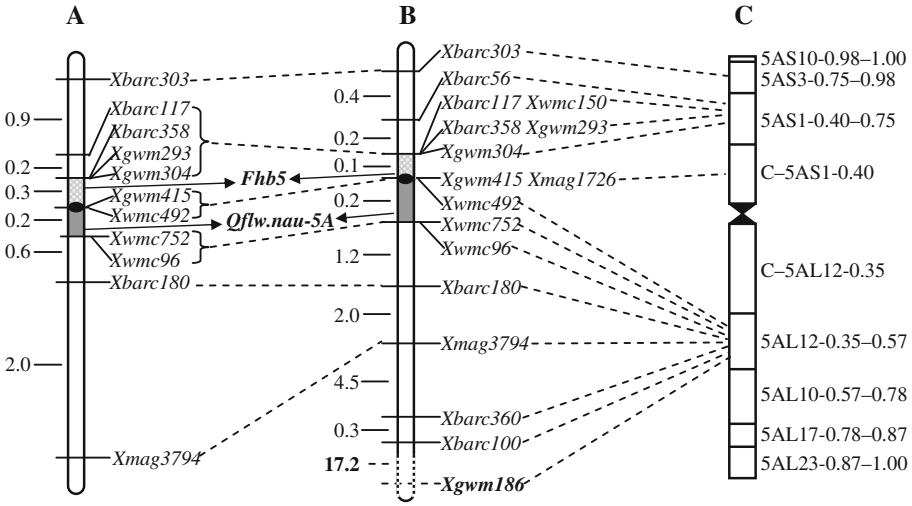

Fig. 3 Marker maps of the *Qflw.nau-5A* region: **a**, **b** constructed using the NIL/MY-derived and NIL/PH-derived F_2 populations, respectively, **c** established through mapping CS deletion lines of chromosome 5A. Markers in different maps were aligned with *dashed lines*. The short arm is at the *top*. The *black oval* indicates the approximate position of the centromere. *Numbers* to the *left* of the maps are interval distances (cM). The highlighted *Xgwm186* was placed on the map according to Xue et al. (2008b). The bin assignments delineated by neighboring deletion breakpoints are shown to the *right* of the deletion bin map. The genetic maps are not drawn to scale

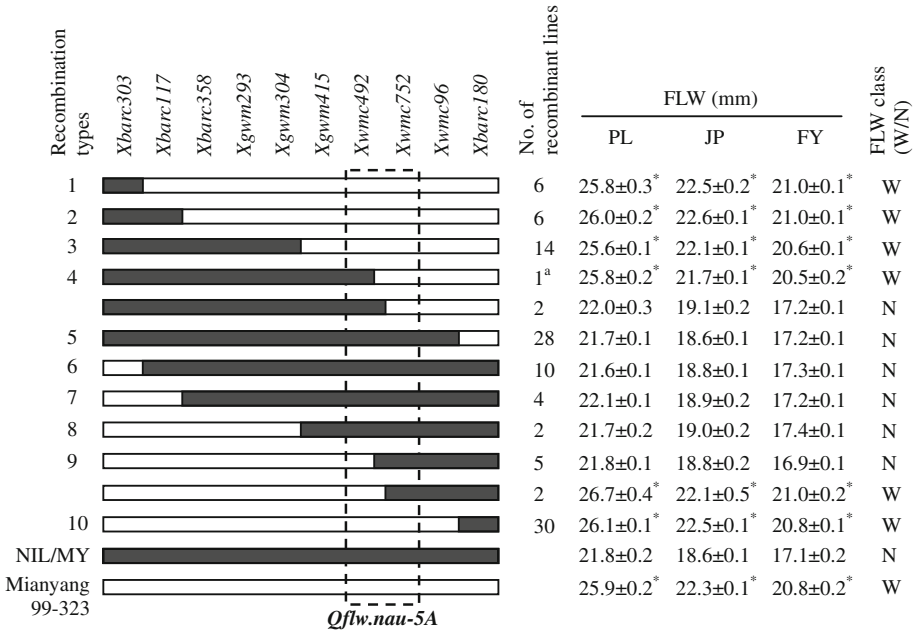

Fig. 4 Graphical illustration of the genotype types of NIL/MY-derived recombinant lines and their phenotypes in the Pailou (PL), Jiangpu (JP) and Fengyang (FY) trials. The *black rectangles* indicate Wangshuibai chromatin; the *open rectangles* indicate Mianyang 99-323 chromatin. The *broken rectangle* defines the interval harboring *Qflw.nau-5A*. *W* wide leaves, *N* narrow leaves. *Significantly different from NIL/MY at $P \leq 0.01$.
[a] The recombinant line HR015

ranged from 20.8 to 23.4 mm and from 18.4 to 19.4 mm, respectively. On the whole, the FLW of the W groups was over 16 % wider than the N group.

Of the ten NIL/MY-derived recombinant genotypes, four had wide flag leaves and four had narrow flag leaves (Fig. 4). Common to the wide-leaf types, the chromatin between *Xwmc492* and *Xwmc752* was derived from Mianyang 99-323; while common to the narrow-leaf types, the chromatin between *Xwmc492* and *Xwmc752* was derived from Wangshuibai. Thus, *Qflw.nau-5A* fell in the 0.2 cM *Xwmc492–Xwmc752* interval. Noting that in three type 4 recombinant lines that had Wangshuibai genotype at *Xwmc492* and Mianyang 99-323 genotype at *Xwmc752*, two had narrow leaves and one had wide leaves, and in seven type 9 recombinant lines that had Mianyang 99-323 genotype at *Xwmc492* and Wangshuibai genotype at *Xwmc752*, five had narrow leaves and two had wide leaves, it was concluded that *Qflw.nau-5A* resided between

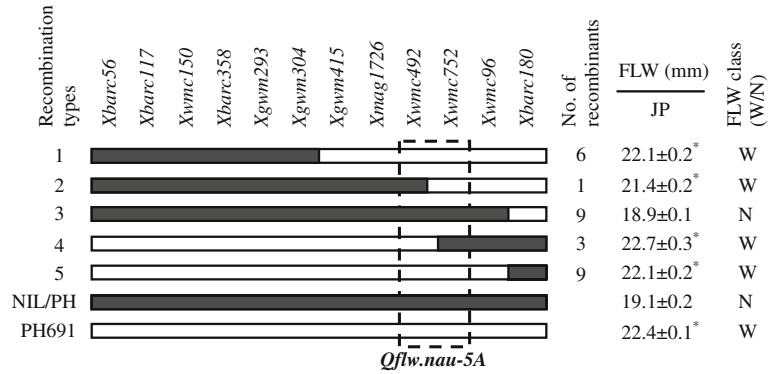

Fig. 5 Graphical illustration of the genotype types of NIL/PH-derived recombinants and their phenotypes in the Jiangpu (JP) trial. The *black rectangles* indicate Wangshuibai chromatin; the *open rectangles* indicate PH691 chromatin. The *broken rectangle* defines the interval harboring *Qflw.nau-5A*. *W* wide leaves, *N* narrow leaves. *Significantly different from NIL/PH at $P \leq 0.01$. The standard errors were estimated based on the pooled data from plants (3–6 tillers measured per plant) with the same genotypes

Table 2 Analysis of variance using a mixed linear model for FLW (mm) of NIL/MY-derived recombinant lines across three environments

Variables	DF	Mean square	*F*-value	Probability
Environment	2	1,341.150		
Block (environment)	3	1.282	4.880	0.003
Genotype	109	21.389	76.076	<0.0001
Genotype × environment	215	0.307	1.168	0.107
Error	303	0.263		

Xwmc492 and *Xwmc752*. With the NIL/PH recombinants, the same results were obtained (Fig. 5).

Deletion bin mapping of *Qflw.nau-5A*

Xue et al. (2011) have assigned 11 markers mapped to the *Xbarc303–Xbarc100* interval shown in Fig. 3b to CS deletion bins on chromosome 5A. To determine the physical position of *Qflw.nau-5A*, the remaining six markers, including *Xwmc150*, *Xmag1726*, *Xwmc492*, *Xwmc752*, *Xbarc360* and *Xgmw186*, were mapped using the CS chromosome 5A ditelosomic and deletion lines. As shown in Fig. 3c, the marker order in the deletion bin map was consistent with that in the genetic maps (Fig. 3a, b). All markers located on the top of *Xgwm415*, *Xmag1726* and *Xwmc492* were assigned to deletion bins on the short arm, while markers below these three markers were assigned to deletion bins on the long arm. *Xgwm415*, *Xmag1726* and *Xwmc492*, though co-segregating in the two populations, were placed to different bins. *Xgwm415* and *Xmag1726* were in deletion bin C–5AS1-0.40 in the short arm. *Xwmc492*, as well as *Xwmc752*, was in deletion bin 5AL12-0.35–0.57 in the long arm (Fig. 3c). Thus, *Qflw.nau-5A*, as its two flanking markers, resided in the 5AL12-0.35–0.57 bin.

Qflw.nau-5A is in close linkage with *Fhb5*

Since Xue et al. (2011) have mapped *Fhb5* to deletion bins on the short arm, *Qflw.nau-5A* was in close linkage with the FHB resistance gene (Fig. 3). If this holds true, phenotypic recombinants could exist in the recombinant lines obtained in this study. To prove this assumption, the 110 NIL/MY-derived recombinant lines were evaluated for FHB resistance. As expected, the majority of these lines, as well as the positive control, were either resistant to FHB with narrow flag leaves or susceptible with wide flag leaves. However, there were six lines displaying phenotypic recombination (Table 3). The FHB resistance of recombinant line HR015 was similar to NIL/MY, but with a significantly wider flag leaf.

Discussion

In this study, the major FLW QTL *Qflw.nau-5A* in wheat cultivar Wangshuibai was fine mapped using two NIL-derived secondary F_2 populations. Hereafter, this QTL is designated as *TaFLW1*. Based on the genotypes at the QTL interval and phenotypes of the identified recombinants, *TaFLW1* was placed in the 0.2 cM *Xwmc492–Xwmc752* interval. In barley, there is a major FLW QTL in the corresponding region of chromosome 5H (Xue et al. 2008a). The homology of both QTLs is intriguing. Yang et al. (2007) mapped a QTL for chlorophyll content of flag leaves in a similar chromosome region in wheat.

It has been shown that *TaFLW1* acted like a semi-dominant gene in the F_2 populations. Semi-dominance of genes or QTLs associated with agronomical traits seems very common. Some of the documented examples include dwarfing genes *Rht1*, *Rht2* and *Rht8* (Peng et al. 1999; Gasperini et al. 2012) in wheat, *qFLW4* for FLW (Chen

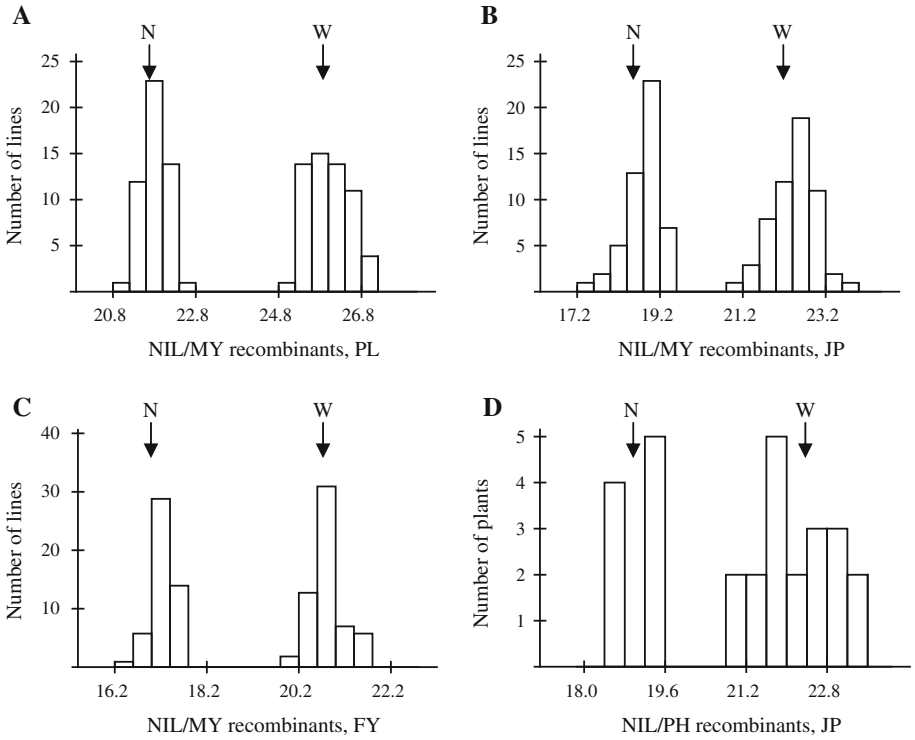

Fig. 6 FLW (mm) distribution of the identified NIL/MY-derived recombinant lines (**a**, **b**, **c**) and NIL/PH-derived recombinants (**d**). *W* wide-leaf control (Mianyang 99-323 or PH691), *N* narrow-leaf control (NIL/MY or NIL/PH)

Table 3 Flag leaf width (FLW) and percentage of diseased spikelets (PDS) of part of the NIL/MY-derived recombinant lines

Line	FLW (mm)[a]			PDS (%)[a]	
	PL	JP	FY	JP	FY
NIL/MY	21.8 ± 0.2	18.6 ± 0.2	17.1 ± 0.2	16.1 ± 1.6	24.9 ± 1.4
HR001	22.1 ± 0.2	18.8 ± 0.2	16.5 ± 0.1	44.1 ± 2.9*	48.4 ± 1.4*
HR002	21.6 ± 0.2	19.1 ± 0.2	17.1 ± 0.2	39.8 ± 1.5*	48.3 ± 1.7*
HR003	21.6 ± 0.2	18.8 ± 0.2	17.2 ± 0.2	40.5 ± 1.5*	51.1 ± 1.7*
HR015	25.8 ± 0.2*	21.7 ± 0.1*	20.5 ± 0.2*	15.7 ± 1.4	22.7 ± 1.8
HR130	22.1 ± 0.2	18.3 ± 0.2	17.2 ± 0.2	41.5 ± 1.6*	48.9 ± 1.4*
HR131	21.5 ± 0.2	18.9 ± 0.3	16.7 ± 0.2	42.7 ± 1.7*	49.0 ± 1.6*
Mianyang 99-323	25.9 ± 0.2*	22.3 ± 0.1*	20.8 ± 0.2*	44.5 ± 1.5*	49.6 ± 2.1*

* Significantly different from NIL/MY at $P \leq 0.01$

[a] Data given as mean ± SE ($n = 20$)

et al. 2012), *qSPP7* for the number of spikelets per panicle (Xing et al. 2008), *qSS7* for seed length and width (Qiu et al. 2012), *qPGWC-7* for grain chalkiness (Zhou et al. 2009) and *qGW8* for grain size (Wang et al. 2012) in rice.

The *TaFLW1* NILs had significantly narrower flag leaves compared with their recurrent parents in different trials. However, there were differences among the PL, JP and FY trials in FLW ranges and means of the recombinants (Fig. 6), implying that FLW was affected by environment. The environmental effects are likely even stronger when a single plant of each genotype is evaluated. Thus, though *TaFLW1* has a major effect on FLW, due to the semi-dominant nature of *TaFLW1* and environmental effects, it is reasonable to observe a continuous distribution for this trait in the two F_2 populations used in the genetic analysis (Fig. 2). The traits controlled by semi-dominant *qFLW4*, *qSS7* and *qPGWC-7* also displayed a continuous distribution in the respective secondary F_2 populations (Zhou et al. 2009; Chen et al. 2012; Qiu et al. 2012). The effect of genotype × environment on FLW was small (Table 2).

In this study, NIL-derived secondary F_2 populations were used in the construction of a saturated marker map of the target QTL. The two genetic maps constructed for the *TaFLW1* region were consistent in marker orders, though discrepancy of genetic distances existed between some

markers (Fig. 3). The genetic distances of the *Xwmc492–Xwmc752* interval that carries *TaFLW1* were the same in both maps. As a matter of fact, NIL-derived secondary segregating populations are often used in QTL fine mapping and isolation because of the reduced genetic background noise (Xing et al. 2008; Wang et al. 2012). To fine map *TaFLW1*, a total of 138 recombinants were identified in the secondary $F_{2:3}$ populations. These recombinants and recombinant lines homozygous at the *TaFLW1* interval could be clearly classified into two groups based on their FLW (Fig. 6). This allowed us to treat *TaFLW1* as a single gene and place it in the 0.2 cM *Xwmc492–Xwmc752* interval located in the 5AL12-0.35–0.57 deletion bin (Fig. 3). This bin was about 117.0 Mb, based on its proportion over the physical length of chromosome 5AL (Šafář et al. 2010). Since the genetic distance of this bin is at least 25.4 cM (Fig. 3b), the physical length of the 0.2 cM *Xwmc492–Xwmc752* interval is about 922 kb.

QTL fine mapping is not only beneficial for QTL isolation, but also useful in minimizing linkage drag in marker-assisted selection breeding programs (Hospital 2001). Wangshuibai is an indigenous FHB resistance germplasm. It was found that some of its FHB resistance QTL intervals were associated with yield or morphological traits (Ma et al. 2008; Xue et al. 2010), for instance, the *Fhb5* interval with narrow flag leaves. Through QTL fine mapping, it was found that *Fhb5* and *TaFLW1* were at two adjacent genetic intervals, but one on the short arm and the other on the long arm (Fig. 3). Within the identified recombinants, a line with good FHB resistance and wider leaves was obtained (Table 3). This demonstrates that, with the help of tightly linked molecular markers, breakage of linkage drag is an achievable goal.

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