Euphytica (2010) 175:331–342
DOI 10.1007/s10681-010-0175-2

Identification and molecular mapping of a dwarfing gene in barley (*Hordeum vulgare* L.) and its correlation with other agronomic traits

Junmei Wang · Jianming Yang · David L. McNeil · Meixue Zhou

Received: 13 January 2010 / Accepted: 15 April 2010 / Published online: 27 April 2010

Abstract Ninety-two doubled haploid (DH) lines, generated from a cross between Franklin and TX9425 (a Chinese Landrace), were grown in three environments to identify quantitative trait loci (QTLs) controlling agronomic traits including heading date, plant height and spike characteristics. The DH lines showed a wide range of variations for all the agronomic traits tested. Most of the traits were controlled by one or two major QTLs which explained 9.5–80.9% of the phenotypic variation. Two dwarfing genes were identified from the cross. One of the dwarfing genes was from Franklin, which is the same as the previously reported *denso* gene. The other dwarfing gene was from the Chinese landrace variety. Both dwarfing genes were temperature and/or day length sensitive. The dwarfing gene from Franklin was more effective in early sowing trials (shorter day length and lower temperature) while the gene from TX9425 was more effective in later sown trials. The dwarfing gene from TX9425 was located at a similar position to the *uzu* gene. However, it differed from this gene being temperature sensitive with very close links to short spikes, awns and high grain density which is more like a *brh* gene. To effectively use this gene in a breeding program, it is necessary to break the linkage between the dwarfing gene and the unfavourable spike traits.

Keywords Barley (*Hordeum vulgare* L.) · Quantitative trait loci (QTLs) · Dwarfing gene · Agronomic trait

Introduction

Barley (*Hordeum vulgare* L.) is an important cereal crop grown widely throughout the world's temperate agricultural zones. To improve grain yield is one of the most important objectives in breeding programs. Yield is determined by many agronomic traits. Most of these traits are controlled by quantitative trait loci (QTLs) and are hard to select for since they exhibit a continuous range of phenotypic variation and can easily be affected by environmental factors. Modern molecular biological techniques and recent advances in quantitative trait locus (QTL) analysis methods enable QTLs controlling complex traits to be mapped and elucidated (Paterson et al. 1988). QTL analysis not only provides a better understanding of the genetic

Electronic supplementary material The online version of this article (doi:10.1007/s10681-010-0175-2) contains supplementary material, which is available to authorized users.

J. Wang · D. L. McNeil · M. Zhou (✉)
Tasmanian Institute of Agricultural Research, University of Tasmania, P.O. Box 46, Kings Meadows, TAS 7249, Australia
e-mail: mzhou@utas.edu.au

J. Wang · J. Yang
Institute of Crop and Nuclear Technology Utilization, Zhejiang Academy of Agricultural Sciences, Hangzhou 310021, China

factors influencing these traits but also helps identify chromosome regions and molecular markers linked to these traits. The use of molecular markers associated with these traits can greatly improve selection efficiency by avoiding environmental effects.

Diversity Array Technology (DArT) has been shown to be very efficient for whole-genome profiling (Jaccoud et al. 2001; Wenzl et al. 2004). Although this technique is still limited to only a few laboratories at this stage, barley consensus maps (Wenzl et al. 2006) have been constructed to link DArT markers with many SSR and RFLP markers which have been previously developed and applied widely in barley mapping studies. Due to the known sequence for each DArT marker, these markers can also be effectively used in breeding programs.

Among agronomic traits, heading date is one of the important quantitative traits in adapting cereal varieties to variable environments and in maximizing yield potential (Bezant et al. 1996). Heading date is determined by three genetic factors: vernalization response, photoperiodic response, and earliness in a narrow sense (Takhashi and Yasuda 1970). It is controlled by a number of genes with a large number of QTLs being detected under field conditions (Hayes et al. 1993; Tinker et al. 1996; Marquez-Cedillo et al. 2001; Pillen et al. 2003; Sameri and Komatsuda 2004). Controlled plant height was used to reduce yield loss arising from lodging and to increase the harvest index (Bezant et al. 1996). Plant height can be a qualitative or a quantitative trait, and its inheritance is complex and involves numerous genetic factors that interact with environmental conditions (Kjær et al. 1995). QTLs conferring plant height are reported to be located on all seven chromosomes (Baghizadeh et al. 2007; Backes et al. 1995; Hori et al. 2003; Kjær et al. 1995; Sameri et al. 2006; von Korff et al. 2006; Pillen et al. 2003). In some of the populations, for example Pillen et al. (2003), plant height was controlled by more than 10 QTLs, which makes it hard for plant breeders to use molecular markers to select for this trait. Thus, it is very important to find new useful genes, preferably major genes, to control plant height in breeding programs.

Yield is generally controlled by many genes and can be dissected into a series of components including spike number, kernel number and kernel weight. Spike length and spike density are the other two important traits. These traits are not components of yield (Abeledo et al. 2002), but could indirectly affect yield and most importantly, affect the grain quality, such as plumpness and screenings. Increased spike length and spike density result in a high number of spikelets per spike. However, too high a spike density will lead to twisting of spikes, leading to lower grain quality. There are many QTL mapping studies on spike numbers per plant, kernel numbers per spike, kernel weight and spike length (Kjær et al. 1995; Hori et al. 2003; Li et al. 2005; Sameri et al. 2006; Baghizadeh et al. 2007), but little is known about the inheritance of spike density (Zhang and Sun 1993). Both grain density and twisty spike are very important for grain quality (especially malting quality), but there is no report on the QTLs controlling these two traits.

TX9425 is a Chinese landrace barley variety and was originally introduced as a source of waterlogging tolerance (Pang et al. 2004; Zhou et al. 2007), but it also showed some good agronomic traits such as shorter plant height and also some unfavourable agronomic traits such as twisty spike and too high grain density. In the current study, a doubled haploid population originating from the cross between TX9425 and an Australian malting barley Franklin, was used to investigate QTLs controlling different agronomic and yield traits. The objectives of the study were to: (1) identify the dwarfing gene(s) in TX9425 and discuss their possible use in a breeding program and (2) identify QTLs for heading date, grain density in a spike and twisty spike.

Materials and methods

Plant material

A population of 92 doubled haploid (DH) lines was produced from F_1 plants of the barley cross between Franklin and TX9425 by the anther culture method (Davies and Morton 1998). Franklin is an Australian two-rowed malting barley, and TX9425 is a Chinese two-rowed feed variety.

Field and glasshouse experiment

The DH lines and parents were grown in three different environments. Two field experiments were at Mt Pleasant Laboratories (MTP) and Forthside

Vegetable Research Station (FVRS), Tasmania, in 2007. Both field trials were sown in winter (early winter for MTP trial and late winter for FVRS) and each line was grown in a 2-m row plot with 0.4 m between rows. All agronomic management methods, including fertilization (DAP: 125 kg/ha), weed control (3 l/ha roundup before sowing), were in accordance with local practice. A glasshouse (GH) trial was conducted at the MTP in 2007. The GH trial was sown in spring and five seeds from each line were planted in 13 cm × 13 cm plastic pots filled with a pine bark/loam based potting mix with premixed slow release fertiliser. All experiments were arranged as a randomized complete block design with two replications. To confirm the effect of sowing time on plant height, the population was sown in the same site (Mt Pleasant Laboratories) at early autumn, late autumn and spring in 2009.

Phenotypic evaluation of agronomic Traits

Plant height, heading date, spike length, awn length, grain number per spike, grain density, kernel weight and the degree of spike twist were evaluated. The recorded traits and methods of measurement are listed in Table 1.

QTL analysis

A genetic linkage map was constructed using 412 DArT markers, 80 AFLP markers and 28 microsatellite markers (Wenzl et al. 2006; Li et al. 2008). For DArT markers, genomic representations and preparation of the "discovery arrays" and "polymorphism-enriched arrays" were the same as explained by Wenzl et al. (2006). Twenty-eight polymorphic primers were selected using four well-separated SSRs from each of the seven chromosomes. These SSRs were amplified using fluorescent dUTPs (Molecular Probes, Eugene, Oregon, USA). Amplification reactions were performed in a total volume of 12.5 µl containing 1× Buffer, 1.5 mM of $MgCl_2$, 0.2 mM of dNTPs, 0.2 µM of unlabeled primer, 0.6 µM fluorescent dUTPs, 0.5 U of taq polymerase and 20 ng of template DNA. SSR name and amplification conditions were those published in the Genetics supplemental data site at http://www.genetics.org/cgi/content/full/156/4/1997/DC1. Gel electrophoresis was performed on an automated Gel scanner (Gel-Scan 2000, Corbett Research). Samples were electrophoresed on 18-cm-long 4% polyacrylamide gel containing 7 M urea. Allele sizes were calculated by comparison with a 350 (TAMRA) size standard.

Using the software package MapQTL5.0 (Van Ooijen and Kyazma 2004), fsQTLs were first analysed by interval mapping (IM). The closest marker at each putative QTL identified using interval mapping was selected as a cofactor and the selected markers were used as genetic background controls in the approximate multiple QTL model (MQM) of MapQTL5.0. Logarithm of the odds (LOD) threshold values applied to declare the presence of a QTL were estimated by performing the genome wide permutation tests implemented in MapQTL version 5.0 using at least 1000

Table 1 List of agronomic trait investigated in three environments

Abbreviation	Trait	Method of measurement	Unit
PH	Plant height	Plant height measured from soil surface to tip of spike (Excluding awns)	cm
HD	Heading date	Days after the start of heading of the earliest lines in the population from same location.	d
SL	Spike length	Length from the base of spike to the tip of the terminal spikelet (Excluding awns)	cm
GN	Grain number	Number of grains per spike	–
GD	Grain density	Grain number per cm spike length	–
AL	Awn length	Length of awn in the central spikelet	cm
TI	Spike twist	Degree of twist using scaling points from 1 to 5: 5 = highest degree of spike twist; 1 = 0 degree of spike twist	–
KW	Kernel weight	Average weight of 1000 kernels	g

permutations of the original data set for each trait, resulting in a 95% LOD threshold between 2.7 and 3.0. For the measurements and comparisons of variability among the traits, we calculated the standard deviation (SD). Analysis of variance (ANOVA) was carried out with the software developed by Tang and Feng (2007).

Results

Trait analysis

The mean values of eight agronomic traits are shown in Table 2. Franklin showed higher values than TX9425 for plant height (PH), heading date (HD), spike length (SL), grain number (GN) and awn length (AL) over all three environments. TX9425 had higher grain density (GD) and degree of spike twist than Franklin. DH lines derived from the cross of these two cultivars showed significant differences for all traits measured in this experiment (Table 2), including plant height even though the difference between the two parents was non-significant in two out of three environments. Analysis of variance of all traits also showed significant effects of location and interaction between genotype and location (Table 3). For example, the plant height of the parents and DH lines was much higher at MTP than the other two environments with the lowest value in the GH. Greater differences between the two parents were found in the GH than at the other two locations (Table 2). As expected many of the traits correlated with each other (Table 4). Short spike length and high grain density can cause the twist of the spike. Awn length also showed significant correlation with spike length and grain density. Most of the lines with a shorter awn showed shorter spike length and greater grain density, thus the twisty spike was associated with short awns.

Table 2 Mean values for agronomic traits of TX9425, Franklin and the DH lines from this cross

Traits	Plant height (cm)	Heading date (d)	Spike length (cm)	Grain number	Grain density	Awn length (cm)	Spike twist	Kernel weight (g)
MTP								
TX9425	110	4.5	5.36	32.8	6.1	5.93	4.5	46.7
Franklin	115	24.5	10.22	33.8	3.3	14.22	1.0	42.3
DH lines-minimum	58	0.0	4.28	27.2	3.3	5.39	1.0	28.6
DH lines-maximum	143	29.0	10.59	42.8	7.7	17.11	5.0	53.0
DH lines-mean	107	14.1	6.75	35.5	5.5	8.11	2.4	42.6
SD	18	8.0	1.47	3.32	1.1	3.08	1.2	0.48
FVRS								
TX9425	90	0.0	5.83	33.4	5.7	4.70	4.0	
Franklin	95	16.0	10.04	35.2	3.5	12.74	1.0	
DH lines-minimum	48	0.5	4.48	27.0	3.4	3.70	1.0	
DH lines-maximum	125	19.0	11.30	40.8	7.4	14.63	5.0	
DH lines-mean	89	10.3	6.66	34.7	5.4	7.47	2. 5	
SD	16	5.2	1.57	2.9	1.1	2.85	1.1	
GH								
TX9425	60	5.0	4.69	27.4	5.84	4.68	3.0	
Franklin	88	15.5	10.22	29.8	2.92	14.27	1.0	
DH lines-minimum	38	0.0	2.90	19.6	2.64	3.82	1.0	
DH lines-maximum	113	33.0	9.74	33.6	6.76	16.68	5.0	
DH lines-mean	71	13.1	5.54	26.0	4.99	7.33	2.3	
SD	16	9.3	1.69	3.18	1.12	4.34	0.9	

SD standard deviation, *MTP* Mt Pleasant Laboratories, *FVRS* Forthside Vegetable Research Station, *GH* glasshouse experiment

Table 3 Analysis of variance on agronomic traits in DH lines from Franklin × TX9425 cross

Source of variation	Plant height	Heading date	Spike length	Grain number	Grain density	Awn length	Spike twist
Block	21.79**	1.11	0.03	0.90	2.83	5.35	25.67**
Genotype	19.78**	99.72**	64.68**	10.64**	76.55**	236.56**	29.67**
Location	695.19**	5427.14**	303.36**	1006.06**	114.85**	77.45**	12.25**
Genotype × Location	2.01**	12.30**	2.42**	2.10**	2.06**	6.99**	1.86**

** Significant at the 1% level

Table 4 Correlation coefficients between agronomic traits in DH lines from Franklin × TX9425 (calculated from the average value of three locations)

	Plant height	Heading date	Spike length	Grain number	Grain density	Awn length
Heading date	−0.29*					
Spike length	0.60**	0.1				
Grain number	0.13	0.77**	0.30*			
Grain density	−0.59**	0.21	−0.91**	0.07		
Awn length	0.50**	0.12	0.83**	0.17	−0.74**	
Spike twist	−0.37**	−0.14	−0.84**	−0.2	0.83**	−0.70**

* Significant at the 5% level

** Significant at the 1% level

Identification of QTLs associated with different traits

The QTL names (Fig. 1) for plant height were given by a "Q", followed by "Ph", a ".", "sowing time", and the number of the QTL if two QTLs were identified. QTL positions were the distances from the first marker at the end of the short arm.

Plant height (PH)

Two significant QTLs were identified for PH at MTP, which explained more than 50% of the phenotypic variation. Both are located on chromosome 3H in the positions of 58 and 83 cM, with bPb-0716 and bPb-2433 being the closest markers to these two QTLs (Fig. 1). Both alleles (bPb-0176 from Franklin and bPb-2433 from TX9425) contributed to shorter plant height. Two QTL were also found on chromosome 3H at FVRS. One of the QTL (in the position of 83 cM) is in the same position as found at MTP and the other one is in the position of 69 cM. In the glasshouse experiment, only one significant QTL was identified which is located on chromosome 3H in the same position (69 cM) as the second one found at FVRS. It explained 53% of the phenotypic variation, with the nearest marker being bPb-0079, this allele from TX9425 also contributed to shorter plant height (Table 5, Fig. 1). Further field experiments in 2009 showed similar results (Table 6). Combined analysis was conducted and QTLs for plant height in different sowing date are shown in Fig. 1.

Heading date (HD)

Two QTLs were found from different environments (Table 5, Fig. S1). At both MTP and FVRS, one QTL was located on chromosome 3H (83 cM) with the nearest marker being bPb-0716 and the other one was located on chromosome 2H (20 cM) with the nearest marker being bPb-4523. These two QTLs explained 39 (MTP)–42% (FVRS) of the phenotypic variation. Both alleles from Franklin contributed to the late heading. However, the effects of the individual QTLs on late heading were different from different environments. At MTP, which is an early sowing trial, the QTL on chromosome 3H showed greater effect while at FVRS experiments, the effect of the QTL on

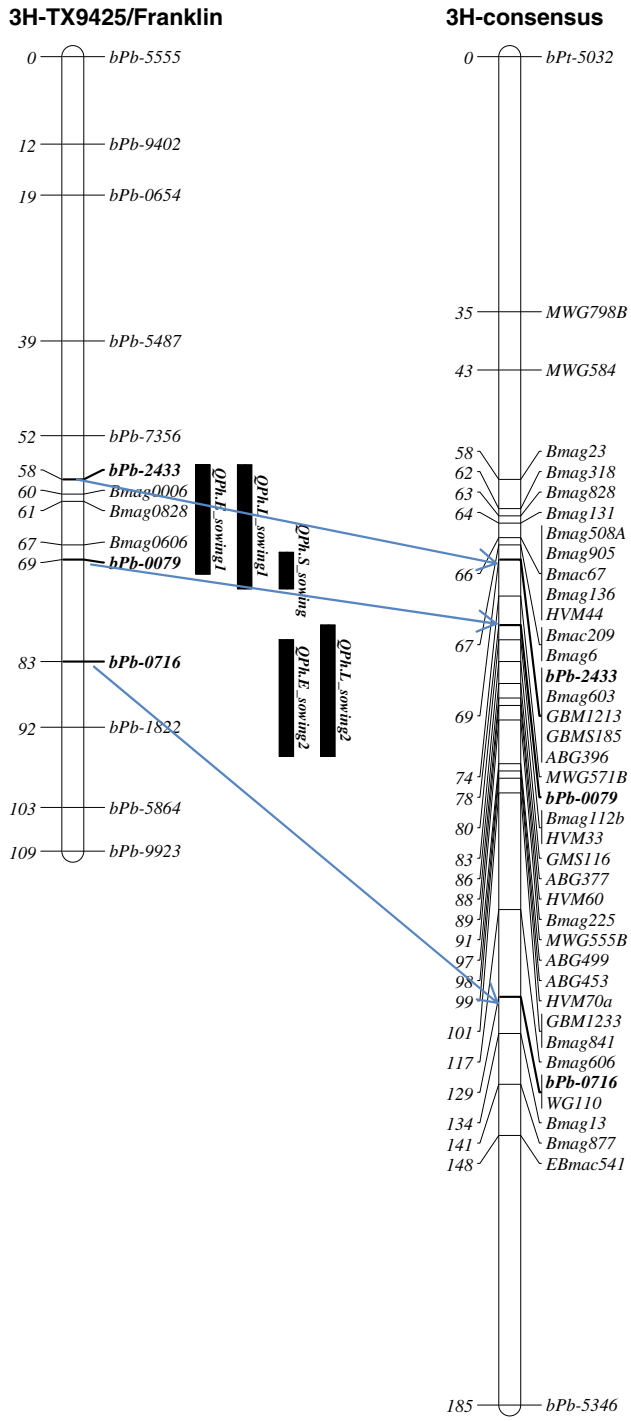

Fig. 1 Quantitative trait loci (QTLs) identified for plant height in the DH population of TX9425 × Franklin. This figure is for chromosome 3H. Only SSR markers and a few DArT markers are shown in the map. For detailed map, please refer to Li et al. (2008). Consensus map (Wenzl et al. 2006) was added to the right for comparison. In the consensus map, only some common markers shared with SSR consensus map by Varshney et al. (2007) were shown. Arrows point out the position of closest DArT markers for two dwarfing genes in two different maps

chromosome 2H was much greater. In the glasshouse experiment, one QTL on chromosome 2H with the same position as that identified at MTP and FVRS showed dominant effect on heading date, explaining 51% of the phenotypic variation. The second QTL on chromosome 3H identified in the glasshouse experiment

Table 5 QTLs for agronomic traits detected in the DH population of Franklin × TX9425 from three environments

Trait	Location	Chromosome	Marker interval	Nearest marker	Position (cM)	LOD	R^2 (%)	Additive effect
Plant height	MTP	3H	80.2–85.6	bPb-0716	82.6	8.95	34.8	−11.07
		3H	55.9–60.8	bPb-2433	57.6	6.00	21.0	−8.41
	FVRS	3H	78.2–84.6	bPb-0716	82.6	7.28	31.4	−9.12
		3H	66.7–74.5	p18b2	68.7	5.10	21.0	−8.77
	GH	3H	67.7–73.2	bPb-0079	69.2	11.70	52.7	−14.72
Heading date	MTP	2H	9.6–29.0	bPb-4523	19.9	3.58	16.8	3.31
		3H	77.2–85.6	bPb-0716	82.6	4.97	21.9	3.93
	FVRS	2H	14.8–23.2	bPb-4523	19.9	6.50	32.7	3.01
		3H	80.2–89.6	bPb-0716	82.6	2.71	9.5	1.75
	GH	2H	17.8–25.6	bPb-4523	19.9	10.48	51.1	6.69
		3H	55.9–61.8	bPb-2433	57.6	3.65	13.8	3.41
Spike length	MTP	2H	128.6–132.1	bPb-4094	130.8	11.66	30.7	−0.81
		3H	67.7–73.2	bPb-0079	69.2	12.68	34.6	−1.09
	FVRS	2H	129.2–135.1	bPb-7211	130.0	15.57	28.1	−0.83
		3H	67.7–73.2	bPb-0079	69.2	21.22	47.5	−1.37
	GH	2H	129.2–132.1	bPb-6087	130.2	5.18	10.7	0.55
		3H	67.7–73.5	bPb-0079	69.2	17.66	57.3	−1.61
Grain number	MTP	–	–	–	–	–	–	–
	FVRS	2H	15.8–20.9	bPb-8750	18.9	6.00	32.3	1.62
	GH	2H	16.8–28.0	bPb-7229	25.6	6.31	33.2	−1.82
Grain density	MTP	2H	129.2–132.1	bPb-7211	130.0	16.16	37.9	0.70
		3H	59.8–73.2	bPb-0079	69.2	14.93	33.5	0.83
	FVRS	2H	127.8–132.1	bPb-7211	130.0	11.85	29.8	0.58
		3H	67.7–73.2	bPb-0079	69.2	13.70	36.8	0.81
	GH	2H	129.2–132.1	bPb-5619	129.8	10.63	21.1	0.51
		3H	67.7–73.2	bPb-0079	69.2	18.93	51.1	1.01
Awn length	MTP	3H	67.7–73.5	bPb-0079	69.2	19.29	70.9	−3.26
	FVRS	3H	67.7–73.5	bPb-0079	69.2	14.43	69.5	−2.96
	GH	3H	66.7–73.5	bPb-0079	69.2	25.88	80.9	−4.90
Spike twist	MTP	2H	127.8–132.1	bPb-6047	130.6	5.43	19.7	0.53
		3H	67.7–73.2	bPb-0079	69.2	7.16	27.6	0.79
	FVRS	2H	129.2–142.9	bPb-7211	130.0	10.40	28.1	0.58
		3H	67.7–73.2	bPb-0079	69.2	12.26	35.4	0.82
	GH	2H	127.8–142.9	bPb-7211	130.0	4.98	15.8	0.37
		3H	67.7–73.2	bPb-0079	69.2	9.80	36.7	0.71
Kernel weight	MTP	5H	5–30.6	bPb-6051	20.4	3.67	21.8	−0.22

The position is that of the nearest marker; R^2 means percentage genetic variance explained by the nearest marker; Abbreviations for traits are given in Table 1

MTP Mt Pleasant Laboratories, *FVRS* Forthside Vegetable Research Station, *GH* glasshouse experiment

was different from those found at MTP and FVRS, which was in the position of 58 cM with bPb-2433 being the nearest marker (Table 5). This allele from TX9425 also contributed to late heading.

Spike length (SL)

At all three locations, two QTLs were found to be associated with this trait. The first QTLs on

Table 6 QTLs for plant height detected in the DH population of Franklin × TX9425 from different sowing time in 2009

Sowing time	Chromosome	Nearest marker	Position (cM)	LOD	R^2 (%)	Additive effect
Early Autumn	3H	bPb-0716	82.6	3.90	22.1	−8.0
	3H	bPb-2433	57.6	2.76	16.2	−6.6
Late Autumn	3H	bPb-0716	82.6	6.32	23.9	−8.3
	3H	p18b2	68.7	7.28	29.1	−10.8
Spring	3H	bPb-0079	69.2	12.21	54.2	−14.3

chromosome 2H was at the position of around 130 cM, explaining 11–31% of the phenotypic variation. The position of the second QTL was 69 cM on chromosome 3H, explaining 35–57% of the phenotypic variation (Table 5, Fig. S1).

Grain number (GN) and kernel weight (KW)

One QTL associated with the number of grains per spike was found at both FVRS and in the glasshouse. This QTL was on chromosome 2H and located at the position of around 20 cM, explaining more than 30% of the phenotypic variation. Surprisingly, no QTL was identified at MTP. The kernel weight was only measured at MTP and one QTL, with bPb-6051 being the nearest marker, was identified (Table 5, Fig. S1).

Grain density (GD) and spike twist (TI)

Grain density and the degree of spike twist were closely related to each other and both traits were also closely related with spike length. The QTLs identified for these two traits were the same as or similar to those identified for spike length. The QTL on chromosome 2H located at the position of 130 cM and the other QTL on chromosome 3H was at 69 cM with the molecular marker, bPb-0079 (Table 5, Fig. S1).

Awn length (AL)

Awn length acted as a qualitative trait and all the DH lines fell into two distinct groups, short awn from 5 to 8 cm and long awn from 11 to 15 cm. One significant QTL on chromosome 3H was identified in all locations. This QTL, with bPb-0079 being the nearest marker, explained 70–81% of the phenotypic variation of the trait (Table 5, Fig. S1).

Discussion

TX9425 is a Chinese landrace variety with several useful traits that can be used in our breeding program. These traits include waterlogging tolerance (Pang et al. 2004, 2006, 2007a, b; Zhou et al. 2007; Li et al. 2008); salinity tolerance (Chen et al. 2007), and some disease resistances. However, TX9425 also showed very low malting quality (Zhou et al. 2008) with some unique agronomic traits such as twisted spike due to an excessively high grain density in the spike leading to non-uniform grains. This variety also showed different reactions to sowing time (different day-length and temperature). In this study, the growth of TX9425 was significantly affected by the environment. Under winter-sown conditions (MTP and FVRS), the plant height was only slightly shorter than Franklin, while under spring-sown conditions (GH), TX9425 was much shorter than Franklin. To effectively use this variety in the breeding programs it is crucial to study the genetic behaviour of agronomically important traits and possibly find more useful genes.

Most of the agronomic traits are affected by environment. The use of molecular markers for selecting these traits can greatly increase selection efficiency. Great effort has been made by a number of researchers (Backes et al. 1995; Kjær et al. 1995; Hori et al. 2003; Pillen et al. 2003; Sameri et al. 2006; von Korff et al. 2006; Baghizadeh et al. 2007) and many QTLs have been found for different agronomic traits (Table 7). In this study, two QTLs for plant height (PH) were identified from different environments. Both alleles (one from Franklin and one from TX9425) contributed to the short plant height. Franklin carries the *denso* dwarfing gene from Triumph which is allelic to the *sdw* dwarfing gene (Laurie et al. 1993; Hellewell et al. 2000). The dwarfing

Table 7 Comparison of QTLs found in this study to QTLs detected in other studies

Trait	QTLs in this study		QTLs in other studies	
	Chromosome	Position (cM)	Chromosome	Position (cM)
Plant height			1H	8[2]; 144, 162[7]; 57, 62[5]
			2H	2[3]; 15, 73, 86[4]; 42, 86[7]
	3H	82(111)[a], 69(62) 58(54)	3H	18(139)[3]; 65.5(43)[6]; 49(48), 155(135)[7]
			4H	39[1]; 17[2]; 73.4[6]; 180[7]; 24, 26, 45, 62, 88, 108[5]
			5H	14[1]; 43[7]; 141, 148[5]
			6H	16[1]
			7H	18[3]; 84, 137.8[6]; 27, 62, 146[7]; 98, 107, 109, 118, 139, 145, 165[5]
Heading date			1H	88, 143[6]; 130[7]; 17, 28, 57, 62[5]
	2H	20(32)	2H	50(60)[1]; 29, 74, 32, 57(30-80)[4]; 49(50)[6]; 42(40), 146(130)[7]; 5(11), 48(66), 50(68), 103(122)[5]
	3H	82(111), 58(54)	3H	25 (20), 155(135)[7]
			4H	190[7]; 24, 26, 62, 118[5]
			5H	106[6]; 41, 91, 141, 148, 187[5]
			6H	6, 107[7]
			7H	11, 36[1]; 19, 146[7]; 98, 107, 118, 145, 167[5]
Spike length	2H	130(135)	2H	2(90)[3]; 30, 75(30–80)[4]; 47(47), 122(122)[6]
	3H	69(62)	3H	66(65)[6]
			4H	0, 9[2]
			5H	14[3]
			7H	84[6]
Grain number			1H	17[5]
	2H	20(32)	2H	–[2]
Grain density	2H	130(135)		
	3H	69(62)		
Awn length			2H	80[6]
	3H	69(62)	3H	66(65)[6]
			7H	84[6]
Spike twist	2H	130(135)		
	3H	69(62)		
Kernel weight			1H	6[2]
			2H	20[1]; 4[3]; 27[7]; 5, 48, 50[5]
			3H	110, 175[7]
			4H	13[1]; 95, 130[7]; 24, 26, 45[5]
	5H	20(0)	5H	165(145)[7]; 35(45), 91, 141(126)[5]
			6H	40, 103[7]
			7H	6[1]; 146[7]

References: [1]Backes et al. 1995, [2]Baghizadeh et al. 2007, [3]Hori et al. 2003, [4]Kjær et al. 1995, [5]Pillen et al. 2003, [6]Sameri et al. 2006, [7]von Korff et al. 2006

[a] The numbers in bracket is the estimated position in the SSR consensus map (Varshney et al. 2007)

allele from Franklin identified in this study is at the same position as *denso/sdw* gene based on the nearest markers of the consensus map (Varshney et al. 2007; Wenzl et al. 2006). This gene showed temperature and/or day length sensitivity in this experiment, making it less effective in spring sowing trials. QTLs

on chromosome 3H for plant height were also reported by several researchers (Sameri et al. 2006; Hori et al. 2003; von Korff et al. 2006), but the positions of the reported QTLs were different from the QTLs found in this study (Table 7), by locating different QTLs on the consensus map (Varshney et al. 2007). The new QTL found in this study also had much greater effect than previously reported, indicating that the gene in TX9452 could be a major dwarfing gene. This gene is located at a similar position to the *uzu* gene (GrainGenes: A database for *Triticeae* and *Avena*). The *uzu* gene was reported to be temperature sensitive (Takahashi and Yamamoto 1951) but there are no reports on the linkage between this gene and short spike length, short awn and high grain density. The gene from TX9425 showed strong links with short spike length, short awn and high grain density, in which case it acted more like a type of *brh* gene (Dahleen et al. 2005)).

Like plant height, QTLs for heading date (HD) have been found to be located on every chromosome (Backes et al. 1995; Pillen et al. 2003; Kjær et al. 1995; Sameri et al. 2006; von Korff et al. 2006) and heading date was always controlled by several different QTLs at the same time. Pillen et al. (2003) found 22 putative QTLs controlling heading date, distributed on chromosomes 1H, 2H, 4H, 5H and 7H, all contributing a small amount of the phenotypic variation. In this DH population, growth conditions showed marked effects on the QTLs identified for heading date, even though two QTLs were identified for all the different environments. At MTP (early winter sown), the QTL on chromosome 3H showed greater effect on heading date while under glasshouse conditions (spring sown) and at FVRS (late winter sown), the QTL on chromosome 2H showed much greater effect on heading date. The QTL on chromosome 3H identified in the glasshouse experiment was in a different position to that found at MTP and FVRS. A similar QTL position on chromosome 2H was reported earlier (Kjær et al. 1995) but the QTL position on chromosome 3H seems to be different from that reported by von Korff et al. (2006). For most of the QTLs identified in this population, the alleles from the late maturity variety Franklin contributed to late maturity. Surprisingly, the QTL on chromosome 3H identified from glasshouse experiment which is a TX9425 allele contributed to late maturity. This is probably due to the great response to growth conditions of the parent variety TX9425. Further research may be needed to clarify the genes controlling heading date in this population.

Two QTLs for spike length (SL) were located on chromosomes 2H and 3H. They are not affected by the growth conditions with both found in the same position in three different environments. These two QTLs are also in similar positions to those reported by Sameri et al. (2006) but different from those reported by other researchers (Table 6). There are very few reports on the QTLs for awn length. Sameri et al. (2006) found three QTLs for awn length with a major one on chromosome 3H (explaining 76% of the genotypic variation), which is in the same position to the QTL identified in this population (also explaining more than 70% of the variation). Twisty spike is one of the distinct characters of the Chinese landrace variety. There is no report on the genetics of this trait. Not surprisingly, this trait is closely correlated with spike length and grain density (Table 4). Two QTLs controlling both grain density and twisty spike were in the same chromosome and position as those for spike length. The alleles from TX9425 contributed all the unfavourable traits (short spike, high grain density and twisty spike). Kernel weight was only measured from one site (MTP) and one QTL was identified, which is different from earlier reports (Table 7).

In conclusion, a dwarfing gene from the Chinese landrace variety was identified in this experiment. This gene showed temperature and/or day length sensitivity and was very closely linked with other unfavourable agronomic traits such as spike length and grain density. To effectively use this dwarfing gene in a breeding program, it is necessary to break the linkage. Since this gene showed too strong an effect under high temperature and/or long day length, it is not suitable to be used for spring sown barley.

Acknowledgements This work was supported by Grains Research and Development Corporation of Australia (UT8) grant to M.Z. and National Natural Science Foundation of China (30700485) grant to J.W.

References

Abeledo LG, Calderini DF, Slafer GA (2002) Physiological changes associated with genetic improvement of grain yield barley. In: Slafer GA, Molina-Cano JL, Savin R,

Araus JL, Romagosa I (eds) Barley science: recent advances from molecular biology to agronomy of yield and quality. The Haworth Press Inc., Binghamton, New York, pp 361–386

Backes G, Graner A, Foroughi-Wehr B, Fischbeck G, Wenzel G, Jahoor A (1995) Localization of quantitative trait loci (QTL) for agronomic important characters by the use of a RFLP map in barley (*Hordeum vulgare* L.). Theor Appl Genet 90:294–302

Baghizadeh A, Taleei AR, Naghavi MR (2007) QTL analysis for some agronomic traits in barley (*Hordeum vulgare* L). Int J Agr Biol 9:372–374

Bezant J, Laurie D, Pratchett N, Chojecki J, Kearsey M (1996) Marker regression mapping of QTL controlling flowering time and plant height in a spring barley (*Hordeum vulgare* L.) cross. Heredity 77:64–73

Chen Z, Zhou M, Newman IA, Mendham NJ, Zhang G, Shabala S (2007) Potassium and sodium relations in salinised barley tissues as a basis of differential salt tolerance. Funct Plant Biol 34:150–162

Dahleen LS, LJvd Wal, Franckowiak JD (2005) Characterization and Molecular Mapping of genes determining semidwarfism in barley. J Hered 96:654–662

Davies PA, Morton S (1998) A comparison of barley isolated microspore and anther culture and the influence of the cell culture density. Plant Cell Rep 17:206–210

Hayes PM, Liu BH, Knapp SJ, Chen F, Jones B, Blake T, Franckowiak J, Rasmusson D, Sorrells M, Ullrich SE, Wesenberg D, Kleinhofs A (1993) Quantitative trait locus effects and environmental interaction in a sample of North American barley germplasm. Theor Appl Genet 87: 392–401

Hellewell KB, Rasmusson DC, Gallo-Meagher M (2000) Enhancing yield of semidwarf barley. Crop Sci 40: 352–358

Hori K, Kobayashi T, Shimizu A, Sato K, Takeda K, Kawasaki S (2003) Efficient construction of high-density linkage map and its application to QTL analysis in barley. Theor Appl Genet 107:806–813

Jaccoud D, Peng KM, Feinstein D, Kilian A (2001) Diversity arrays: a solid state technology for sequence information independent genotyping. Nucleic Acids Res 29:e25

Kjær B, Jenson J, Giese H (1995) Quantitative trait loci for heading date and straw characters in barley. Genome 38:1098–1104

Laurie DA, Pratchett N, Romero C, Simpson E, Snape JW (1993) Assignment of the denso dwarfing gene to the long arm of chromosome 3 (3H) of barley by use of RFLP markers. Plant Breed 111:198–203

Li JZ, Huang XQ, Heinrichs F, Ganal MW, Roder MS (2005) Analysis of QTLs for yield, yield components, and malting quality in a BC3-DH population of spring barley. Theor Appl Genet 110:356–363

Li H, Vaillancourt R, Mendham N, Zhou M (2008) Comparative mapping of quantitative trait loci associated with waterlogging tolerance in barley (*Hordeum vulgare* L.). BMC Genomics 9:401

Marquez-Cedillo LA, Hayes PM, Kleinhofs A, Legge WG, Rossnagel BG, Sato K, Ullrich SE, Wesenberg DM (2001) QTL analysis of agronomic traits in barley based on the doubled-haploid progeny of two elite North American varieties representing different germplasm groups. Theor Appl Genet 103:625–637

Pang J, Zhou M, Mendham N, Shabala S (2004) Growth and physiological responses of six barley genotypes to waterlogging and subsequent recovery. Aust J Agric Res 55:895–906

Pang JY, Mendham N, Zhou MX, Newman I, Shabala S (2006) Microelectrode ion and O_2 flux measurements reveal differential sensitivity of barley root tissues to hypoxia. Plant Cell Environ 29:1107–1121

Pang J, Cuin T, Shabala L, Zhou M, Mendham N, Shabala S (2007a) Effect of secondary metabolites associated with anaerobic soil conditions on ion fluxes and electrophysiology in barley roots. Plant Physiol 145:266–276

Pang J, Ross J, Zhou M, Mendham N, Shabala S (2007b) Amelioration of detrimental effects of waterlogging by foliar nutrient sprays in barley. Funct Plant Biol 34: 221–227

Paterson AH, Lander ES, Hewitt JD, Peterson S, Lincoln SE, Tanksley SD (1988) Resolution of quantitative traits into Mendelian factors by using a complete linkage map of restriction fragment length polymorphisms. Nature 335: 721–726

Pillen K, Zacharias A, Leon J (2003) Advanced backcross QTL analysis in barley (*Hordeum vulgare* L.). Theor Appl Genet 107:340–352

Sameri M, Komatsuda K (2004) Identification of quantitative trait loci (QTLs) controlling heading time in the population generated from a cross oriental and occidental barley cultivars (*Hordeum vulgare* L). Breed Sci 54:327–334

Sameri M, Takeda K, Komatsuda T (2006) Quantitative trait loci controlling agronomic traits in recombinant inbred lines from a cross between oriental- and occidental-type barley cultivars. Breed Sci 56:243–252

Takahashi R, Yamamoto J (1951) Studies on the classification and geographycal distribution of the Japanese barley varieties. III. On the linkage relation and the region of "*uzu*"or semi-brachytic character in barley. Ber Ohara Inst Landow Forsch 9:399–410

Takhashi R, Yasuda S (1970) Genetics of earliness and growth habit in barley. In: Nilan RA (ed) Barley Genetics II, Proceedings of the Second International Barley Genetics Symposium. Washington State University Press, Pullman, WA, pp 388–408

Tang QY, Feng MG (2007) DPS Data processing system: experimental design statistical analysis and data mining. Science Press, Beijing

Tinker NA, Mather DE, Rossnagel BG, Kasha KJ, Kleinhofs A, Hayes PM et al (1996) Regions of the genome that affect agronomic performance in two-row barley. Crop Sci 36:1053–1062

Van Ooijen JW, Kyazma BV (2004) MapQTL® 5 Software for the mapping of quantitative trait loci in experimental populations. Wageningen, The Netherland

Varshney RK, Marcel TC, Ramsay L, Russell J, Roder MS, Stein N, Waugh R, Langridge P, Niks RE, Graner A (2007) A high density barley microsatellite consensus map with 775 SSR loci. Theor Appl Genet 114: 1091–1103

Von Korff M, Wang H, Léon J, Pillen K (2006) AB-QTL analysis in spring barley: II. Detection of favourable

exotic alleles for agronomic traits introgressed from wild barley. Theor Appl Genet 112:1221–1231

Wenzl P, Carling J, Kudrna D, Jaccoud D, Huttner E, Kleinhofs A, Kilian A (2004) Diversity arrays technology (DArT) for whole-genome profiling of barley. Proc Nat Acad Sci 101:9915–9920

Wenzl P, Li H, Carling J, Zhou M, Raman H, Paul E, Hearnden P, Maier C, Xia L, Caig V, Ovesna J, Cakir M, Poulsen D, Wang J, Raman R, Smith KP, Muehlbauer GJ, Chalmers KJ, Kleinhofs A, Huttner E, Kilian A (2006) A high-density consensus map of barley linking DArT markers to SSR, RFLP and STS loci and agricultural traits. BMC Genomics 7:206–227

Zhang J, Sun LJ (1993) Inheritance of spike length and density of barley in three crosses. Hereditas (Beijing) 15:23–26

Zhou MX, Li HB, Mendham NJ (2007) Combining ability of waterlogging tolerance in barley (*Hordeum vulgare* L.). Crop Sci 47:278–284

Zhou MX, Li HB, Chen ZH, Mendham NJ (2008) Combining ability of barley flour pasting properties. J Cereal Sci 48:789–793