Identification of QTLs for Yield Related Traits in *Indica* Type Rice Using SSR and AFLP Markers

Babak RABIEI [1(✉)]
Mojtaba KORDROSTAMI [2]
Atefeh SABOURI [1]
Hossein SABOURI [3]

Summary

This research was carried out to identify quantitative trait loci (QTLs) controlling yield and yield components in rice using 196 $F_{2:4}$ lines derived from a cross between two rice varieties of *indica*, Sepidrood and Gharib. Quantitative trait loci analysis using composite interval mapping was carried out by 105 SSR and 111 AFLP markers. Results showed that 8 chromosomes contain 28 regions (QTLs) controlling 11 studied traits. One QTL was mapped for the number of spikelet per panicle on chromosome 12, three QTLs for number of filled grains per panicle on chromosomes 1, 6 and 11, three QTLs for empty spikelets per panicle on chromosomes 2, 3 and 12, five QTLs for plant height on chromosomes 1, 7 (2 QTLs), eight and 11, four QTLs for days to 50% flowering on chromosomes 2, 3 (2 QTLs) and 6, one QTL for panicle length on chromosome 1, two QTLs for 1000 grain weight on chromosomes 1 and 2, three QTLs for number of panicles per plant on chromosomes 1, 3 and 6, one QTL for grain yield on chromosome 3, four QTLs for days to maturity on chromosomes 2, 3 (2 QTLs) and 6 and one QTL for fertility percentage on chromosome 11. The identified QTLs on specific chromosome regions explaining high phenotypic variance can be considered for use in marker-assisted selection (MAS) programs.

Key words

AFLP and SSR markers, linkage map, QTL mapping, yield related traits

[1] University of Guilan, Dept. of Agronomy and Plant Breeding,
Faculty of Agricultural Sciences, P.O. Box: 41635-1314, Rasht, Iran
✉ e-mail: rabiei@guilan.ac.ir
[2] University of Guilan, Dept. of Biotechnology, Faculty of Agricultural sciences,
P.O. Box: 41635-1314, Rasht, Iran
[3] Gonbad Kavous University, Dept. of Plant Production, P.O. Box: 49717-99151, Gonbad, Iran

Received: December 8, 2014 | Accepted: April 1, 2015

ACKNOWLEDGEMENTS
Assistance provided by Agricultural Biotechnology Research Institute of North of Iran was greatly appreciated.

Introduction

The major challenge to rice breeders for increasing yield potential of cultivars is to improve the agronomic traits contributing to rice yield. It is difficult to achieve this goal by using conventional breeding technologies because of the epistatic interaction of different yield-contributing genes (Jena and Mackill, 2008). Recent advances in high-throughput technologies for the quantification of biological molecules have shifted the focus in quantitative genetics from single traits to comprehensive large-scale analyses (Xu, 2010). The major requirement for application of molecular markers in rice improvement is that a gene or QTL of significant effect has to be mapped with a high degree of accuracy, and that the gene is effective in the desired genetic background. Furthermore, the gene should not have negative effects on other traits. In initial mapping studies, a gene is generally mapped at low resolution using small populations (< 200) (McKill and McNally, 2004).

Rice (*Oryza sativa* L.) is one of the most important food crops worldwide. Rice has also been adopted as an important model system for plant science research (Xing and Zhang, 2010), and a lot of research has been carried out on this model crop. Mapping quantitative trait loci (QTLs) become an important approach to study quantitative traits, and has been widely employed for important agricultural crops such as rice (Jiming *et al*., 2001). Because rice is an important food crop in Iran, this research was carried out to identify quantitative trait loci (QTLs) controlling yield and yield components in rice using 196 $F_{2:4}$ lines derived from a cross between two rice varieties of *indica*, 'Sepidrood' and 'Gharib'.

Materials and methods

Plant material and Field evaluation

The rice mapping populations used in this study included a set of 196 F_4 individuals derived from a cross between two Iranian cultivars, 'Sepidrood' and 'Gharib'. 'Sepidrood' is an improved rice variety from IRRI, a dwarf and high yielding plant that is resistant to blast and lodging. 'Gharib' is a native rice variety that is cultivated mainly in Guilan province. It has a low yield and is susceptible to lodging. The phenotyping experiment was conducted at the experimental farm of the College of Agriculture, Guilan University, Rasht, Iran. F_4 plants consisting of 196 individuals were grown in an experimental field with a spacing of 25 cm × 25 cm and were scored for: number of filled grains per panicle (NGP), days to 50% flowering (FD), days to maturity (DM), plant height (PH), panicle length (PL), number of panicles per plant (NPP), number of spikeletes per panicle (NSP), 1000-grain weight (GW), fertility percentage (FP), number of empty spikelets per panicle (NES) and grain yield (GY).

DNA extraction, SSR and AFLP analysis

Fresh leaves were used for DNA extraction according to CTAB method (Saghai-Maroof *et al*., 1984) with slight modifications. In this study a total of 550 SSR DNA markers distributed throughout the rice genome were selected and primer sequences were obtained from Gramene (http://www.gramene.org). Sixty AFLP primer combinations were tested to find the polymorphic markers between the parents. From these, 105 SSR and 10 AFLP polymorphic markers (these 10 primer combinations made 107 polymorphic bands) were chosen. The selected SSR DNA primers were tested for polymorphism on the parents and the F_2 population. PCR conditions were as described in Panaud *et al*. (1996). In summary, 10 µl PCR reactions contained 0.4 mM of each primer, 10 µM deoxyribonucleotides, 50 mM KCl, 10 mM TRIS-Cl (pH 8.3), 1.5 mM $MgCl_2$, 0.01% gelatin, 50–100 ng of DNA, and 1 unit of Taq polymerase. The PCR was performed with a profile of 94°C for 5 min, followed by 35 cycles at 94°C for 1 min, at 55°C for 1 min, at 72°C for 2 min, and finally for 5 min at 72°C for the final extension. Next, a total of 3µl PCR products were denatured and run on 6% polyacrylamide denaturing gels, and marker bands were revealed using the silver staining as that described by Panaud *et al*. (1996). The PCR products were separated on denaturing 6% polyacrylamide gels and the bands were revealed using the silver-staining protocol (Panuad *et al*., 1996). The primers used for AFLP analysis are listed in Table 1.

Table 1. AFLP primers used for QTL mapping.

Primer	Sequence
MseI	5'-GATGAGTCCTGAGTAA-3'
EcoRI	5'-GTAGACTGCGTACCAATTC-3'
MseI adaptor	5'-GACGATGAGTCCTGAG-3'
MseI adaptor	3'-TACTCAGGACTCAT-5'
EcoRI adaptor	5'-CTCGTAGACTGCGTACC-3'
EcoRI adaptor	3'- CATCTGACGCATGGTTAA -5'
E32 Primer	5'-GTAGACTGCGTACCAATTC**AAC**-3'
E33 Primer	5'-GTAGACTGCGTACCAATTC**AAG**-3'
E35 Primer	5'-GTAGACTGCGTACCAATTC**ACA**-3'
M44 Primer	5'-GATGAGTCCTGAGTAA**ATC**-3'
M42 Primer	5'-GATGAGTCCTGAGTAA**AGT**-3'
M39 Primer	5'-GATGAGTCCTGAGTAA**AGA**-3'
M62 Primer	5'-GATGAGTCCTGAGTAA**CTT**-3'
M61 Primer	5'-GATGAGTCCTGAGTAA**CTG**-3'
M33 Primer	5'-GATGAGTCCTGAGTAA**AAG**-3'
M59 Primer	5'-GATGAGTCCTGAGTAA**CTA**-3'
M60 Primer	5'-GATGAGTCCTGAGTAA**CTC**-3'
M93 Primer	5'-GATGAGTCCTGAGTAA**TTG** -3'
M90 Primer	5'-GATGAGTCCTGAGTAA**TGT** -3'

Linkage map construction and QTL analysis

The linkage map of the F_2 population was constructed using Map manager QTXb17 (Manly and Olson, 1999), based on the segregation data of 105 SSR and 107 AFLP loci in the F_2 population. The related genomic distances (cM) were calculated from recombination values using the Kosambi mapping function (Kosambi, 1944). In order to map the QTLs, composite interval mapping was conducted using QTL cartographer version 2.5 (Basten *et al*., 2001). To determine empirical significance thresholds for declaring a QTL, 1000 permutations were done to calculate LOD thresholds for each trait at p=0.05 and p=0.01. Permutation testing (using QTL cartographer) indicated that a logarithm of the odds ratio (LOD) score of ≥2.5 is a suitable threshold for this data.

Results

Phenotypic evaluation of F_4 population

Table 2 shows the phenotypic values of studied traits in each parent (Sepidrood and Gharib) and F_4 lines. The t-tests showed that there were significant differences between the parents for

Table 2. Phenotypic values of parents, F_4 population, and heritability of measured traits in rice

Traits	Parents		F4 ($m \pm sx$)	t ($\|P_1\text{-}P_2\|$)
	Sepidrood ($m \pm sx$)	Gharib ($m \pm sx$)		
NSP	147.37 ± 0.77	109.32 ± 0.88	127.80 ± 2.07	32.58**
NGP	126.12 ± 0.78	95.74 ± 0.63	66.44 ± 1.58	30.19**
NES	21.37 ± 0.57	13.58 ± 0.54	61.38 ± 1.92	9.92**
GW (gr)	28.82 ± 0.73	24.14 ± 0.35	24.43 ± 0.37	5.73**
GY (ton/ha)	5.82 ± 0.09	3.84 ± 0.09	3.88 ± 0.13	16.41**
PH (cm)	98.43 ± 0.95	139.31 ± 0.50	113.54 ± 1.28	38.07**
PL (cm)	28.01 ± 0.22	30.74 ± 0.39	26.07 ± 0.29	6.04**
NPP	20.58 ± 0.36	12.88 ± 0.57	15.70 ± 0.29	11.38**
FP (%)	85.51 ± 0.37	87.61 ± 0.42	52.65 ± 1.08	3.78**
DF	73.48 ± 0.28	65.74 ± 0.21	69.79 ± 0.43	21.97**
DM	118.68 ± 0.25	110.63 ± 0.22	116.09 ± 0.43	23.99**

The symbols of traits are: number of spikelet per panicle (NSP), number of filled grain per panicle (NGP), number of empty spikelets per panicle (NES), 1000-grain weight (GW), grain yield (GY), plant height (PH), number of panicles per plant (NPP), fertility percentage (FP), days to 50% flowering (DF) days to maturity (DM).

Table 3. Genotypic (above diagonal) and phenotypic (below diagonal) correlation coefficients among traits in the 196 F_4 rice lines

Traits	GP	FG	HG	GW	GY	PH	PL	PP	FR	DH	MD
NSP	1	0.50	0.67	-0.16	0.35	0.12	0.17	0.05	-0.16	-0.01	-0.01
NGP	0.49	1	-0.31	-0.09	0.67	0.17	0.12	-0.07	0.75	-0.02	-0.03
NES	0.69	-0.38	1	-0.1	-0.19	-0.01	-0.09	0.12	-0.82	0.01	0.01
GW	-0.15	-0.08	-0.09	1	0.18	0.07	0.11	-0.14	0.02	-0.31	-0.3
GY	0.34	0.66	-0.19	0.19	1	-0.03	0.09	0.55	0.51	0.02	0.01
PH	0.12	0.17	-0.01	0.07	-0.03	1	0.46	-0.28	0.07	-0.29	-0.32
PL	0.17	0.12	0.08	0.1	0.09	0.46	1	-0.05	0.01	-0.06	-0.08
NPP	0.05	-0.07	0.12	-0.13	0.55	-0.28	-0.05	1	-0.09	0.2	0.19
FP	-0.18	0.74	-0.81	0.02	0.51	0.07	0.01	-0.91	1	0	-0.01
DH	0	-0.02	0.01	-0.29	0.2	-0.28	-0.06	0.19	0	1	0.96
DM	-0.01	-0.03	0.01	-0.29	0.01	-0.31	-0.08	0.08	-0.01	0.97	1

The symbols of traits are: number of spikelet per panicle (NSP), number of filled grain per panicle (NGP), number of empty spikelets per panicle (NES), 1000-grain weight (GW), grain yield (GY), plant height (PH), number of panicles per plant (NPP), fertility percentage (FP), days to 50% flowering (DF) days to maturity (DM).

all studied traits at 1% probability level. For grain weight and grain yield, 'Sepidrood' was superior to 'Gharib'. On the other hand 'Gharib' had a relative superiority to 'Sepidrood' for fertility percentage, plant height and panicle length traits. Skewness and kurtosis tests were applied to the distribution of the traits studied. In the F4 families, all the traits showed continuous variation with transgressive segregation (Figure 1). This type of study can give information about the inheritance of the traits. Therefore, continuous and normal distribution of phenotypic traits can be the reason for the quantitative inheritance of them.

Trait correlation

Correlation among traits was evaluated at P<0.05 and P<0.01. As summarized in Table 3, the strongest correlation was found between yield and number of spikeletes per panicle, with significant correlations also found between yield and number of filled grains per panicle, and fertility rate. As expected, a negative correlation was found between yield and number of hollow grains per panicle. The frequency distribution of phenotypes for each trait in the F_4 lines is shown in Figure 1. Most of the traits showed continuous variation. Some of the lines fell within the range of the parents, however, transgressive segregations were observed for most of the traits.

QTL mapping

The linkage map was constructed using Map manager QTXb17 software. The integrated molecular linkage map consisted of 107 AFLP markers and 105 SSR markers covering 1861.3 cM in length with an average distance of 8.95 cM between adjacent markers. A total of 28 QTLs were mapped for 11 traits of yield, yield component and plant morphology (Table 4, Figure 2). These QTLs were detected on chromosomes 1, 2, 3, 6, 7, 8, 11 and 12 using CIM analysis and explained 0.16–26.12% of the phenotypic variances. Nine of the QTLs had high general contribution of over 10%. One QTL was mapped for number of spikeletes per panicle (NSP). This QTL (*NSP*-12) was located on chromosome 12 (between RM2197-RM212) and explained 22.58% of phenotypic variation. Increased number of spikeletes per plant was associated with 'Gharib' alleles. Three QTLs were detected for number of filled grains per panicle (NGP): these QTLs were mapped on chromosomes 1, 6 and 11, respectively. Among these, *qNGP-1* on chromosome 1 (between RM246-RM1268) explained 21.03% of phenotypic variation and became the major QTL for this trait. Except *qNGP-6,* for other QTLS, the allele from 'Sepidrood' increased the value for number of filled grains per panicle (NGP). Three QTLs for number of empty spikelets per panicle (NES) were

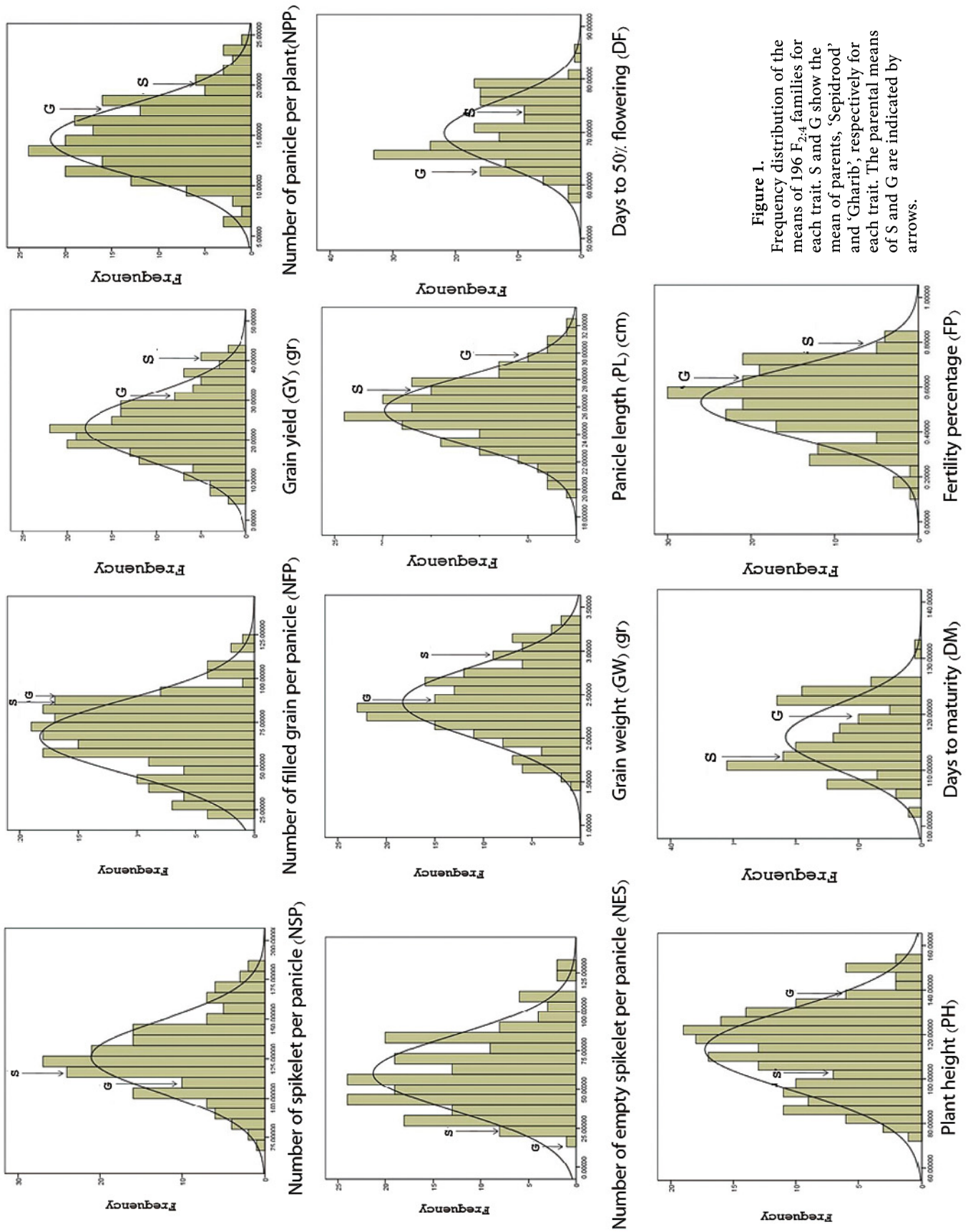

Figure 1.
Frequency distribution of the means of 196 $F_{2:4}$ families for each trait. S and G show the mean of parents, 'Sepidrood' and 'Gharib', respectively for each trait. The parental means of S and G are indicated by arrows.

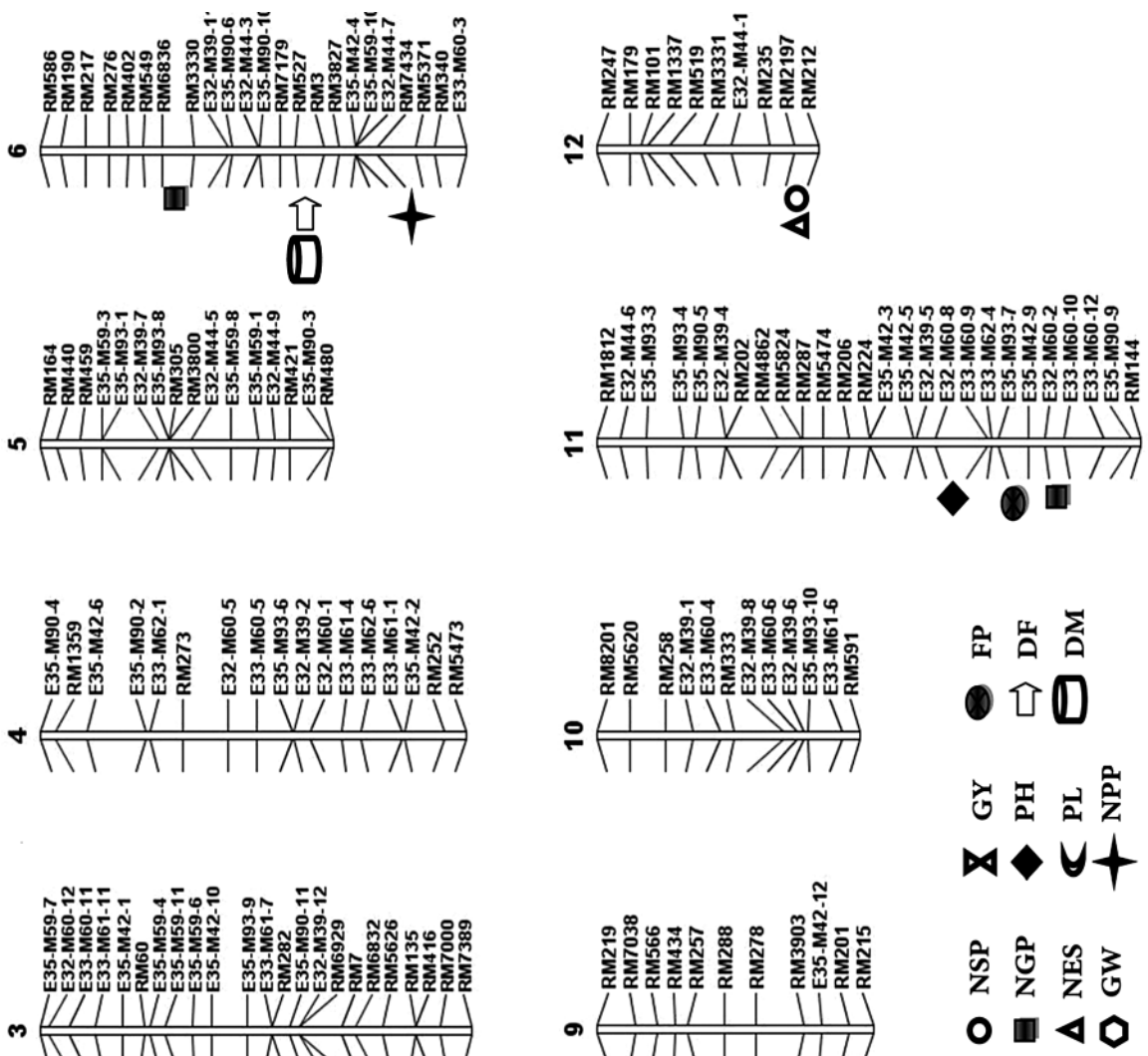

Figure 2.
Linkage map and QTLs related to agronomic traits in the $F_{2:4}$ population from the cross GHB × SPD. The number of chromosomes is shown at the top and markers are indicated at the right sides of the chromosomes, respectively. The QTLs for 11 studied traits are indicated on the chromosomes. Number of spikelet per panicle (NSP), number of filled grain per panicle (NGP), number of empty spikelets per panicle (NES), 1000-grain weight (GW), grain yield (GY), plant height (PH), number of panicles per plant (NPP), fertility percentage (FP), days to 50% flowering (DF) days to maturity (DM)

Table 4. Putative QTLs for agronomic traits in $F_{2:4}$ population derived from the cross of two rice varieties (Gharib × Sepidroud)

Trait	QTL[a]	Flanking markers	Chr	LOD	a[b]	d[c]	d/a	PEV[d]
NSP	*qNSP-12*	RM2197 - RM212	12	11.66	-13.92	-1.18	-0.08	22.58
NGP	*qGP-1*	RM246 - RM1268	1	11.95	-9.06	-26.35	-2.91	21.03
	qGP-6	RM6836 - RM3330	6	2.86	1.62	-18.21	-11.24	2.13
	qGP-11	E32-M60-2 - E33-M60-10	11	3.8	9.78	-23.45	-2.39	7.19
NES	*qES-2*	E32-M39-9 - E32-M60-6	2	2.77	-7.73	-21.89	-3.02	5.05
	qES-3	E33-M61-11 - E35-M42-1	3	9.07	-7.55	-9.57	-1.27	15.23
	qES-12	RM2197 - RM212	12	2.59	-3.75	-12.10	-3.23	2.6
GW	*qGW-1*	RM1 - RM283	1	2.89	1.48	0.44	0.29	28
	qGW-2	E32-M39-9 - E32-M60-6	2	11.89	0.55	0.95	1.72	9.68
GY	*qGY-3*	RM5626 - RM135	3	5.57	-2.79	-7.76	-2.81	10.26
PH	*qPH-1*	RM237 - RM246	1	2.03	-10.56	-5.73	-0.54	2.97
	qPH-7a	RM3608 - RM234	7	3.83	-0.19	-23.64	-124.4	5.62
	qPH-7b	RM1243 - RM8263	7	6.25	6.36	-22.03	-3.46	26.12
	qPH-8	RM3572 - RM404	8	2.06	-2.09	-17.53	-8.38	2.44
	qPH-11	E32-M60-8 - E33-M60-9	11	3.81	9.38	12.24	1.30	4.03
PL	*qPL-1*	RM283 - RM8132	1	4.21	-0.57	3.01	5.28	9.61
NPP	*qNPP-1*	RM283 - RM8132	1	6.64	0.15	-3.84	-25.6	19.11
	qNPP-3	E32-M39-10 - E32-M44-1	3	4.93	1.89	-2.93	-1.55	10.54
	qNPP-6	RM7434 - RM5371	6	3.83	-3.69	1.86	0.51	9.65
FP	*qFP-11*	E35-M93-7 - E35-M42-9	11	4.82	0.11	0.07	0.63	6.21
DF	*qDF-2*	RM3340 - RM154	2	2.63	3.33	5.52	1.67	0.17
	qDF-3a	RM6832 - RM5626	3	5.69	-4.29	4.6	1.07	10
	qDF-3b	RM7000 - RM7389	3	3.12	-0.67	10.40	15.75	9.74
	qDF-6	RM527 - RM3	6	7.45	2.83	-8.51	-3	19.09
DM	*qDM-2*	RM3340 - RM154	2	2.51	3.35	5.55	1.65	0.16
	qDM-3a	RM6832 - RM5626	3	5.48	-4.37	4.43	1.01	9.94
	qDM-3b	RM7000 - RM7389	3	3.03	-0.71	9.93	13.08	8.78
	qDM-6	RM527 - RM3	6	6.49	-2.63	-8.59	-3.26	16.21

[a] QTLs are named by abbreviations plus chromosomal number; [b] Additive effect; [c] Dominance effect; [d] Percentage of total phenotypic variance explained by the QTL; Number of spikelet per panicle (NSP), number of filled grain per panicle (NGP), number of empty spikelets per panicle (NES), 1000-grain weight (GW), grain yield (GY), plant height (PH), number of panicles per plant (NPP), fertility percentage (FP), days to 50% flowering (DF) days to maturity (DM)

identified. These QTLs were mapped on chromosomes 2, 3 and 12 respectively. The QTL (*NES*-2) on chromosome 2 (between E33-M61-11-E35-M42-1), with LOD= 9.07 was the major effective QTL for this trait (R^2 = 15.23%). The alleles from 'Gharib' increased the empty spikelets for this QTL. Two QTLs for 1000 grain weight were detected. The first QTL on chromosome 1 (*qGW*-1) between RM1-RM283, having an increasing allele from 'Sepidrood', explained 28.34% of phenotypic variation and showed the greatest effects on this trait. The second QTL on chromosome 2 (*qGW*-2) explained 9.68% of phenotypic variation. The alleles from Sepidrood decreased 1000 grains weight. One QTL was identified for grain yield. The mentioned QTL was located on chromosome 3. *qGY*-3, explained 10.26% of the phenotypic variation and the alleles from 'Gharib' increased grain yield. Five QTLs were detected for plant height on chromosomes 1, 7 (two QTLs), 8 and 11 respectively: the 'Gharib' allele increased the plant height at two loci (*qPH-7b* and *qPH-11*) and 'Sepidrood' alleles increased plant height at other loci. The QTL (*qPH-7b*) mapped on chromosome 7 (between RM1243-RM8263), explained 26.12% of the phenotypic variation and it means that it was the major QTL for this trait. Only one QTL was mapped for panicle length. This QTL was mapped on chromosome 1 (between RM283-RM8132). *qPL-1* on chromosome 1 explained 9.61% of phenotypic variance. 'Sepidrood' allele increased the panicle length for this QTL. Three QTLs were mapped for number of panicle per plant. These QTLs were mapped on chromosomes 1, 3 and 6. Among them, *qNPP-1* with LOD = 6.64, located on chromosome 1, explained 19.11% of phenotypic variation. In this study QTL allele from 'Sepidrood' increased NPP by 0.15. Only one QTL was mapped for fertility rate having an LOD scores of 4.82 and contribution of 6.21%. Four QTLs were detected for days to 50% flowering on chromosomes 2, 3 (two QTLs) and 6, accounting for 0.17%, 10%, 9.74% and 19.09% of the total trait variances, respectively. The major effect QTL (*qDF-6*) on chromosome 6 (between RM527-RM3) explained 19.09% of the phenotypic variation. Four QTLs were mapped for days to maturity. These QTLs were located on chromosomes 2, 3 (two QTLs) and 6, respectively. The alleles contributing to the increase were from 'Gharib' except for loci *qMD-2*.

Discussion

Recognizing the enormous potential of DNA markers in plant breeding, many agricultural research centers have adopted the capacity for marker-assisted selection (MAS). However, due to rapid developments in marker technology, statistical methodology for identifying quantitative trait loci (QTLs) and the jargon used by molecular biologists, the utility of DNA markers in plant breeding may not be clearly understood by non-molecular biologists (Collard *et al*., 2005). In present study, the $F_{2:4}$ population was evaluated for eleven yield related traits. The absolute value of skewness and kurtosis for all traits was less than 1 (data not shown) indicating their normal distribution and thereby being

suitable for QTLs analysis. A total of 28 QTLs were identified with the significance threshold equivalent to LOD>2.5. One putative QTL was mapped for number of spikelets per panicle (NSP). This QTL (*qNSP-12*) explained 22.58% of phenotypic variation. Thompson *et al*. (2003) mapped QTLs for yield, yield components and morphological traits in an advanced backcross population between an accession of *Oryza rufipogon* (IRGC 105491) and the U.S. cultivar Jefferson (*Oryza sativa* ssp. japonica). The QTL on chromosome 12 may represent the same locus *qNSP-12* in the present study and explained 7.4% of total phenotypic variations. Our results showed that this QTL had additive effects. This result is contrary to that of Thompson *et al*. (2003) and is the same as those reported by Xiao *et al*. (1998) and Fu *et al*. (2010).

Three QTLs were mapped for number of filled grains per panicle (NGP). QTL (*NGP-1*) on chromosome 1 explained 21.03% of phenotypic variation. Moncada *et al*. (2001) reported the same chromosomal region for NGP. Mu *et al*. (2008) detected that the NGP related QTLs are distributed on chromosomes 1, 2, 3, 4, 6, 7 and 11, respectively. Our results showed that the QTLs controlling NGP had partial dominance effects that are contrary to the results of Fang-Ming *et al*. (2008). They believed that the NGP related QTLs have over dominance effects. Our results are the same as those of Sabouri *et al*. (2010). They believe that the related QTLs have partial dominance effects. The number of QTLs mapped in this research was lower than that reported in other studies. Sabouri *et al*. (2010) and Tan *et al*. (2008) mapped an average number of 5 QTLs for this trait. This can be the result of population type and genetic background, type and the number of molecular markers that were used in this study. Three QTLs for empty spikelets per panicle (NES) were mapped. These three QTLs were mapped on chromosomes 2, 3 and 12, respectively. The previous studies (Sabouri *et al*., 2009) showed that the related QTLs are distributed on chromosomes 1, 7 and 10. In other studies, no QTLs were found on chromosome 12 (in this location) that show the novelty of this QTL.

Two putative QTLs were mapped for 1000 grain weight on chromosomes 1 and 2. These QTLs had both partial dominance (*qGW-1*) and over dominance effects (*qGW-2*). One QTL for grain yield was located on chromosome 3 that explained 10.26% of the phenotypic variation. As seen, the number of QTLs for this trait is limited. Our results were similar to those of Xiao *et al*. (1998), Lin *et al*. (1996) and Benmousa *et al*. (2005). The locations of the QTLs for grain yield are different in other researches. Biological complexity of grain yield can cause these different results. Five QTLs were mapped for plant height (PH) on chromosomes 1, 7 (two QTLs) and 11, respectively. The QTL on chromosome 7 showed the major effect. These findings conform to that of Sabouri *et al*. (2010). Only one QTL was mapped for PL on chromosome 1, while Sabouri *et al*. (2010) reported four QTLs for the trait PL in the $F_{2:3}$ population from 'Sepidrood' × 'Gharib' cross. They mapped these QTLs on chromosomes 1, 4, 8 and 12. This shows stability of the *qPL-1* in this population. Ahmadi *et al*. (2008) reported a QTL with major effects for this trait in the same location on chromosome 1 indicating the expression and stability of *qPL-1* in different populations as well as environments.

Three QTLs were mapped for NPP. The QTL with major effects (*qNPP-1*) was identified on chromosome 1. Yuan *et al*. (2003) reported that NPP related QTLs are distributed on chromosomes 2, 3, 4, 7, 11 and 12. Sabouri *et al*. (2010) mapped a QTL for NPP on the same chromosome (in the same population), but the marker interval was different. The differences may be from environmental factors and their effects on QTL mapping. Austin *et al*. (1998) believed that different alleles at a locus are responsible for genetic variation under different environmental conditions. One QTL was identified for FP on chromosome 11 with minor effects. Tan *et al*. (2008) and Wen-Qiang *et al*. (2009) mapped the same QTLs. Several studies also found that different chromosomal regions are responsible for controlling these traits (Rahman *et al*., 2007; Hittalmani *et al*., 2003). So it can be concluded that the genes controlling fertility percentage (FP) have not been expressed in all environments to the same extent. Four QTLs were mapped for DF (*qDF-2*, *qDF-3a*, *qDF-3b* and *qDF-6*). Of all cereals, the genes that control flowering time are best understood in rice. Control of flowering in response to day-length has been analyzed extensively, and interestingly the genes cloned to date relate directly to known genes in Arabidopsis (Graham *et al*., 2006). Rabiei (2007) mapped six QTLs for DF on chromosomes 1, 3, 6, 7, 8 and 11. The *qDF-3* explaining 11.7% of phenotypic variation was in the same location as in this study. Moncada *et al*. (2001) using the genetic background of *Oryza rufipogon* reported four QTLs controlling DF, located on chromosomes 2, 3, and 7 explaining 6-14% of the phenotypic variation. Cui *et al*. (2004) using Zenshan97/ Minghui63 reported six QTLs controlling DF on chromosomes 6, 7, 10, and 11 explaining 59.69% of phenotypic variation. Lin *et al*. (2011) believed that the diversity of positions associated with the heading date QTLs in different studies comes from allelic variations. Allelic variations on flowering exist in many cultivars under artificial or natural selection. So attempts to identify and determine QTL associated with these traits using different populations and pedigrees, not only reveal new genes, but also provide the conditions for effective identification of genes for map-based cloning. Four QTLs were identified for DM located on chromosomes 2, 3 (two QTLs), and 6. The QTL analysis of DF and DM showed that these traits are controlled by one major effect QTL and multiple minor effects QTLs. Similar results were reported by Lin *et al*. (1998) and Yamatoto *et al*. (2001).

Conclusions

The results showed that most of the studied traits in this study were controlled by one or more major QTLs explaining high percentage of phenotypic variance and several minor QTLs. The major QTLs can be considered in breeding programs. Several genomic regions were associated with more than one trait, indicating linkage and/or pleiotropic effects. For example, on chromosome 12 there are QTLs for number of spikelets per panicle and number of empty spikelets per panicle, or on chromosome 1, there are QTLs for number of panicles per plant and panicle length. The significant correlations between studied traits can be explained by these genomic regions containing pleiotropic or tight linkage QTLs.

References

Chun O. K. (2005). Daily consumption of phenolics and total antioxidant capacity from fruit and vegetables in the American diet. J Sci Food Agric 85(2):1715-1724.

Austin D. F., Lee M. (1998). Detection of quantitative trait loci for grain yield and yield components in maize across generations in stress and nonstress environments. Crop Sci 38(5): 1296-1308.

Basten C. J., Weir B. S., Zeng Z. B. (2001). QTL Cartographer: a reference manual and toturial for QTL mapping. North Carolina State University. USA, 163p.

Benmoussa M, Achouch A, Snoussi SA, Zhu J (2005). Conditional QTL analysis of genetic main effects and genotype × environment interaction effects for yield in rice (*Oryza sativa* L.). Food, Agr & Environ 4 (1): 157-162.

Collard B. C. Y., Jahufer M. Z. Z., Brouwer J. B., Pang E. C. K. (2005). An introduction to markers, quantitative trait loci (QTL) mapping and marker assisted selection for crop improvement: The basic concepts. Euphytica 142: 169–196.

Collard B. C. Y., Vera Cruz C. M., McNally K. L., Virk P. S., Mackill D. J. (2008). Rice molecular beeding laboratories in the genomics era: current status and future considerations. Int J Plant Genomics. Article ID 524847, 25 pp.

Cui K., Peng S., Ying Y., Yu S., Xu C. (2004). Molecular dissection of the relationship among tiller number, plant height and heading date in rice. Plant Prod Sci 7(3): 309-318.

Fang ming Z., Gui F. L., Hai T. Z., Xiao H. D., Rui H. Z., Ze M. Z., Wen tao L., Gui quan Z. (2008). Unconditional and conditional QTL mapping for tiller numbers at various stages with single segment substitution lines in rice (*Oryza sativa* L.). Plant and Soil 236: 237–242.

Fu Q., Zhang P., Tan L., Zhu Z., Ma D., Fu Y., Zhan X., Cai H., Sun C. (2010). Analysis of QTLs for yield-related traits in Yuanjiang common wild rice (*Oryza rufipogon* Griff.). J Genet Genomics 37: 147–157.

Graham J., Smith K., Tierney I., MacKenzie K., Hackett C. A. (2006). Mapping gene H and association with resistance to cane botrytis and spur blight, rust and cane spot. Theor Appl Genet 112: 818-831.

Hittalmani S., Huang N., Courtois B., Venuprasad R., Shashidhar H. E., Zhuang J. Y., Zheng K. L., Liu G. F., Wang G. C., Sidhu J. S., Srivantaneeyakul S., Singh V. P., Bagali P. G., Prasanna H. C., McLaren G., Khush G. S. (2003). Identification of QTL for growth and grain yield related traits in rice across nine locations of Asia. Theor Appl Genet 107: 679–690.

Jena K. K and Mackill D. J. (2008). Molecular markers and their use in marker assisted Selection in Rice. Crop Sci 48:1266–1276.

Jiming G., Xianwu Z., Baoxing D., Qian Q., Shouyi C., Lihuang Z., Ping H. (2001). Comparative study of QTLs for agronomic traits of rice (Oriza sativa L.) between salt stress and nonstress environment. Sci China 44:1 73-82.

Jing Z., Qu Y., Chen Y., Pan D., Fan Z., Chen J., Li C. (2010). QTL Analysis of Yield-relatedTraits using an Advanced Backcross Population Derived from Common Wild Rice (*Oryza rufipogon L*). Mol Plant Breeding 1: 1-10.

Kosambi D. D. (1944). The estimation of map distances from recombination values. Ann Eugenic 12: 172–175.

Lin H.X., Qian H.R., Zhuang J.Y., Lu J., Min S.K., Xiong Z.M., Huang N., Zheng K. L. (1996). RFLP mapping of QTLs for yield and related characters in rice (*Oryza sativa* L.). Theor Appl Genet 92: 920-927.

Lin Y. R., Wu S. C., Chen S., Tseng T. H., Chen C. S., Kuo S. C., Wu H. P., Hsing Y. C. (2011). Mapping of quantitative trait loci for plant height and heading date in two inter-subspecific crosses of rice and comparison across *Oryza* genus. Bot Stud 52: 1-14.

Mackill D. J. and McNally K. L. (2004). A model crop species: molecular markers in rice. In: Lörz H Wenzel G (ed.) Molecular Marker Systems in Plant Breeding and Crop Improvement. Springer Verlag, Heidelberg, p. 39–54.

Manly K. F., Olson J. M. (1999). Overview of QTL mapping software and introduction to map manager QTL. Mamm Genome b: 327–334.

Meudt H. M., Clarke A. C. (2007). Almost forgotten or latest practice? AFLP applications, analyses and advances. Trends Plant Sci 12: 106–117.

Moncada P., Martinez C. P., Borrero J., Chatel M., Gauch H., McCouch S. R. (2001). Quantitative trait loci for yield and yeild components in an *Oryza sativa* x *Oryza rufipogon* BC_2F_2 population evaluated in an upland environment. Theor Appl Genet 102: 41-52.

Mu P., Huang C. J., Li X. L., Liu F., Li Z. C. (2008). Yield trait variation and QTL mapping in a DH population of Rice under phosphorus deficiency. Acta Agron. Sin 34 (7): 1137-1142.

Panaud O., Chen X., McCouch S. R. (1996). Development of Microsatellite Markers and Characterization of Simple Sequence Length Polymorphism (SSLP) in Rice (*Oryza sativa* L.). Mol Gen Genet 252: 597–607.

Rabiei B. (2007). Linkage map of SSR markers and QTLs detection for heading date of Iranian rice cultivars. J Agr Sci Technol 9: 235-242.

Rahman L., Chu S. H., Choi M. S., Qiao Y. L., Jiang W., Piao R., Khanam S., Cho Y. I., Jeung J. U., Jena K. K., Koh H. J. (2007). Identification of QTLs for some agronomic traits in Rice using an introgression line from *Oryza minuta*. Mol Cells 24(1): 16-26.

Saghai-Maroof M. A., Soliman K. M., Jorgesen R. A. Allard R. W. (1984). Ribosomal DNA spacer-length polymorphisms in barley: Mendelian inheritance, chromosomal location and population dynamics. Proceedings of National Academy of Sciences of the United States of America. 81: 8014-8018.

Sabouri H., Sabouri A., Dadras A. R. (2009). Genetic dissection of biomass production, harvest index and panicle characteristics in indica-indica crosses of Iranian rice (*Oryza sativa* L.) cultivars. Australian J Crop Sci 3(3):155-166.

Sabouri A., Toorchi H., Rabiei B., Aharizad S., Momoeni A., Singh R. K. (2010). Identification and Mapping of QTLs for Agronomic Traits in indica – indica Cross of Rice (*Oryza sativa L.*). Cereal Res Commun 38(3): 317–326.

Tan L., Zhang P., Liu F., Wang G., Ye S., Zhu Z., Fu Y., Cai H., San C. (2008). Quantitative trait loci underlying domestication and yield related in an *Oryza sativa* × *Oryza rufipogon* advanced backcross population. Genome 51: 692-704.

Thompson M. J., Tai T. H., McClung A. M., Lai X. H., Hinga M. E., Lobos K. B., Xu Y., Martinez C. P., McCouch S. R. (2003). Mapping quantitative trait loci for yield, yield components and morphological traits in an advanced backcross population between *Oryza rufipogon* and the *Oryza sativa* cultivar Jefferson. Theor Appl Genet 107: 479–493.

Vos P., Hogers R., Bleeker M., Reijans M., Van de Lee T., Hornes M., Frijters A., Pot J., Peleman J., Kuiper M., Zabeau M. (1995). AFLP: a new technique for DNA fingerprinting. Nucl Acid Res 23: 4407–4414.

Wen-qiang L., Ye-yang F., Jie C., Yong-feng S., Jian-li W. (2009). Avoidance of Linkage Drag Between Blast Resistance Gene and the QTL Conditioning Spikelet Fertility Based on Genotype Selection Against Heading Date in Rice. Rice Sci 16(1): 21–26.

Xiao J., Li J., Grandillo S., Ahn S. N., Yuan L., Tanksley S. D., McCouch S. R. (1998). Identification of trait improving quantitative trait loci alleles from a wild rice relative, *Oryza rufipogon*. Genetics 150: 899-909.

Xing Y., Zhang Q. (2010). Genetic and Molecular Bases of Rice Yield. Annu Rev Plant Biol 61: 421-442.

Xu Y. B. (2010) Molecular Plant Breeding, 1st Ed. CAB International, 734 p.

Yamamoto T., Taguchi-Shiobara F., Ukai Y., Sasaki T., Yano M. (2001). Mapping quantitative trait loci for days to heading, and culm, panicle and internode lengths in a BC1F3 population using an elite rice variety, Koshihikari, as the recurrent parent. Breeding Sci 51: 63-71.

Yuan A. P., Cao L. Y., Zhuang J. Y., Li R. Z., Zheng K. L., Zhu J. (2003). Analysis of additive and AE interaction effects of QTLs controlling plant height, heading date and panicle number in rice (Oryza sativa L.). Acta Genet Sinica 30: 899-906.

acs80_13