Theor Appl Genet (1998) 96: 997–1003

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Mapping quantitative trait loci controlling seed dormancy and heading date in rice, *Oryza sativa* L., using backcross inbred lines

Received: 10 October 1997 / Accepted: 12 January 1998

Abstract To detect quantitative trait loci (QTLs) controlling seed dormancy, 98 BC_1F_5 lines (backcross inbred lines) derived from a backcross of Nipponbare (*japonica*)/Kasalath (*indica*)//Nipponbare were analyzed genetically. We used 245 RFLP markers to construct a framework linkage map. Five putative QTLs affecting seed dormancy were detected on chromosomes 3, 5, 7 (two regions) and 8, respectively. Phenotypic variations explained by each QTL ranged from 6.7% to 22.5% and the five putative QTLs explained about 48% of the total phenotypic variation in the BC_1F_5 lines. Except for those of the QTLs on chromosome 8, the Nipponbare alleles increased the germination rate. Five putative QTLs controlling heading date were detected on chromosomes 2, 3, 4, 6 and 7, respectively. The phenotypic variation explained by each QTL for heading date ranged from 5.7% to 23.4% and the five putative QTLs explained about 52% of the total phenotypic variation. The Nipponbare alleles increased the number of days to heading, except for those of two QTLs on chromosomes 2 and 3. The map location of a putative QTL for heading date coincided with that of a major QTL for seed dormancy on chromosome 3, although two major heading-date QTLs did not coincide with any seed dormancy QTLs detected in this study.

Communicated by G. Wenzel

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Key words Rice · Seed dormancy · Heading date · Backcross inbred line · QTL analysis

Introduction

Seed dormancy is one of the most important traits in the breeding programs of cereal crops because it is associated with pre-harvest sprouting (Seshu and Sorrells 1986). Seed dormancy is defined as the inability of viable seed to germinate under environmental conditions favorable to germination. However, it is difficult to measure such inability in seeds. Seed dormancy is usually estimated by germination rate, but complex inheritance of seed dormancy has been noted by breeders and geneticists (Takahashi 1997). In barley and wheat, seed dormancy is affected by environmental conditions during the ripening stage and by storage conditions, as well as by complex genetic factors (Anderson et al. 1993; Laura et al. 1995). The recent development of molecular markers has made it possible to identify individual genetic factors controlling such complex traits as seed dormancy (Tanksley 1993), and several genes affecting seed dormancy in barley and wheat which have been identified using these markers (Anderson et al. 1993; Flintham and Gale 1995; Laura et al. 1995; Larson et al. 1996).

In rice, however, only a few reports have been published on the genetics of seed dormancy (Seshu and Sorrels 1986; Das 1995; Takahashi 1997). No gene for seed dormancy with its chromosomal location has been reported. This might also be due to complex inheritance and environmental interactions. Recently, four rice molecular linkage maps have been developed by independent research groups (McCouch et al. 1988; Saito et al. 1991; Causse et al. 1994; Kurata et al. 1994). These linkage maps have facilitated analyses of quantitative trait loci (QTLs) controlling several complex traits, such as yield-related traits (Xiao et al. 1996), blast

resistance (Wang et al. 1994), heading date and plant height (Li et al. 1995; Yano et al. 1997), root morphology (Champoux et al. 1995), and seedling vigor (Redona and Mackill 1996). In the present study, we have developed BC_1F_5 lines (backcross inbred lines: BILs) as a permanent mapping population to facilitate QTL analysis in rice. We describe here the construction of a framework linkage map of the BILs and the identification of QTLs associated with seed dormancy and heading date using these BILs.

Materials and methods

Plant materials

A *japonica* variety, Nipponbare, was crossed with an *indica* variety, Kasalath. The resultant F_1 plant was crossed with Nipponbare to produce BC_1F_1 seeds. Ninety eight BC_1F_5 lines (backcross inbred lines: BILs) were developed from the resultant BC_1F_1 plants by the single-seed descent method. The 98 BILs and their two parental lines were sown on 19 April 1995. Their seedlings were transplanted on 26 May and were grown under natural conditions in Tsukuba, Japan (latitude 38°N). One individual from each BIL was selected and leaf tissue was collected for the extraction of total DNA. Heading date and seed dormancy were measured based on selected individuals.

Evaluation of seed dormancy and heading date

Seed dormancy was evaluated in terms of germination rate. Two panicles were collected from the selected individuals on the 40th day after heading and were immediately placed under conditions of 30°C and 100% relative humidity for 7 days. The germination rate of each selected individual was scored as a percentage of germinated seeds in the total numbers of seeds on the two panicles. To normalize the variances, the percentage germination rate of each individual was transformed to the arc sine [$= \arcsin(\chi)^{1/2}$] and the transformed data were used for QTL analysis. The day when the first panicle became exposed in the selected individual was scored as the heading date of the individual. The number of days required from seeding to heading (days to heading) was used in the QTL analysis.

RFLP analysis

Total DNA was extracted from the leaf tissue of each selected individual according to the CTAB method (Murray and Thompson 1980). The DNA was digested with eight restriction enzymes, *Bam*HI, *Bgl*II, *Eco*RV, *Hin*dIII, *Apa*I, *Dra*I, *Eco*RI and *Kpn*I. Electrophoresis and Southern blotting were performed according to Kurata et al. (1994). From the high-density linkage map we selected as probes 245 clones that were distributed evenly in the 12 chromosomes. Southern hybridization and detection were carried out according to the protocols of the ECL direct nucleic acid labelling and detection system (Amersham International).

Linkage analysis

Linkage groups and the order of markers were determined using MAPMAKER/EXP (Lander et al. 1987). Since MAPMAKER/EXP did not have an option for backcross inbred lines, the 'f2 intercross' mode was employed to determine the linkage groups and the marker orders in each linkage group. To avoid detecting pseudo-linkages of markers, relatively higher threshold levels (LOD > 3.0 and genetic distances < 18cM) were employed to establish linkage groups. The recombination frequency (r) of one meiosis between two markers was estimated from the formula, $r = 2w/(3 - 4w)$ (where 'w' is the frequency of observed recombinants between two markers). This formula was developed based on that for recombinant inbred lines (Haldane and Waddington 1931) with a slight modification. Genotype information of heterozygous individuals was treated as missing data for the estimation of recombination frequency.

The Kosambi mapping function was used to transform the recombination frequency to genetic distances (Kosambi 1944). The chi-square test was used to check the segregation ratios of individual markers against expected Mendelian inheritance ratios. The proportions of chromosome regions that were heterozygous or homozygous were estimated using the computer software HYPERGENE (Young and Tanksley 1989).

QTL analysis

Chromosomal locations of putative QTLs were determined by single-point analysis using the general linear model (GLM) procedure of SAS (SAS Institute 1989). The one-way ANOVA involved testing the significance of association at each locus between two genotype groups (homozygous for Nipponbare and Kasalath alleles) for two traits, seed dormancy and heading date. A probability level of 0.01 was employed as a threshold to detect significant mean differences between the two genotype groups for the two traits. MAPMAKER/QTL software (Lander and Botstein 1989; Lincoln et al. 1993) was also used to estimate the effects of Kasalath alleles on detected QTLs in the 'f2 backcross' mode. Two-way analysis of variance with the PROC GLM in SAS (SAS Institute 1989) was also performed to detect epistatic interaction between detected QTLs using genotype data of the nearest marker for each putative QTL.

Results

Linkage map and genome composition of the BILs

All 245 markers were grouped into 17 linkage groups using the 'f2 intercross' mode of MAPMAKER/EXP. The 17 linkage groups were further arranged into 12 linkage groups according to known linkage information on the high-density linkage map of Kurata et al. (1994). A linkage map with a total distance of 1179.9 cM was constructed (Fig. 1). The average distance between markers was 4.8 cM. All determined marker orders agreed with those of the published rice genetic map (Kurata et al. 1994).

The percentage of the Nipponbare genome in each line ranged from 49.3% to 91.3% with an average of 74.2%, which was not significantly different from the expected 75% (Fig. 2). The percentage of heterozygous regions ranged from 0.5% to 20.7% with a mean value of 4.6%, not significantly different from the expected 3.1% (Fig. 2).

The segregation ratios of the two genotype classes in most loci fitted the expected Mendelian ratio of 3 (Nipponbare) : 1 (Kasalath). Segregation distortion was observed for 12 loci on chromosomes 2, 3, 4 and 5, at $P < 0.01$ (Fig. 1). The frequency of Kasalath alleles

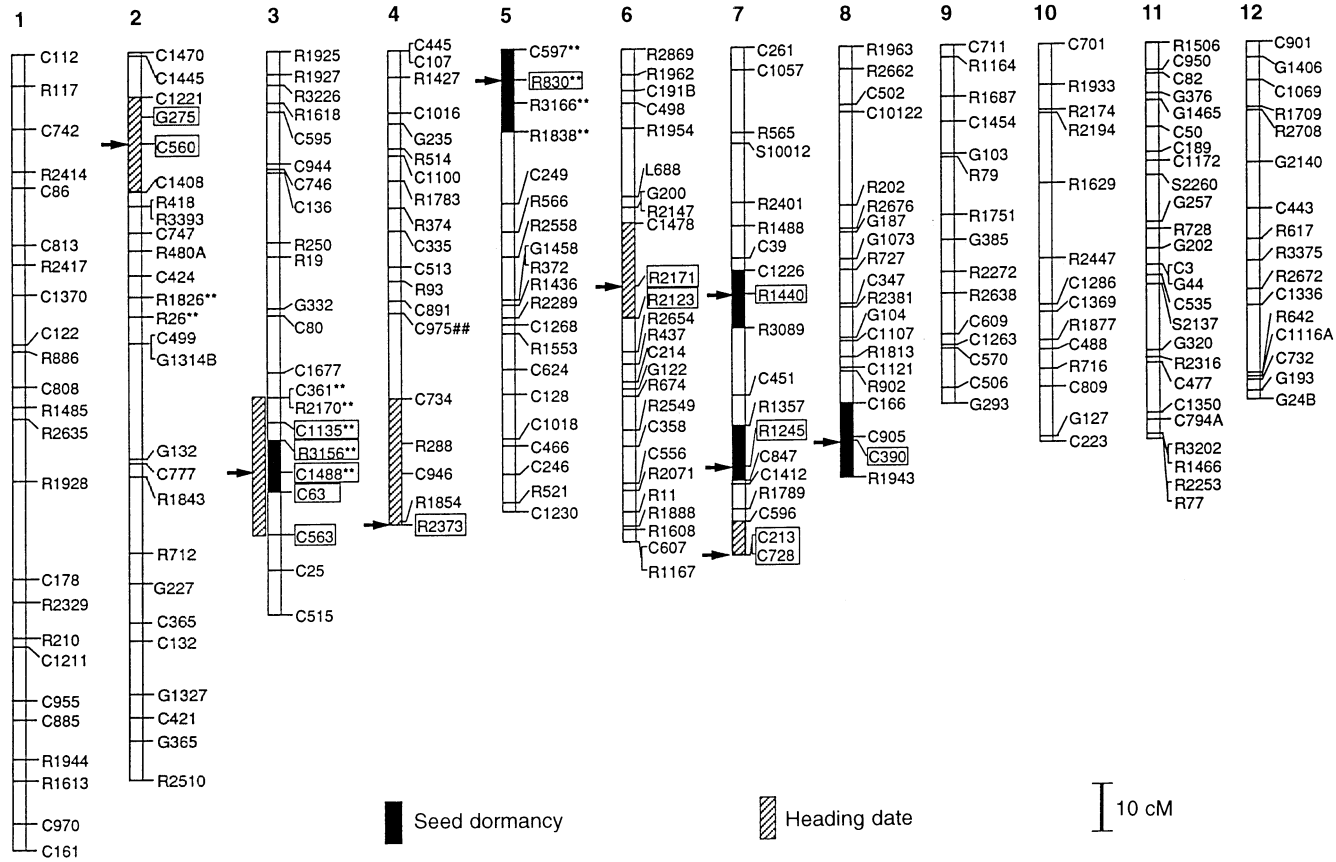

Fig. 1 Linkage map and the positions of QTLs for seed dormancy and heading date. *Black and striped bars* represent putative region of QTLs for seed dormancy and heading date, respectively. A reduction of 0.5 LOD value from LOD peaks was used to define left and right borders of the confidence interval in MAPMAKER/QTL. Based on ANOVA, markers which were significant at the 0.01 probability level are indicated in a *box*, *and arrows* indicate the nearest marker locus to the QTL in the markers analyzed. Loci with significant segregation distortion from the expected segregation ratio ($P < 0.01$) are marked with ** *or* # #; ** and # # show that the frequency of the Kasalath allele and the Nipponbare allele was increased in the region, respectively

increased in regions on chromosome 2, 3 and 5, while segregation distortion on chromosome 4 showed that the frequency of Nipponbare alleles increased.

Variations in seed dormancy and heading date in BILs

The germination rates of the two parents, Nipponbare and Kasalath, were 92% and 6.3%, respectively. The germination rates of BILs ranged from 0% to 100% with continuous variation (Fig. 3). Days to heading in Nipponbare and Kasalath were 122 and 113 days, respectively. Days to heading in BILs ranged from 106 to 146 days, also with continuous variation (Fig. 3). Transgressive segregants with late heading were observed. A significant negative correlation ($r = -0.49$, $P < 0.01$) was evident between germination rate and days to heading.

QTLs for seed dormancy

Five putative QTLs associated with the seed dormancy were detected based on ANOVA. These putative QTLs were located in the vicinity of C1488 on chromosome 3, R830 on chromosome 5, R1440 and R1245 on chromosome 7, and C390 on chromosome 8 (Fig. 1, Table 1). MAPMAKER/QTL also confirmed the existence of three putative QTLs in the vicinity of markers C1488, R1440 and R1245 using an empirical threshold of LOD = 2.0. However, the other two putative QTLs in the vicinity of markers R830 and C390 were not confirmed based on this threshold. The additive effects of Kasalath alleles in the five putative QTLs ranged from -6.9 to -12.8 in the arc-sine transformation of germination rate. Nipponbare alleles increased the germination rate of the putative QTLs on chromosome 3, 5 and 7, while Kasalath alleles increased the germination rate of the QTL on chromosome 8. The percentage of phenotypic variation explained by each QTL ranged from 7.0% to 25.6% based on GLM/SAS, and from 6.7% to 22.5% based on MAPMAKER/QTL. Total phenotypic variation explained by the five putative QTLs was 48.2% based on the multiple QTL model in

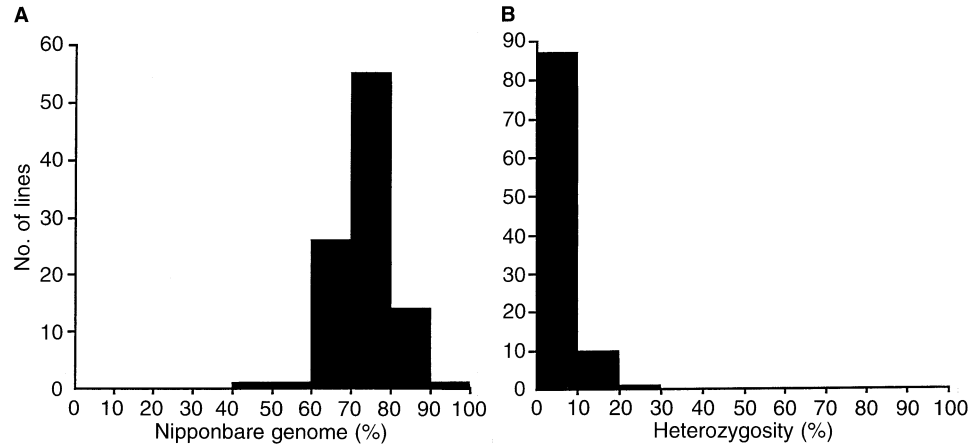

Fig. 2 Frequency distributions of the percentage of the Nipponbare genome (**A**) and heterozygosity (**B**) in the 98 backcross inbred lines

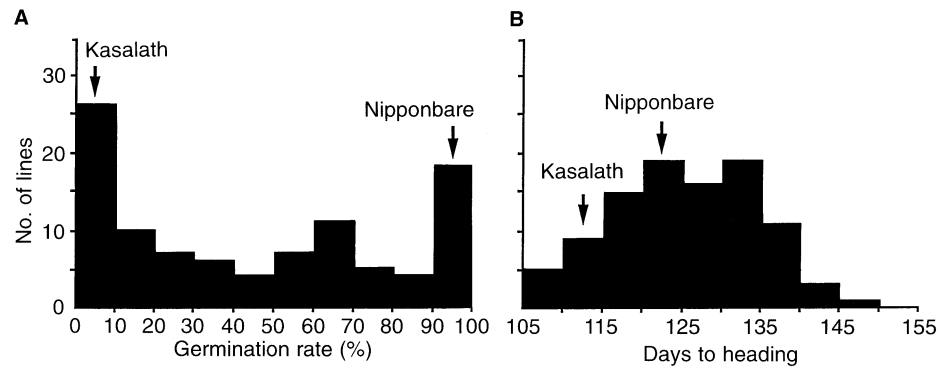

Fig. 3 Frequency distributions of germination rate (**A**) and days to heading (**B**) in 98 backcross inbred lines

Table 1 Putative QTLs of seed dormancy and heading date

	NML[a]	Chr	GLM/SAS		MAPMAKER/QTL			DPE[e]
			Probability	R^{2b}	LOD	% Variation[b]	AE	
Seed dormancy	C1488	3	< 0.0001	0.256	5.147	22.5	− 12.8[c]	N
	R830	5	0.0071	0.075	1.436	6.7	− 6.9	N
	R1440	7	0.0009	0.112	2.534	11.2	− 10.3	N
	R1245	7	0.0012	0.111	2.254	10.2	− 9.4	N
	C390	8	0.0091	0.070	1.500	6.9	9.5	K
					Total[f]	48.2		
Heading date	C560	2	0.0088	0.072	1.121	5.7	2.4[d]	K
	C1488	3	0.0035	0.094	1.377	6.4	2.5	K
	R2373	4	0.0051	0.081	1.427	6.6	− 2.6	N
	R2171	6	< 0.0001	0.276	5.141	23.4	− 5.9	N
	C728	7	< 0.0001	0.169	3.489	15.4	− 4.1	N
					Total[f]	52.3		

[a] Nearest marker locus of putative QTLs
[b] Phenotypic variation explained by each QTL
[c] Additive effects (1/2 weight) of Kasalath allele by arc sine of germination rate
[d] Additive effects (1/2 weight) of Kasalath allele by days to heading
[e] Direction of phenotypic effect. N and K indicate Nipponbare and Kasalath allele increased that values, respectively
[f] Estimates obtained from a multiple QTL model

MAPMAKER/QTL. Significant two-way interaction was not observed among the five putative QTLs.

QTLs for heading date

Five putative QTLs for heading date were identified based on ANOVA (Fig. 1, Table 1). These putative QTLs were located in the vicinities of C560 on chromosome 2, C1488 on chromosome 3, R2373 on chromosome 4, R2171 on chromosome 6, and C728 on chromosome 7. The two putative QTLs on chromosomes 6 and 7 were also confirmed by MAPMAKER/QTL. However, the other three putative QTLs on chromosomes 2, 3 and 4 were not confirmed using the empirical threshold of LOD = 2.0. The additive effects of the Kasalath alleles for the three QTLs on chromosomes 4, 6 and 7 were -2.6, -5.9 and -4.1 days, respectively. Additive effects of the QTLs on chromosomes 2 and 3 were 2.4 and 2.5 days, respectively. The percentage of phenotypic variation explained by the five detected QTLs ranged from 7.2% to 27.6% based on GLM/SAS. The percentage of phenotypic variation explained by these QTLs ranged from 5.7% to 23.4% and the total phenotypic variation explained was 52.3% based on the multiple QTL model in MAPMAKER/QTL (Table 1). Significant two-way interaction ($P < 0.01$) was observed between two QTLs located on chromosomes 6 (R2171) and 7 (C728). The lines exhibiting Nipponbare and Kasalath homozygotes at locus C728 showed little difference (115 and 116 days) in days to heading when both lines had a Kasalath homozygote at the R2171 locus, but large differences (121 and 131 days) were evident between these homozygotes at locus C728 when both lines had a Nipponbare homozygote in locus R2171. No other significant two-way interactions were found among the detected heading-date QTLs.

Discussion

Seed dormancy is usually affected by environmental factors such as temperature, dryness and degree of maturity (Seshu and Sorrells 1986; Takahashi 1995). These factors make the genetic study of seed dormancy complex because the grains of each individual in a given segregation population usually ripen at different times in different weather conditions. Recently, QTL analysis with molecular marker has made it possible to detect multigenes affecting a trait such as seed dormancy. In the present study, we successfully detected five chromosomal regions controlling seed dormancy based on QTL analysis. The results reported here are to our knowledge the first seed-dormancy genes located in rice chromosomes. The five QTLs detected accounted for 48.2% of the total phenotypic variation in seed dormancy. The unexplained remainder of the phenotypic variation might be due to environmental effects or to undetected QTLs with relatively smaller phenotypic effects.

The statistical threshold is most important in QTL analysis because the numbers of detected QTLs would be different when using different thresholds in QTL analysis; however, to detect QTLs is the final objective of genetic analysis. In previous reports, low thresholds ($P < 0.05$ in ANOVA) were used to avoid false-negatives, whereas relatively high threshold levels (LOD > 2.4 or > 3.0) were employed to avoid false-positives (Yano and Sasaki 1997). The empirical statistical thresholds employed in QTL analyses should be adjusted, therefore, when using segregating populations with different sizes or different genetic structures. The low threshold used here was chosen in the light of the relatively small size of the BIL population, because it is difficult to detect QTLs using a high threshold in smaller populations. Most of the thresholds employed in published QTL analyses of rice have been between LOD 2.0 and 3.0 in MAPMAKER/QTL, and between 0.05 and 0.01 in ANOVA (Yano and Sasaki 1997). Because our goal in this study was to identify all possible putative QTLs, we used the low threshold, and confirmation of these putative QTLs using advanced backcross progeny will still be necessary.

The control of seed dormancy is very important for controlling pre-harvest sprouting in rice breeding. Kasalath exhibits a desirable seed dormancy, stronger than that of Nipponbare and sufficient to prevent pre-harvest sprouting. The seed dormancy of Kasalath can be broken by drying the panicle for 24 h at room temperature, but seeds so treated never germinate in the field (unpublished data). Thus, alleles of Kasalath at the QTLs detected on chromosomes 3, 5 and 7 will be useful gene sources for the improvement of pre-harvest sprouting resistance in rice, as well as for genetic and physiological studies of rice seed dormancy.

Yasue and Asai (1968) noted that pre-harvest sprouting is associated with heading date. Ikehashi (1973) reported that the temperature during the grain-filling period affects seed dormancy. To investigate the relationship between seed dormancy and heading date, we compared the detected QTLs for seed dormancy with those of heading date in the BILs. We found that the major QTL of seed dormancy on chromosome 3 (C1488) coincided with one of the QTLs for heading date. However, two major heading-date QTLs were located on different chromosomes (6 and 7) from that of the major seed-dormancy QTLs. This suggests that natural environmental factors, such as temperature or weather changes during grain filling, did not affect the phenotypes of seed dormancy in this population. Therefore, it is difficult to explain the significant negative correlation between the two QTLs on chromosome 3 for seed dormancy and heading date by gene

pleiotropy, though this correlation might be due to the tight linkage of these QTLs with each other.

The phenotype of an individual is conditioned not only by its genotype, but by the interaction of that genotype with the environment (Tanksley 1993). Yano et al. (1997) reported on the QTLs for heading date using an F_2 population that was also derived from a cross between Nipponbare and Kasalath grown at Niigata, Japan (latitude 378° N). It is interesting to compare their results with those of our study. We found some new QTLs for heading date in the BILs while other QTLs were not detected. The locations of two major QTLs on chromosomes 6 and 7 detected in the present study were also detected in the F_2 population of Yano et al. (1997). Significant digenic interaction between the two major QTLs was also observed in both their F_2 population and our BILs. However, QTLs linked to C226A on chromosome 6, to L538T3 on chormosome 7, and to R902 on chromosome 8, that were detected in the analysis of the F_2 population of Yano et al. (1997), were not detected in our study. The new putative QTLs for heading date linked to C560 on chromosome 2, to C1488 on chromosome 3, and to R2373 on chromosome 4 that were detected in the present study, were not detected in the F_2 population studied by Yano et al. (1997).

The differences between the results of the two studies might be due to genotype-environment interactions, to differences in the genetic structure of the populations studied or the thresholds employed in the two studies. The comparison suggests that segregation distortion might affect QTL detection. Thus, the lack, in the F_2 population, of the heading-date QTL on chromosome 3 detected in our study, might be caused by segregation distortion on chromosome 3 in the F_2 population. The QTL of heading date was linked to C1488 on chromosome 3, close to a factor (*ga-2*) that produces segregation distortion (Nakagahra 1972; Harushima et al. 1996). In the region near *ga-2*, the frequency of *indica* alleles was greater than the Mendelian ratio when the segregating population was derived from a specific *japonica*/*indica* cross. The number of homozygous Nipponbare individuals in the region of *ga-2* was only 3.2% (the Mendelian ratio is 25%) of the 186 F_2 individuals (Harushima et al. 1996). Therefore, it is difficult to reliably compare the phenotypes between the two genotype groups based on RFLP markers in this region. This segregation distortion was also detected in our BIL population, where the Mendelian ratio of the Nipponbare type is 75% and that of the Kasalath type is 25%. The segregation ratio of the Kasalath type of marker near *ga-2*, observed in the BIL population, was more than 25% and resulted in a segregation ratio near 1 : 1 (the segregation ratio of the nearest marker, C1488, was Nipponbare : Kasalath = 55 : 34; the proportion of the Kasalath type was in excess of the Mendelian ratio). The ability to detect QTLs in this BIL was therefore increased because treating the sample data with a ratio near 1 : 1 is statistically favorable.

Acknowledgements We thank all the members of Rice Genome Research Program for their contributions to this research which was supported by funds from the Ministry of Agriculture, Forestry and Fisheries of Japan (MAFF), and from the Japan Racing Association (JRA). Seed materials of all inbred lines (BC_1F_6) are available at the National Institute of Agrobiological Resources (Please contact Dr. Masahiro Yano, e-mail: myano@abr.affrc.go.jp; fax: + 81-298-38-7468). Genotype data used in this study are available on the internet: http: //www.dna.affrc.go.jp: 84/

References

Anderson JA, Sorrells ME, Tanksley SD (1993) RFLP analysis of genomic regions associated with resistance to pre-harvest sprouting in wheat. Crop Sci. 33 : 453–459

Causse MA, Fulton TM, Cho GY, Ahn SN, Chunwonges J, Wu K, Xiao J, Yu Z, Ronald PC, Harrington SE, Second G, McCouch SR, Tanksley SD (1994) Saturated molecular map of the rice genome based on an interspecific backcross population. Genetics 135 : 1251–1274

Champoux M, Wang G, Sarkarung S, Huang N, Mackill DJ, O'Toole JC, McCouch SR (1995) Locating genes associated with root morphology on the molecular linkage map of rice. Theor Appl Genet 90 : 969–981

Das T (1995) Genetic relation between dormancy and aroma in rice. Rice Genet Newslett 12 : 209–211

Flintham JE, Gale MD (1995) Dormancy gene maps in homologous cereal genomes. In: Noda K, Mares DJ (eds) Pre-harvest sprouting in cereals. Center for Academic Societies Japan, Osaka (CASJO), p 143

Haldane JBS, Waddington CH (1931) Inbreeding and linkage. Genetics 16 : 357–374

Harushima Y, Kurata N, Yano M, Nagamura Y, Sasaki T, Minobe Y, Nakaghara M (1996) Detection of segregation distortions in an indica-japonica rice cross using a high-resolution molecular map. Theor Appl Genet 92 : 145–150

Ikehashi H (1973) Studies on environmental and varietal differences of germination habits of the rice seed, from the special view point of plant breeding (in Japanese). Bull Nat Agri Exp Sta 19 : 1–60

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

Kurata N, Nagamura Y, Yamamoto K, Harushima Y, Sue N, Wu J, Antonio BA, Shomura A, Shimizu T, Lin S-Y, Inoue T, Fukuda A, Shimano T, Kuboki Y, Toyama T, Miyamoto Y, Kirihara T, Hayasaka K, Miyao A, Monna L, Zhong HS, Tamura Y, Wang Z-X, Momma T, Umehara Y, Yano M, Sasaki T, Minobe Y (1994) A 300-kilobase interval genetic map of rice including 883 expressed sequences. Nature Genet 8 : 365–372

Lander ES, Botstein D (1989) Mapping Mendelian factors underlying quantitative traits using RFLP linkage maps. Genetics 121 : 185–199

Lander ES, Green P, Abrahamson J, Barlow A, Daly MJ, Lincoln SE, Newburg L (1987) MAPMAKER: an interactive computer package for constructing primary genetic linkage maps of experimental and natural populations. Genomics 1 : 174–181

Larson S, Bryan G, Dyer W, Blake T (1996) Evaluating gene effects of a major barley seed dormancy QTL in reciprocal backcross populations. J Quant Trait Loci: vol. 2, article 4. On the internet: http: //probe.nalusda.gov: 8000/otherdocs/jqtl

Laura O, Blake TK, Dyer WE, Ullrich SE (1995) Genetic analysis of seed dormancy in barley (*Hordeum vulgare* L.). J Quant Trait Loci, vol. 1, article 5. On the internet: http: //probe.nalusda.gov: 8000/otherdocs/jqtl

Lincoln S, Daly M, Lander E (1993) Mapping genes controlling quantitative traits with MAPMAKER/QTL 1.1: a tutorial and reference manual, 2nd edn. Whitehead Institute Technical Report

Li Z, Pinson SRM, Stansel JW, Park WD (1995) Identification of quantitative trait loci (QTLs) for heading date and plant height in cultivated rice (*Oryza sativa* L.). Theor Appl Genet 91 : 374–381

McCouch SR, Kochert G, Yu ZH, Wang ZY, Khush GS, Coffman WR, Tanksley SD (1988) Molecular mapping of rice chromosomes. Theor Appl Genet 76 : 815–829

Murray MG, Thompson WF (1980) Rapid isolation of high-molecular-weight plant DNA. Nucleic Acids Res 8 : 4321–4325

Nakagahra M (1972) Genetic mechanism on the distorted segregation of marker genes belonging to the eleventh linkage group in cultivated rice. Jpn J Breed 22 : 232–238

Redona ED, Mackill DJ (1996) Mapping quantitative trait loci for seedling vigor in rice using RFLPs. Theor Appl Genet 92 : 395–402

Saito A, Yano M, Kishimoto N, Nakagahra M, Yoshimura A, Saito K, Kuhara S, Ukai Y, Kawase M, Nagamine T, Yoshimura S, Ideta O, Ohsawa R, Hayano Y, Iwata N, Sugiura M (1991) Linkage map of restriction fragment length polymorphism loci in rice. Jpn J Breed 41 : 665–670

SAS Institute (1989) SAS/STAT user's guide: version 6, vol 2, 4th edn. SAS Institute Inc., Cary, North Carolina

Seshu DV, Sorrells ME (1986) Genetic studies on seed dormancy in rice. In: Rice genetics. IRRI, Philippines, pp 369–382

Takahashi N (1995) Physiology of dormancy. In: Matsuo T, Kumazawa K, Ishii R, Ishihara K, Hirata H (eds) Science of the rice plant. 2. Physiology. Food and Agriculture Policy Research Center, Tokyo, pp 45–57

Takahashi (1997) Inheritance of seed germination and dormancy. In: Matsuo T, Futsuhara Y, Kikuchi F, Yamaguchi H (eds) Science of the rice plant. 3. Genetics. Food and Agriculture Policy Research Center, Tokyo, pp 348–359

Tanksley SD (1993) Mapping polygenes. Annu Rev Genet 27 : 205–233

Wang GL, Mckill DJ, Bonman JM, McCouch SR, Champoux MC, Nelson RJ (1994) RFLP mapping of genes conferring complete and partial resistance to blast in a durably resistant rice cultivar. Genetics 136 : 1421–1434

Xiao J, Li J, Yuan L, Tanksley SD (1996) Identification of QTLs affecting traits of agronomic importance in a recombinant inbred population derived from a subspecific rice cross. Theor Appl Genet 92 : 230–244

Yano M, Sasaki T (1997) Genetic and molecular dissection of quantitative traits in rice. Plant Mol Biol 35 : 145–153

Yano M, Harushima Y, Nagamura Y, Kurata N, Minobe Y, Sasaki T (1997) Identification of quantitative trait loci controlling heading date of rice using a high-density linkage map. Theor Appl Genet 95 : 1025–1032

Yasue T, Asai Y (1968) Studies on the dormancy and viviparous germination in rice seed. Res Bull Fac Agri Gifu Univ 26 : 1–12

Young ND, Tanksley SD (1989) Restriction fragment length polymorphism maps and the concept of graphical genotype. Theor Appl Genet 77 : 95–101