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Flowering response of rice to photoperiod and temperature: a QTL analysis using a phenological model

Abstract In this study we have attempted to quantify the thermal and photoperiodical responses of rice (*Oryza sativa* L.) flowering time QTLs jointly by a 'date-of-planting' field experiment of a mapping population, and a 'phenological model' analysis that separately parameterizes the two responses, based on daily temperature, daily photoperiod and flowering date. For this purpose, the 'three-stage Beta model', which parameterizes the sensitivity to temperature (parameter α), the sensitivity to photoperiod (parameter β), and earliness under optimal conditions (10 h photoperiod at 30°C) (parameter G), was applied to 'Nipponbare' × 'Kasalath' backcross inbred lines that were transplanted on five dates. QTLs for the β value were detected in the four known flowering time QTL (*Hd1*, *Hd2*, *Hd6* and *Hd8*) regions, while QTLs for the G value were detected only in the *Hd1* and *Hd2* regions. This result was consistent with previous reports on near-isogenic lines (NILs) of *Hd1*, *Hd2* and *Hd6*, where these loci were involved in photoperiod sensitivity, and where *Hd1* and *Hd2* conferred altered flowering under both 10 and 14 h photoperiods, while *Hd6* action was only affected by the 14 h photoperiod. *Hd8* was shown to control photoperiod sensitivity for the first time. Interestingly, *Hd1* and *Hd2* were associated with a QTL for the α value, which might support the previous hypothesis that the process of photo-induction depends on temperature. These results demonstrate that our approach can effectively quantify environmental responses of flowering time QTLs without controlled environments or NILs.

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Introduction

Understanding the flowering response of crops to their natural field environment is essential for manipulating productivity through management and breeding. In the last decade, this issue has been greatly facilitated by quantitative trait locus (QTL) analyses for flowering time. Rice (*Oryza sativa* L.) is a crop in which such studies have been pioneered. To date, flowering (heading) time QTLs have been identified in more than 20 regions of the rice genome (Li et al. 1995; Tsunematsu et al. 1996; Xiao et al. 1996; Yano et al. 2001). For many of these QTLs, near-isogenic lines (NILs) have been developed via the marker-aided selection technique. Using these NILs, the photoperiodical response of QTLs have been characterized under controlled environmental regimes (Yano et al. 2001). However, this type of work is laborious and time-consuming. Thus, a comprehensive approach that analyzes the responses of individual QTLs to the natural field environment in situ is necessary for understanding the flowering response to complex natural field environments.

Because most crops respond to environmental factors in a quantitative manner, these responses have been analyzed most successfully with mathematical models that simulate their phenology, or 'phenological models'. For rice, the 'developmental rate' (DVR)-type phenological model (de Wit et al. 1970; Horie 1994) has proved particularly useful in simulating reproductive initiation and flowering (Horie and Nakagawa 1990; Gao et al. 1992; Summerfield et al. 1992; Yin et al. 1997b). In the

DVR model, the progress of developmental stages from seedling emergence (defined as stage 0) to flowering (stage 1) is quantified by integrating the daily increment of developmental stages, or the DVR (by definition, flowering occurs when the sum of daily DVR = 1). The DVR is principally a function of temperature [$f_1(T)$, holding the units of DVR (day^{-1})], while expression of $f_1(T)$ is modified by the function $f_2(P)$, a scalar function of photoperiod that ranges from 0 to 1. Thus, DVR is set as: DVR = $f_1(T) f_2(P)$ Different functions could be used for $f_1(T)$ and $f_2(P)$. Yin et al. (1997a, b) showed that the Beta function (Johnson and Leone 1964) is particularly useful for both $f_1(T)$ and $f_2(P)$ in rice. Specific parameter values that characterize $f_1(T)$ and $f_2(P)$ are determined based on experiments, the most popular of which is the classical 'date-of-planting' experiment, where the cultivar of interest is planted at different dates under natural field conditions (Horie and Nakagawa 1990). These models provide a practical guide for determining cultivars and management practices. In addition, the model parameters could serve as effective means for quantifying genetic differences in phenological responses to environmental factors. For example, Yin et al. (1997b) successfully quantified the flowering responses of nine rice cultivars to temperature and photoperiod using DVR model parameters based on field data. Recently, several reports have successfully integrated QTL and such ecophysiological-model analyses for understanding and predicting agronomic traits such as yield components (Yin et al. 2000) and leaf growth (Reymond et al. 2003). Identification of QTLs that control these phenological model parameters would be very helpful in genetically dissecting plant responses to environmental factors.

We quantified the thermal and photoperiodical responses of individual flowering time QTLs in rice jointly by: (1) a date-of-planting experiment using a mapping a population grown under natural field conditions, and (2) a DVR model analysis that parameterized these two responses. For this purpose, we used 'Nipponbare' × 'Kasalath' backcross inbred lines (BILs) (Lin et al. 1998). This cross is considered the best choice for validating our approach because many flowering time QTLs (*Hd1* to *Hd14*) have been well characterized using NILs (Yano et al. 2001). The model-parameter values for each BIL were estimated from the five planting experiments, and then subjected to QTL analyses to identify genetic factors for thermal and photoperiodical responses. The results were compared with the physiological information from the NILs obtained from previous studies.

Materials and methods

Mapping population

Ninety-eight BILs (BC$_1$F$_9$) derived from the cross between Nipponbare and Kasalath were used. Nipponbare is a photoperiod-sensitive *japonica* variety developed in Japan, while Kasalath is a photoperiod-insensitive *indica* landrace from India. The 98 lines were developed from BC$_1$F$_1$ (Nipponbare/Kasalath//Nipponbare) plants using the single-seed descent method (Lin et al. 1998).

Phenotypic evaluation

The experiment was conducted in a paddy field at the experimental farm of The University of Tokyo, Nishitokyo, in 2003. Five 21-day-old seedlings, grown in a daylight phytotron with a day/night temperature of 30°C/25°C, were transplanted at one plant per hill on 28 April, 12 May, 26 May, 9 June and 23 June with a 30 cm row spacing, a 15 cm hill spacing, and no replication. A chemical compound fertilizer (60-40-66 kg of N-P-K ha^{-1}) was applied basally just before transplanting. Days to flowering (the number of days from seedling emergence to opening of the first flower) were recorded for five plants per entry. In early May 2003 (i.e., immediately after the first date of transplanting), Tokyo experienced strong windstorms, which caused severe damage to plant rooting. As a result, only one or two plants survived until flowering in 12 entries that included the *indica* parent Kasalath. For these entries, only the data from the plants that survived were recorded.

Model construction and parameter estimation

We used a modified 'three-stage Beta model' (Yin et al. 1997b), a DVR model for rice flowering that assumes that the pre-flowering development of a rice plant is divided into three subphases: (1) the 'juvenile phase', when it is not yet sensitive to the flowering stimulus; (2) the 'photoperiod sensitive phase' (PSP), when the plant is able to show responses to photoperiodic flowering stimulus; and (3) 'post-PSP' (after completion of the PSP to flowering). The model quantifies the progress of developmental stages (DVS) from seedling emergence (DVS 0) to flowering (DVS 1) by integrating the daily DVR (day^{-1}) as:

$$DVS = \sum_{i=0}^{n} DVR_i$$

where i is the number of days after seedling emergence. The DVR is assumed to be a multiplicative function of responses to temperature and photoperiod as:

$$DVR = \begin{cases} f(T)/G & (DVS < DVS_1 \text{ or } DVS > DVS_2) \\ f(T)g(P)/G & (DVS_1 \leqslant DVS \leqslant DVS_2) \end{cases}$$

where T is the daily mean temperature (°C) and P is the photoperiod (in h, calculated as the duration of possible sunshine). The parameter G characterizes the earliness of flowering (scaled by the number of days from seedling emergence to flowering) under optimal photoperiod and temperature. DVS_1 and DVS_2 are the DVS values at the end of the juvenile and photoperiod-sensitive phases, respectively. The response to temperature is described by the following Beta function:

$$f(T) = \begin{cases} \left[\left(\frac{T-T_b}{T_0-T_b} \right) \left(\frac{T_c-T}{T_c-T_0} \right)^{(T_c-T_0)/(T_0-T_b)} \right]^{\alpha}, & (T_b \leqslant T \leqslant T_c) \\ 0, & (T < T_b, T > T_c) \end{cases}$$

where T_b and T_c are the base and ceiling temperatures, respectively (DVR $= 0$ at both temperatures), and T_o is the optimum temperature. Parameter α ($\alpha > 0$) is the sensitivity coefficient that characterizes the response to temperature (the temperature-sensitivity increases with an increase in the α value) (Fig. 1). The response to photoperiod is described as:

$$g(P) = \begin{cases} \left[\left(\frac{P-P_b}{P_0-P_b} \right) \left(\frac{P_c-P}{P_c-P_0} \right)^{(P_c-P_0)/(P_0-P_b)} \right]^{\beta}, & (P \geqslant P_0) \\ 1, & (P < P_0) \end{cases}$$

where P_b, P_o and P_c are the base, optimum and ceiling photoperiods, respectively. Parameter β ($\beta > 0$) is the sensitivity coefficient that characterizes the response to photoperiod (the photoperiod sensitivity increases with an increase in the β value) (Fig. 1). Among the nine parameters, T_b, T_c, P_b, P_o and P_c are fixed at 8°C, 42°C, 0 h, 10 h and 24 h, respectively. These fixed values can sufficiently describe a wide range of flowering responses of rice cultivars (Yin et al. 1997b). Although optimum temperature, T_o, is important in describing cultivar differences in rice phenology under tropical environments (Yin et al. 1997b), it is mostly out of range for temperatures in temperate regions, such as in the current study. Thus, T_o is fixed at 30°C, as reported for Nip-

ponbare by Horie and Nakagawa (1990). To minimize the numbers of parameters, we employed the empirical relationship between parameters G, DVS_1 and DVS_2 reported for 17 rice cultivars by Yin et al. (1997b):

$$DVS_1 = 0.145 + 0.005G$$
$$DVS_2 = 0.345 + 0.005G$$

The values of parameters α, β and G were selected to describe photothermal responses of individual BILs, as a set that best fitted observed days to flowering for each entry. The SIMPLEX method was used for the iterative calculation to optimize the three parameters.

Map construction and QTL analysis

Genotypic data of 245 RFLP markers (http://www.rgrc.dna.affrc.go.jp/index.html.en) were used for the QTL analysis of flowering time and the model parameters (α, G and β). Linkage analysis was performed using Mapmaker/EXP 3.0 (Lander et al. 1987) with the Kosambi function (Kosambi 1944). QTL analysis was performed using composite interval mapping with Windows QTL Cartographer v2.0 (Wang et al. 2001–2003). A QTL was declared when the LOD value exceeded the threshold of 3.0. In addition, we considered a peak with a LOD value between 2.5 and 3.0 as a putative QTL, because this was also informative for estimating flowering time values from model-parameter QTLs (Yin et al. 2000). The total percentage of variance explained by the detected QTLs was estimated by multiple regression analysis using the nearest flanking markers of each QTL.

The overall explanatory value of the detected model-parameter QTLs was further assessed by QTL-based DVR model prediction, whereby the original parameter (α, G and β) values are replaced by those estimated from identified QTLs for the parameters. Briefly: (1) the parameter values were estimated by QTL genotypes for each BIL and the parents as described by Yin et al. (2000), (2) the values were input into the DVR model, and (3) the performance was evaluated by comparing the predictability (R^2) of outputs (i.e., predicted days to flowering) of the QTL-based model and the original model. The analysis was made on two separate field experiments: the present date-of-planting experiment, whose data were used for model parameterization (see above), and an independent field experiment using the same BILs conducted in Tsukuba (Lin et al. 1998). In both cases, only BILs (66 lines) homozygous for all parameter QTL regions were used for the QTL-based model prediction.
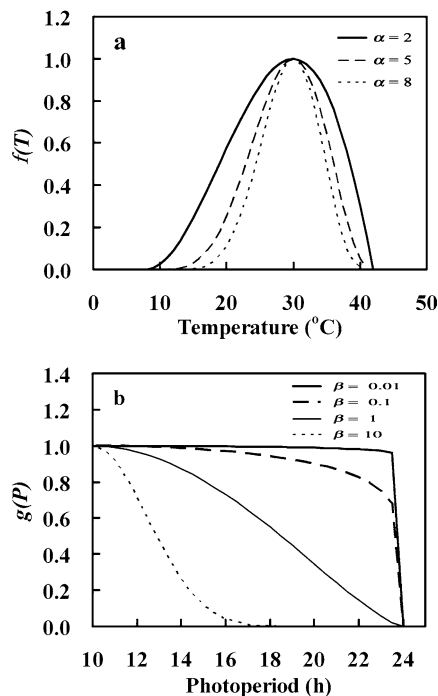

Fig. 1 Plots of two components of the DVR model used in this study as a function of temperature (T) and photoperiod (P). **a** The function $f(T)$ with different values of parameter α. **b** The function $g(P)$ with different values of parameter β

Results

Phenotypic evaluation and QTL mapping of flowering time

In Nipponbare, the flowering date was almost constant across the five planting dates (21, 20, 23, 25 and 30
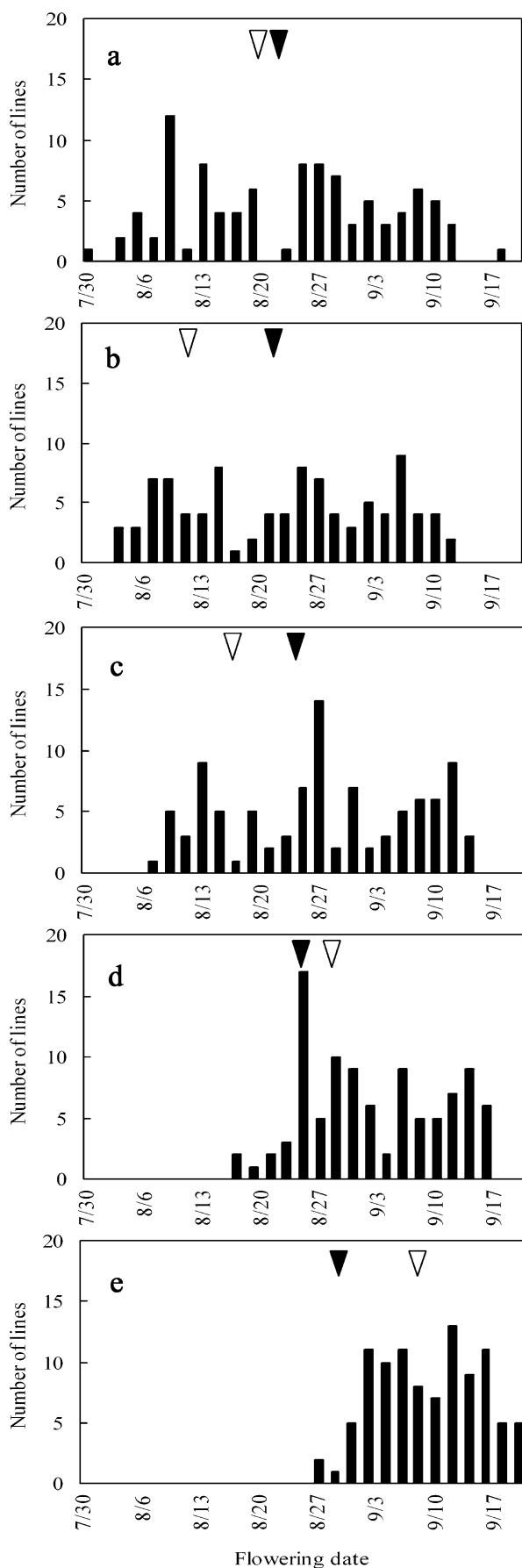
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Fig. 2 Distribution of the flowering dates of 98 BILs. Seedlings were transplanted on 28 April, the first date of planting (**a**), 12 May, the second date of planting (**b**), 26 May, the third date of planting (**c**), 9 June, the fourth date of planting (**d**), and 23 June, the fifth date of planting (**e**). *Open and closed triangles* indicate the flowering dates of Kasalath and Nipponbare, respectively

August for the first to fifth dates of planting, respectively). By contrast, the planting date greatly affected the flowering date of Kasalath (18, 10, 17 and 27 August, and 7 September for the first to fifth date of planting, respectively). The BIL values ranged from 30 July to 20 September, 3 August to 12 September, 8 August to 14 September, 15 August to 17 September and 27 August to 20 September for the first to fifth dates of planting, respectively (Fig. 2). In many lines, including Kasalath, flowering for the first date of planting was delayed to the second date of planting, possibly due to the severe damage caused by the windstorm in early May.

The analysis with QTL Cartographer 2.0 revealed eight QTLs for days to flowering on chromosomes 1 (two QTLs), 3 (two QTLs), 4, 6, 7 and 8 (Table 1). At the QTLs on chromosomes 3 and 8, the Kasalath allele prolonged flowering, whereas the Nipponbare allele prolonged flowering at the QTLs on chromosomes 1, 4, 6 and 7. Based on their map position and the direction of the additive effects, the QTLs on chromosomes 3 (two QTLs), 4, 6, 7 and 8 were considered to be the known flowering time QTLs *Hd8*, *Hd6*, *Hd11*, *Hd1*, *Hd2*, and *Hd5* (Yano et al. 2001), respectively (Fig. 3). The combined percentage of variation explained by all identified QTLs was 62, 71, 66, 54 and 55% for the first to fifth dates of planting, respectively.

Estimation and QTL mapping of model parameters

Based on daily temperature, photoperiod and flowering date, we determined the best parameter (α, G and β) values for each entry so that the DVR model would best fit their observed flowering dates. The resultant model successfully described the flowering date of the BILs with a high accuracy ($R^2 = 0.990$, 0.988, 0.993, 0.991 and 0.985 for the first to fifth dates of planting, respectively). Moreover, the model predicted well the flowering date in independent field experiments in Tsukuba ($R^2 = 0.81$; Fig. 4c). This validated our parameterization. The three parameters clearly differentiated the flowering responses of the two parents (Fig. 5). Parameter α was 6.9 for Nipponbare and 4.4 for Kasalath, which suggested that flowering response to temperature was more sensitive in Nipponbare than in Kasalath. Parameter G was greater in Kasalath ($G = 65$) than in Nipponbare ($G = 46$), which indicated that the growth period (days to flowering) under the optimal (10 h photoperiod at 30°C) condition was much longer in Kasalath than in Nipponbare. Parameter β was 4.6 for Nipponbare and 0.1 for Kasalath, which was consistent with the fact that Nipponbare is

Table 1 Location, peak LOD, additive effects, and percent of the phenotypic variation explained (R^2) for QTLs detected for days to flowering in BILs from the cross Nipponbare × Kasalath

Transplanted date	Chromosome	Marker interval	Peak LOD	Additive effect[a]	R^2
28 April	3	C63-C1488	3.17	−3.21	0.06
	6	R2171-R2123	14.94	9.05	0.33
	7	C213-C728	13.09	7.50	0.30
12 May	3	C63-C1488	4.33	−3.66	0.08
	4	C946-R288	2.60[b]	2.68	0.05
	6	R2171-R2123	16.20	9.79	0.40
	7	C213-C728	13.50	7.55	0.31
	8	R902-C1121	2.97[b]	− 3.42	0.05
26 May	1	C955-C1211	2.56[b]	3.37	0.05
	3	C63-C1488	4.02	−3.63	0.10
	3	C944-C595	2.81[b]	− 3.64	0.06
	6	R2171-R2123	16.25	9.45	0.42
	7	C213-C728	9.92	6.17	0.24
16 June	1	R1928-R2635	2.64[b]	2.14	0.05
	3	C63-C1488	6.53	−3.42	0.15
	3	C944-C595	3.08	−3.01	0.08
	6	R2171-R2123	12.88	6.97	0.37
	7	C213-C728	6.68	3.54	0.15
23 June	3	C63-C1488	4.82	−2.35	0.13
	3	C944-C595	3.00	−2.33	0.09
	6	R2171-R2123	7.97	3.80	0.22
	7	C213-C728	4.92	2.46	0.13
	8	R902-C1121	3.77	−2.53	0.10

[a]Positive value indicates an additive effect of the Nipponbare allele
[b]Putative QTLs with a LOD of 2.5–3.0

photoperiod-sensitive while Kasalath is almost completely photoperiod-insensitive. The BIL values showed a transgressive segregation for all the parameters.

We then detected QTLs for these three parameters (Table 2, Fig. 3). For parameter α, four QTLs were detected at the *Hd1*, *Hd2*, *Hd9* and R117-C112 (chromosome 1) regions. At these QTLs, the Nipponbare alleles increased the α value (i.e., conferred higher sensitivity to temperature). An exception was for the R117-C112 region, where the Kasalath allele increased the α value. For parameter G, two QTLs were detected at the *Hd1* and *Hd2* regions. The Kasalath alleles increased the G value (i.e., conferred late flowering under the optimal condition) at both QTLs. For parameter β, five QTLs, located at the *Hd1*, *Hd2*, *Hd6*, *Hd8* and C178-R1928 (chromosome 1) regions, were detected. The Kasalath alleles increased the β value at the *Hd6* and *Hd8* regions (i.e., conferred higher sensitivity to photoperiod), while the Nipponbare alleles increased the value at the remaining QTLs. No apparent LOD peak of the three parameters was detected at the *Hd5* region on chromosome 8. The combined percentage of variation explained by all identified QTLs was 45, 55 and 65% for α, G and β, respectively.

The overall explanatory value of these QTLs was further assessed using the QTL-based model prediction approach (Yin et al. 2000). The R^2 value between the measured days to flowering and those predicted based on the model-parameter QTLs was lower ($R^2 = 0.818$; Fig. 4b) than that of the original model prediction ($R^2 = 0.994$; Fig. 4a). A similar tendency was observed in the Tsukuba experiment ($R^2 = 0.401$ and 0.810 for QTL-based and original model predictions, respectively; Fig. 4d, c).

Discussion

The flowering time QTLs of the Nipponbare × Kasalath cross have been extensively studied. From this cross (F_2, BILs, and advanced backcross progenies such as BC_3F_2), 15 flowering time QTLs (*Hd1* to *Hd14*, including *Hd3a* and *Hd3b*) have been identified (see Yano et al. 2001 for a review). NILs have been developed for more than half of these QTLs, and have been used to fully characterize the responses to photoperiod (10 and 14 h). Such information is essential for testing the validity of our trial using a phenological model approach. In our field experiment on BILs, we detected seven known flowering time QTLs: *Hd1*, *Hd2*, *Hd5*, *Hd6*, *Hd8*, *Hd9* and *Hd11*. Of the seven QTLs, four (*Hd1*, *Hd2*, *Hd5* and *Hd6*) appeared to respond to photoperiod based on NIL studies (Lin et al. 2000, 2003; Yamamoto et al. 2000). Our results are consistent with these previous reports: we detected QTLs for parameter β (the index for photoperiod-sensitivity) at these photoperiod-sensitive *Hd* regions (except the *Hd5* region, where no significant QTLs were detected for any of the model parameters) (Fig. 3). In our study, a QTL for parameter β was also detected in the *Hd8* region, which suggests that *Hd8* is involved in photoperiod sensitivity. This is the first evidence of the photoperiod-sensitive nature of *Hd8*. Besides the known *Hd* QTLs, we detected two putative flowering time QTLs on chromosome 1. Of the two, the QTL at the R1928 region was associated with a QTL for parameter β, suggesting that this flowering time QTL also controls photoperiod sensitivity.

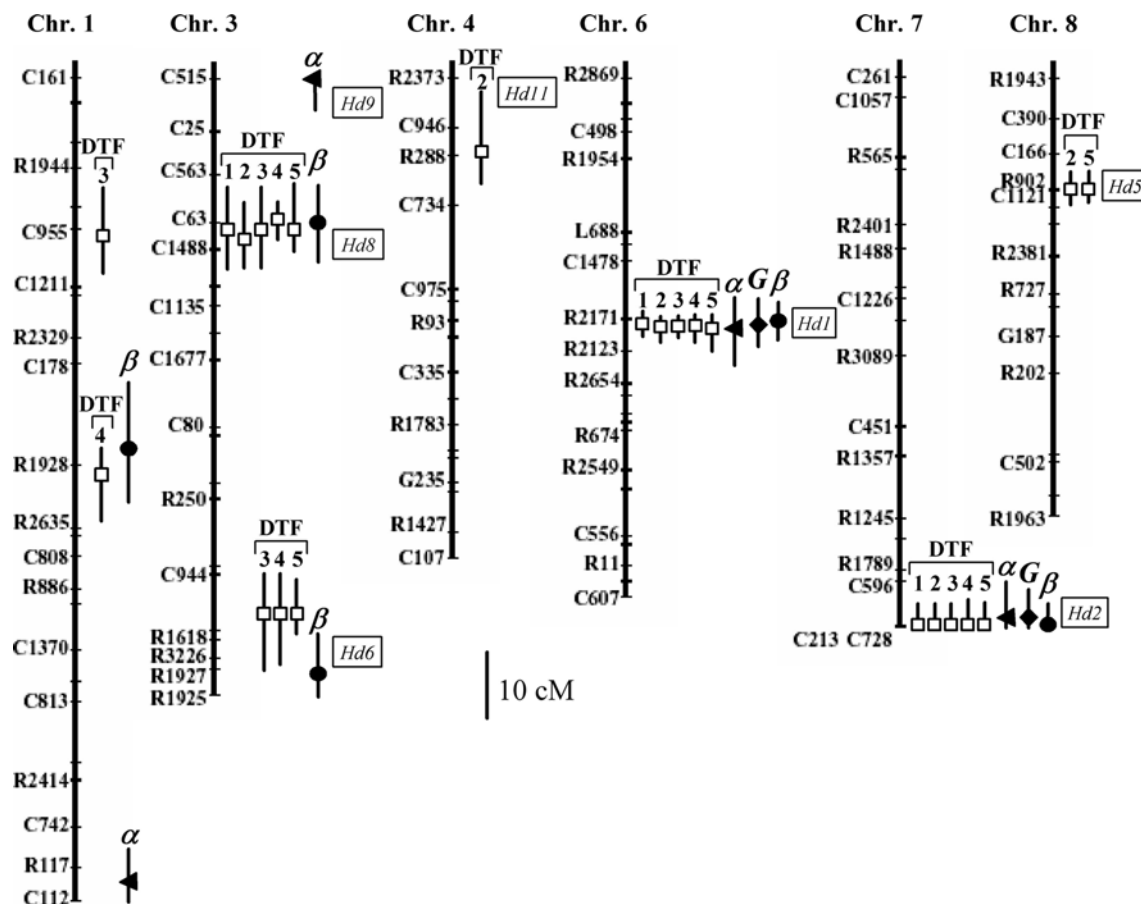Further congruence between the previous and present results was found for parameter G (days to flowering

Fig. 3 Chromosomal location of flowering time and model parameter QTLs. *Bars* to the right of the chromosomes indicate 1-LOD likelihood intervals, and *symbols* indicate the position of the peak LOD in the interval. *DTF* indicates days to flowering for the first (1), second (2), third (3), fourth (4) and fifth (5) dates of planting. α, *G* and β are the model parameters. Some known flowering time QTLs (*Hd 1*, *Hd2*, *Hd5*, *Hd6*, *Hd8*, *Hd9* and *Hd11*) are shown as references

under a 10 h photoperiod at 30°C). Previous studies on NILs showed that there were three different types of photoperiod-sensitive genes. The first responds to long days (14 h) and short days (10 h) in opposite directions (*Hd1* and *Hd2*), the second responds to long days, but not to short days (*Hd3b*, *Hd4*, *Hd5*, *Hd6*, etc.) (Lin et al. 2000, 2002, 2003; Yamamoto et al. 2000; Monna et al. 2002), and the third responds to short days, but not to long days (*Hd3a*) (Monna et al. 2002). In our study, QTLs for parameter *G* were detected at the *Hd1* and *Hd2* regions, but not at the *Hd6* and *Hd8* regions (Fig. 3). This finding was consistent with the fact that *Hd1* and *Hd2* responded to both long- and short-day conditions (i.e., the Kasalath alleles conferred early- and late-flowering under long- and short-day conditions, respectively), while *Hd6* responded to only the long-day conditions (i.e., the Kasalath allele conferred late flowering under long-day conditions). The additive effects of the Kasalath allele at the two *G* value QTLs were 4.61 days (*Hd1* region) and 5.67 days (*Hd2* region) (Table 2). These values were comparable with those in the previous report that the flowering of NILs for *Hd1*, *Hd2* and *Hd1* + *Hd2* were delayed by 9.1, 3.4 and 24.6 days, respectively, under the 10 h photoperiod as

compared with the Nipponbare control (Lin et al. 2000). These additive effects predict well the action of QTLs under the 10 h photoperiod, considering that the photoperiod ranged from 12.2 to 14.5 h during the growing season in our field experiment. Such congruence between the previous and present results suggests that our approach could effectively characterize the photoperiodical responses of individual flowering time QTLs.

In contrast to photoperiod sensitivity, very little research has been undertaken on flowering time QTLs related to temperature responses in rice. In our study, the response to temperature was indexed by parameter α, for which four QTLs were detected. Interestingly, two of these four were associated with the photoperiod-sensitive QTLs, *Hd1* and *Hd2* (Fig. 3). This association might be due to the linkage between an *Hd* and a thermo-sensitive gene. Alternatively, this might be due to pleiotropism, which might be relevant to the 'thermal control of photoinduction' theory (Vergara and Chang 1985; Yin et al. 1997a). In rice, temperature just before floral initiation may strongly affect days to flowering (Vergara and Lilis 1968; Shibata et al. 1973; Yin et al. 1997a). The effect of temperature is stronger in photoperiod-sensitive cultivars than in photoperiod-insensitive cultivars, which suggests

Fig. 4 Comparison between the observed days to flowering and those predicted by the model with original parameters (**a, c**), and between the observed days to flowering and those predicted by the model with QTLs for the model parameters (**b, d**), in 66 BILs and their parents. The *solid line* represents the linear regression of the predicted value (*y*) to the observed value (*x*). **a, b** Field experiment in Tokyo for which the parameterization was made. **c, d** An independent field experiment in Tsukuba (data adopted from Lin et al. 1998)
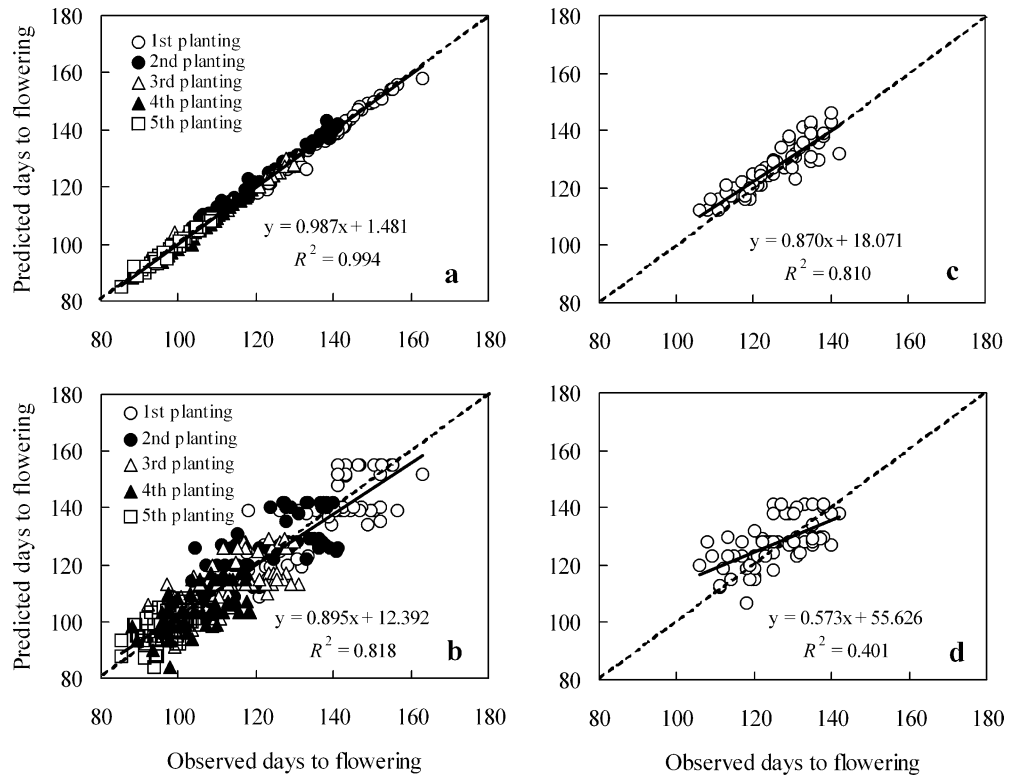
that temperature has an effect on photoinduction as well as its role as a general modifier of developmental rate (Yin et al. 1997a). In fact, different critical photoperiods have been observed under different temperatures in many other plant species (Thomas and Vince-Prue 1997). Our finding that certain photoperiod-sensitive QTLs were associated with thermal-response QTLs might be consistent with the fact that the photoinduction process was highly sensitive to temperature. Recently, *Hd1* was cloned using a map-based strategy and appeared to be the rice homolog of *CONSTANS*, an *Arabidopsis thaliana* gene involved in the signal transduction of floral induction (Yano et al. 2000). This will facilitate investigation of the possible response of *Hd1* gene expression to temperature (including the influence of the allelic difference) in the future.

Not all QTLs for parameter α were associated with those of parameter β. An interesting example is the *Hd9* region on chromosome 3. *Hd9* was first identified using an advanced backcross progeny, and the NIL phenotype (including epistasis with other *Hd* loci) has raised two hypotheses: (1) that *Hd9* controls photoperiod-sensitivity independent of such major genes as *Hd1* and *Hd2*, and (2) that *Hd9* is involved in functions other than photoperiod sensitivity, such as a high temperature requirement (Lin et al. 2002). In our study, a putative QTL for parameter α existed in the *Hd9* region, but not for
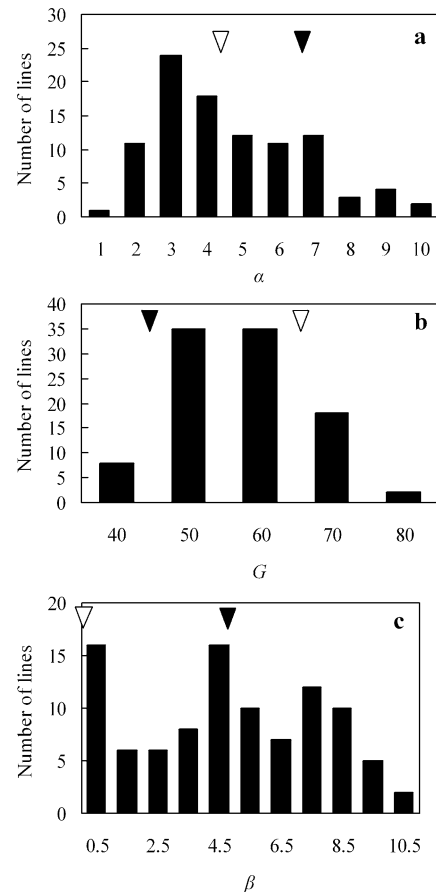
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Fig. 5 Distribution of parameters α (**a**), G (**b**) and β (**c**). *Open and closed triangle* indicate values of Kasalath and Nipponbare, respectively

Table 2 Location, peak LOD, additive effects, and percent of the phenotypic variation explained (R^2) for QTLs detected for phonological model parameters in BILs from the cross Nipponbare × Kasalath

Trait	Chromosome	Marker interval	Peak LOD	Additive effect[a]	R^2
Parameter α	1	R117-C112	3.14	−0.95	0.12
	3	C515-C25	2.99[b]	0.82	0.10
	6	R2171-R2123	3.72	0.95	0.13
	7	C596-C213	3.34	0.82	0.13
Parameter G	6	R2171-R2123	7.06	−4.61	0.17
	7	C596-C213	12.07	−5.67	0.33
Parameter β	1	C178-R1928	3.19	0.99	0.09
	3	C63-C1488	3.17	−0.88	0.07
	3	R1927-R1925	2.53[b]	− 0.94	0.06
	6	R2171-R2123	9.02	1.96	0.23
	7	C213-C728	11.06	1.80	0.29

[a] Positive value indicates additive effect of the Nipponbare allele
[b] Putative QTLs with a LOD of 2.5–3.0

parameter β (Table 2, Fig. 3). This result may support the latter hypothesis that *Hd9* is involved in the thermal response.

Date-of-planting experiments have been widely employed for identifying best cultivar-season combinations in agronomy. Such experiments have also been a rich source of information on the quantitative aspects of phenological responses to environments (Loomis and Connor 1992). Our results demonstrated that a QTL analysis of phenological-model parameters for date-of-planting experiments is an informative approach that could quantify the environmental responses of individual QTLs without the need for controlled environments or NILs. In our study, the explanatory power of the model-parameter QTLs was not very high, as shown by lower R^2 values of QTL-based model predictions compared with the original model prediction (Fig. 4). This may be partly explained by the relatively low resolution power of our QTL detection due to the small population size (98 lines). Further model improvement would also increase the explanatory power of model-parameter QTLs. This could be achieved by: (1) estimating T_o, DVS_1 and DVS_2 values as model parameters (they were set as fixed values or approximated empirically in the present study), and (2) using specific model parameter values for each growth stages (Horie and Nakagawa 1990; Nakagawa and Horie 1997; Yin et al. 1997b). However, such multiple parameterization requires a much larger data set with many planting dates.

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