Identification of QTLs for Plant Height and Its Components by Using Single Segment Substitution Lines in Rice (Oryza sativa)

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Abstract: QTLs for plant height and its components on the substituted segments of fifty-two single segment substitution lines (SSSLs) in rice were identified through t-test (P≤0.001) for comparison between each SSSL and recipient parent Huajingxian 74. On the 14 substituted segments, 24 QTLs were detected, 10 for plant height, 2 for panicle length, 4 for length of the first internode from the top, 5 for length of the second internode from the top and 3 for length of the third internode from the top, respectively. All these QTLs were distributed on nine rice chromosomes except chromosomes 5, 9 and 11. The additive effect ranged from –4.08 to 3.98 cm, and the additive effect percentages varied from –19.35% to 10.43%.

Key words: rice; single segment substitution line; plant height; quantitative trait locus

Plant height is one of the most important traits associated with plant type and yield potential in rice. In the 1960s, the development of semi-dwarf cultivars had made a marked advance in rice productivity. Since then, the genetic basis of plant height was widely studied. Since semi-dwarf gene sd-1 was first characterized in the Chinese variety Dee-geo-woo-gen, more than 60 genes responsible for the dwarf or semi-dwarf growth habit of rice varieties have been identified by using classical genetic analysis [1, 2]. Many dwarf or semi-dwarf genes have been mapped on rice genetic linkage maps [3-5]. Gene sd-1 has been cloned and widely utilized in the breeding of high-yielding varieties [6].

There have been many studies attempting to map QTLs for plant height in rice using molecular marker genetic analysis [7-10]. Partitioning plant height into panicle length and length of upper internodes has significant implications for deeply understanding the genetic basis of plant height in rice [11-13]. Lin et al [11] detected 7 QTLs for plant height, 3 QTLs for panicle length and 17 QTLs related to upper internodes in a F2 population constructed from Tesanai 2/CB. Tan et al [12] identified 4 QTLs for plant height and 12 QTLs controlling the length of upper internodes using a DH population from a cross between Zhaiyeqing 8 and Jingxi 17. Yamamoto et al [13] mapped 6 QTLs for culm length, 4 QTLs for panicle length and 17 QTLs for the length of upper internodes using a BC1F3 population derived from the Koshihikari/Kasalath/Koshihikari cross.

The populations usually used for QTL mapping in self-pollinated crops were F2/F3, BC1, DH or RIL [14]. To facilitate genetic mapping of QTLs, some secondary mapping populations were developed. Eshed and Zamir [15] constructed introgression lines (ILs) and used them to map QTLs for agronomically important traits in tomato. Yu et al [16] studied the heterosis between japonica and indica rice using a set of chromosome segment substitution lines (CSSLs) covering the whole indica genome with the japonica genetic background. Their results clearly demonstrated the validity of using ILs or CSSLs in the genetic analysis of quantitative traits. We have constructed a series of single segment substitution lines (SSSLs), each containing only one marker-defined chromosomal segment introgressed from one donor into a cultivar’s genetic background [17, 18]. Through trait comparison between SSSLs and the recipient parent, QTLs on the substituted segments for important traits were identified and their genetic effects were estimated [19, 20]. In the present study, we identified QTLs for plant height and its components on the substituted segments of 52 SSSLs in rice.
MATERIALS AND METHODS

Plant materials and traits measurements

Fifty-two SSSLs were used in the experiment. These SSSLs were developed via microsatellite marker-assisted selection, from six advanced backcrosses involving recipient Huajingxian 74 and six donors. The average length of the substituted segments in fifty-two SSSLs was 25.0 cM. The total covered length on rice chromosome was 766.1 cM, accounting for 50.4% coverage of rice genome [18].

The experiment was performed at the farm of South China Agricultural University, Guangzhou, China, during second season of 2002. Each SSSL, together with six donors, Suyunuo, IR64, IRAT261, Chenglongshuijingmi, Lemont and IAPAR9, was planted at a single plot in the same field. Three plots of Huajingxian 74 were also randomly planted in the field. The individual plot consisted of four rows with ten plants per row.

The twelve plants in the middle of the two central rows were measured at maturity for plant height (PH), panicle length (PL), length of the first internode from the top (LFI), length of the second internode from the top (LSI) and length of the third internode from the top (LTI).

Analysis of environmental influence

Huajingxian 74 was planted in three replications randomly. The environmental influence on the traits tested was evaluated via F-test for comparison in three Huajingxian 74 plots.

QTL analysis

QTL analysis was conducted as suggested by Liu et al [19]. The QTLs for plant height and its components were detected by t-test for comparisons between each SSSL and the recipient parent Huajingxian 74 with a significance of $P \leq 0.001$. QTLs were nominated as described by McCouch et al [21].

The additive effect and the additive effect percentage of the QTLs were estimated. The additive effect was half of the difference between each SSSL and Huajingxian 74.

QTL substitution mapping approach was employed to detect the exact QTL position as described in previous article [20]. If one QTL could be detected in both SSSLs containing overlapping substituted segment, the QTL was localized to the overlapping interval; If one QTL was detected in one SSSL, but not detected in another SSSL with overlapping segment, the QTL was mapped at the non-overlapping interval of the overlapping segments.

RESULTS

Phenotypic variation among parents

F-tests showed that the differences for PH, PL, LFI, LSI and LTI in three Huajingxian 74 plots were not significant ($P > 0.05$), indicating that the environment had no significant influence on the traits tested. Hence, the phenotypic values in three Huajingxian 74 plots were mixed as a control in t-test.

The differences for plant height, panicle length, length of the first internode from the top, length of the second internode from the top and length of the third internode from the top between Huajingxian 74 and each donor were determined via t-test. As a result, at least two traits were different significantly ($P \leq 0.01$) between Huajingxian 74 and each donor (Table 1).

QTLs detected for plant height and its components

A total of 24 QTLs for plant height and its components were detected on 14 substituted segments (Table 2), but no QTL was detected on other 38 substituted segments. The additive effects of twenty-four QTLs ranged from –4.08 to 3.98 cm, and the additive effect percentages of the QTLs varied from –19.35% to 10.43%.

Ten QTLs for plant height were detected on chromosomes 1, 2, 3, 4, 7, 8, 10 and 12, respectively. The additive effects of 7 QTLs were positive, ranging from 2.06 to 3.98 cm. The alleles increasing plant height were from IR64, Chenglongshuijingmi and IAPAR9, respectively. The additive effects of the other 3 QTLs were negative, with a range from –4.08 to –2.66 cm. The alleles decreasing plant height were from IR64, IRAT261 and Chenglongshuijingmi, respectively.

Two QTLs for panicle length ($qPL-4$ and $qPL-7$) were found on chromosomes 4 and 7. The panicle length
HE Feng-hua, et al. Identification of QTLs for Plant Height and Its Components by Using SSSLs in Rice

The QTLs for Plant Height and Its Components in the SSSLs were increased by the IAPAR9 allele at the qPL-4 locus and IR64 allele at the qPL-7 locus. Their additive effects ranged from 0.92 to 1.23 cm, and the additive effect percentages varied from 4.37% to 5.84%.

Four QTLs on chromosomes 1, 4, 7 and 12 were detected for Length of the first internode from the top. The IAPAR9 allele at qLFI-4 and the IR64 allele at qLFI-7 increased the length of the internode. The additive effects ranged from 1.08 to 1.40 cm, and the additive effect percentages varied from 3.62% to 4.69%. The IRAT261 allele at qLFI-1 and the Suyunuo allele at qLFI-12 decreased the length of the internode. The additive effects were –1.64 and –2.11 cm, and the additive effect percentages were –5.49% and –7.06%, respectively.

For Length of the second internode from the top, five QTLs on chromosomes 3, 8, 10 and 12 were involved. Length of this internode was increased by the Chenglongshuijingmi alleles both at qLSI-3-2 and qLSI-12, and by the IAPAR9 allele at qLSI-10. The additive effects ranged from 0.62 to 1.65 cm, and the additive effect percentages varied from 3.92% to 10.43%. Length of the internode was decreased by the IR64 allele at qLSI-3-1 and the Chenglongshuijingmi allele at qLSI-8. The additive effects were –0.87 and –0.73 cm, and the additive effect percentages were –5.50% and –4.61%, respectively.

The length of the third internode from the top was controlled by three QTLs on chromosomes 3, 6 and 10 respectively. Length of this internode was increased by the IAPAR9 alleles both at qLTI-6 and qLTI-10. The additive effects were 0.67 and 1.01 cm, and the additive effect percentages were 6.79% and 10.23%, respectively. The IR64 allele at qLTI-3 decreased length of the internode. The additive effect was –1.91 cm, and the additive effect percentage was –19.35%.

**QTL substitution mapping**

Nine QTLs were mapped in smaller intervals using substitution mapping strategy (Fig. 1).

qPH-1 and qLFI-1 were detected on the substituted segment of H18-01-03, but not detected in H18-01-01 with overlapping segment, which clearly
indicated that qPH-1 and qLFI-1 were located at the non-overlapping interval of RM243–RM493. The length of this interval was 33.8 cM.

qPH-3-2 and qLSI-3-2 were detected on the substituted segments of H20-03-02 and H20-03-03. Therefore, the two QTLs were located at the 23.7 cM overlapping interval of RM168–RM293.

qLTI-6 was identified on the substituted segment in H27-06-02, but not identified on the substituted segment in H27-06-03. This revealed that qLTI-6 was at the non-overlapping interval of RM508–RM589 with length of 3.0 cM.

The substituted segment of OSR22–RM18 in H08-07-02 can be divided into two intervals. The interval of OSR22–RM70 also was same as the substituted segment in H08-07-01 and the interval of RM234–RM18 was same as the substituted segment in H08-07-03. QTLs for plant height were detected on the substituted segments in the three SSSLs. The results indicated that there were two QTLs for plant height in the substituted segment of OSR22–RM18 in H08-07-02, one at the interval of OSR22–RM70 and the other at the interval of RM234–RM18. The QTL at the 18.7 cM interval of OSR22–RM70 was designated as qPH-7-1, and one at the 13.4 cM interval of RM234–RM18 was qPH-7-2. Both the qPH-7-1 allele and the qPH-7-2 allele from IR64 increased plant height, and their genetic effects were 2.54 and 2.91 cm, respectively. The additive effect for plant height increased to 3.52 cm when the qPH-7-1 and qPH-7-2 were on the same substituted segments in H08-07-02. Furthermore, qPL-7 was detected on both substituted segments in H08-07-01 and H08-07-02, but not detected on the segment in H08-07-03. This suggested that qPL-7 was localized at the interval of OSR22–RM70. qLTI-7 was detected on both segments in H08-07-02 and H08-07-03. Therefore, the qLTI-7 resided at the interval of RM234–RM18.

The 24 QTLs were distributed on 9 chromosomes except chromosomes 5, 9 and 11 (Fig. 2). qPH-2 and qLSI-12 identified in this study. Therefore, they are likely new genes involved in plant height or its components. The discovery of the new QTLs will be useful in rice breeding for improving plant type.

Some researchers have carried out QTL analyses for plant height and its components in rice. Comparison of the chromosomal locations of QTLs in this study with those in other reports suggests that the QTL qPH-4 and Ph4.1 detected by Moncada et al [8], qPL-7 and PLH detected by Brondani et al [9], qLTI-10 and Lti10 detected by Liu et al [19] are mapped in similar chromosomal locations. However, no similar reports are available on qPH-2, qPH-12 and qLSI-12 identified in this study. Therefore, they are likely new genes involved in plant height or its components. The discovery of the new QTLs will be useful in rice breeding for improving plant type.

Currently, several genes controlling plant height in rice have been cloned. Some genes were involved in the gibberellin (GA) metabolic pathway in rice [6, 23]. In comparison, three GA biosynthetic candidate genes OsKS1, OsKS2 and OsKS3 on chromosome 4 characterized by Sakamoto et al [23] were in similar chromosomal regions with qPH-4, qPL-4 and qLFI-4 identified on the substituted segment of H27-04-01 in this study. These results support Robertson’s hypothesis that the major and minor genes are
different alleles of the same loci \[24\].

Compared with conventional QTL analysis approach \[25\], the efficiency of QTL detection may be improved by the use of SSSLs \[19, 20\]. Additionally, research on complex epistatic interactions among QTLs can be performed without the disturbance of undesirable segments \[26\]. We have identified twenty-four QTLs for plant height and its components on the fourteen substituted segments, thereby enabling us to proceed to study their epistatic interactions.

Based on the pattern of decreasing plant height in rice, the genes controlling plant height can be categorized into three classes: (1) genes controlling number of upper internodes, (2) genes controlling length of upper internodes, and (3) genes for both number and length of upper internodes \[12\]. In typical japonica genetic background, semi-dwarf gene \(sd-1\) reduces plant height mainly via shortening length of upper internodes \[27\]. In the present study, nine chromosome segments carrying QTLs for plant height were common with those associated with panicle length and (or) length of upper internodes \[12\]. In typical japonica genetic background, semi-dwarf gene \(sd-1\) reduces plant height mainly via shortening length of upper internodes \[27\]. In the present study, nine chromosome segments carrying QTLs for plant height were common with those associated with panicle length and (or) length of upper internodes, which reflected the genetic basis of panicle length and length of upper internodes as the components of plant height, and indicated that plant height was controlled by QTLs for panicle length and QTLs for length of upper internodes simultaneously. Plant height is one of the important breeding traits for plant type in rice. The QTLs for plant height and its components are useful in rice breeding program for constructing ideotype.

ACKNOWLEDGEMENTS

This work was supported by the key project of National Natural Science Foundation of China (30330370) and the team project of Natural Science Foundation of Guangdong Province (20003023).

REFERENCES


