Mapping quantitative trait loci associated with callus browning in Dongxiang common wild rice (Oryza rufipogon Griff.)

Yibo Wang
China Agricultural University
Xin Yang
Rural Development Institute Chinese Academy of Social Sciences
Guimin Xu
China Agricultural University
Xiao Ye
China Agricultural University
Yiming Ji
China Agricultural University
Xin Lou
China Agricultural University
Jingjing Su
China Agricultural University
Chuanqing Sun
China Agricultural University
Yongcai Fu
China Agricultural University
Zhang Kun (kunzhang@cau.edu.cn)
China Agricultural University

Research Article

Keywords: Common wild rice, Callus browning, Introgression lines, QTL mapping

Posted Date: November 21st, 2022

DOI: https://doi.org/10.21203/rs.3.rs-2262011/v1

Version of Record: A version of this preprint was published at Molecular Biology Reports on January 24th, 2023. See the published version at https://doi.org/10.1007/s11033-023-08279-7.

Abstract

Background: The genetic transformation of *indica*rice (*Oryza sativa* ssp. *indica*) is limited by its poor *in vitro* tissue culturability, especially callus browning. Elucidating the common wild rice (*Oryza rufipogon* Griff.) genes controlling callus browning is a literally fundamental method for improving the tissue culturability of *indica* rice varieties.

Methods and results: In the present study, we used a population of 129 *O. rufipogon* (Dongxiang common wild rice; DXCWR) introgression lines in the elite cultivar GC2 (*Oryza sativa* ssp. *indica*) background and 159 simple sequence repeat (SSR) markers to identify quantitative trait loci (QTLs) associated with callus browning in rice. The callus browning phenotype was evaluated using the indices of the callus browning rate (CBR), callus browning index (CBI), and standard callus browning index (SCBI).

Conclusions: We detected 30 QTLs associated with callus browning across the genotypes, which were located on chromosomes 1, 2, 3, 4, 8, 9, and 12. These were repeatedly associated with differences in CBR, CBI, and SCBI. The alleles from DXCWR had an additive effect in reducing callus browning. Especially, we identified new QTLs near RM247 an RM7003 on chromosome 12, indicating the QTLs were unique in DXCWR. Furthermore, we identified six introgression lines with significantly reduced callus browning, which is expected that these lines will be useful materials for the genetic transformation and fine mapping of the culturability trait.

Introduction

Efficient genetic transformation is crucial for functional studies of genes, molecular breeding, and the development of genetically modified crops. The *Agrobacterium tumefaciens*–mediated transformation of *indica* rice accessions (*Oryza sativa* ssp. *indica*) is hindered by callus browning, which is less of a problem in *japonica* accessions (*O. sativa* ssp. *japonica*). Callus browning is a common feature in many plant species, resulting in a decreased regenerative ability, poor growth, and even death (He et al. 2009; Zhang et al. 2020). In previous studies, various physiological and biochemical indices (Murata et al. 2001; Tang and Newton, 2004; Sarmadi et al. 2018; Zhang et al. 2020; Ogawa et al. 1999) and hormones (Adkins et al. 1990; Kobayashi et al. 1991; Zhang et al. 2020) were shown to affect callus browning; however, the most important factor determining the likelihood of callus browning is the genotype. Elucidating and utilizing alleles associated with a reduction in callus browning should therefore be a major target for functional genetics analyses, genome editing, and molecular breeding (Jia et al. 2007).

Several quantitative trait loci (QTLs) associated with the rice response to tissue culture have already been identified (Taguchi-Shiobara et al. 1997, 2006; Takeuchi et al. 2000; Nishimura et al. 2005; Ozawa and Kawahigashi, 2006; Li et al. 2007; Zhao et al. 2009; Li et al. 2013; Zhang et al. 2016, 2020; Huang et al. 2021); however, only two of the genes responsible for QTLs associated with callus culturability have been isolated. One of these genes encoded ferredoxin-nitrite reductase (NiR), which controls callus differentiation on chromosome 1(Nishimura et al. 2005), and the other was *BROWNING OF CALLUS1*

(*BOC1)*, encoding a SIMILAR TO RADICAL-INDUCED CELL DEATH ONE (SRO) protein involved in callus browning on chromosome 3 (Zhang et al. 2020). However, few QTLs controlling tissue culture ability have been mined from common wild rice (*Oryza rufipogon* Griff.).

Wild rice is an important favorable gene pool (Wang et al. 1992; Sun et al. 2001, 2002). The genetic background of the introgression lines is single, with only a few introgression frangment differences, which lays the foundation for genetic analysis and functional identification (Eshed and Zamir, 1995;Tan et al. 2007). Compared with conventional materials, introgression lines derived from the common wild rice are more accurate and efficient for gene mapping and cloning. In previous studies using map-based cloning, our team identified two important genes derived from Dongxiang common wild rice (DXCWR) on the background of *indica* GC2 that were associated with the number of grains produced (Huo et al. 2017) and cold tolerance at the early seedling stage (Zhao et al. 2020). GC2 is an elite *indica* rice variety; however, this genotype is susceptible to callus browning, making its genetic transformation much more challenging.

In the present study, we investigated the callus browning trait of 129 introgression lines bearing variable genetic material from DXCWR in *indica* cultivar GC2 background at 30 days after inoculation on the induction medium. The introgression lines were genotyped with 159 simple sequence repeat (SSR) markers, and a QTL analysis was performed using the single-point analysis method, laying a solid foundation for the further fine mapping and cloning of the genes involved in tissue culturability.

Materials And Methods

Plant materials

A total of 129 *O. rufipogon* introgression lines (BC_4F_4; named 2DIL), derived from a cross between the *O. rufipogon* accession DXCWR as the donor and the elite *indica* variety GC2 (*O. sativa*) as the recipient, were screened from 265 lines on the basis of their chromosome genotype distributions. These lines were used to assess callus browning.

Media

The unimproved NB (NB1) medium used for the callus induction consisted of N6 macronutrient components (Chu et al. 1975), B5 micronutrient components, and organic components (Gamborg et al.1968), supplemented with 2 mg/L of 2,4-D and 30 g/L of sucrose (Zhang et al. 2020). The pH of the medium was adjusted to 5.8–6.0 with 1 N KOH, and 3 g/L of phytagel was added before autoclaving at 121°C for 20 min.

Tissue Culture Procedure

After hulling, ~ 90 mature, healthy, dehusked seeds from each line were sterilized by immersion in 70% ethanol for ~ 2 min, followed by a 15% sodium hypochlorite solution for 15 min with shaking. They were then rinsed three or four times with sterile water on an ultraclean workbench. The sterilized seeds were placed on the induction medium in three dishes (25 ~ 30 seeds per dish) and incubated in the dark at 28°C for 30 days. The callus browning phenotypes of the calli in each dish were recorded as described below. For each replicate, all the introgression lines were divided into five subgroups, each containing 30 introgression lines and the recurrent parent GC2.

Phenotypic Evaluation Of Callus Browning

The phenotypic observations of callus browning were performed as described by Zhang et al. (2020). The tendency for callus browning was categorized into five levels: (0) less than 1/10 of the callus tissue was brown; (1) 1/10 ~ 1/3 of the callus tissue was brown; (2) 1/3 ~ 2/3 of the callus tissue was brown; (3) 2/3 ~ 1 of the callus tissue was brown; and (4) the callus was completely brown. To minimize phenotyping errors due to observations being made by different people or differences arising from variations in the cultivation environment, the phenotypes were standardized using the callus browning index (SCBI) to avoided the influences of these factors as far as possible. The tendency for callus browning was calculated according to equations (1), (2), and (3):

(1) Callus browning rate (CBR) = (number of calli showing browning / number of produced calli) × 100%;

(2) Callus browning index (CBI) = (Σ of number of calli at each browning level × browning level) / (number of produced calli × highest browning level);

(3) Standard callus browning index (SCBI) = (CBI of the same batch – CBI of GC2 in the same batch) / CBI of GC2 in the same batch.

Qtl Analysis

A total of 159 SSR primers were used to identify the genotypes of the 129 introgression lines. The association between the phenotype and the marker genotype was investigated using a single-point analysis in the software Map Manager QTXb20 (Manly et al. 2001). The statistical threshold for the single-point analysis was $P < 0.05$.

Statistical analysis

The program GGT2.0 (Van Berloo 1999) was used to construct the graphic genotype. The phenotype data were converted into percentages in Microsoft Excel, and were statistically analyzed using SPSS v25.0 software, which included drawing the frequency distributions, performing analyses of variance (ANOVAs), and performing correlation analyses of the three indices between the introgression lines.

Results

Phenotypic variations in callus browning between DXCWR, the 2DIL introgression lines, and the recurrent parent GC2

In our previous study, we constructed a set of introgression lines (named 2DIL) using the *O. rufipogon* accession DXCWR as a donor and the elite *indica* cultivar GC2 (*O. sativa*) as the recurrent parent. DXCWR was found to be relatively resistant to browning, with a CBI of 0.22 (Zhang et al. 2020; Table 1). To identify QTLs involved in the regulation of callus browning in rice, we screened 129 introgression lines by inoculating mature seeds on unimproved NB medium (NB basal medium without additives, named NB1) for 30 days (Fig. 1). Meanwhile, based on the genotypes of the 129 introgression lines, we constructed the graphic genotype used the program GGT2.0 (Fig. 2). We divided the tendency for callus browning into five levels, compared to GC2, 0 level was recorded as no browning, 1 and 2 level was considered as medium browning, but 3 and 4 level was serious browning similar to GC2 (Fig. 1b–f). The recurrent parent GC2 was more susceptible to callus browning than DXCWR (Fig. 1a and Table 1). The callus browning rate (CBR), callus browning index (CBI), and the standard callus browning index (SCBI) of the 2DIL population varied significantly, ranging from 0.00 to 100%, 0.00 to 0.93, and − 1.00 to 0.40, respectively (Table 1 and Fig. 3).

Variance And Correlation Among The Three Callus Browning Indices

The three indices related to callus browning varied substantially among the introgression lines (Table 2). The ANOVA revealed that the highly significant variation in the three indices was due to a genotype effect (Table 2). The correlation among these indices was determined by calculating the correlation coefficients (r), revealing all indices were positively correlated: CBR with CBI (r = 0.895), CBR with SCBI (r = 0.828), and CBI with SCBI (r = 0.938) (Table 3). Consequently, CBR, CBI, and SCBI were all used for QTL analysis.

Identification Of Qtls For Callus Browning (Dup: Abstract ?)

A total of 30 putative QTLs closely related to the callus browning phenotype were identified, including 10 QTLs for CBR (*qCBR1-1 ~ qCBR12-2*), which explained between 3% and 11% of the phenotypic variation in this trait; 10 QTLs for the CBI (*qCBI2-1 ~ qCBI12-2*), explaining 3–8% of CBI variation; and 10 QTLs for SCBI (*qSCBI2-1 ~ qCBI12-2*), explaining 4–8% of the phenotypic variation in this trait (Table 4 and Fig. 4). These QTLs were located on chromosomes 1, 2, 3, 4, 8, 9, and 12 (Table 4 and Fig. 4). The QTLs linked to markers RM335 on chromosome 4, RM189 on chromosome 9, and RM7003 on chromosome 12 were found to be associated with all three indices. The QTLs accounted for 5–11% of the phenotypic variation, and the additive effect of the DXCWR alleles could alleviate callus browning (Table 4).

Screening Elite Introgression Lines With Low Callus Browning Level

We screened six introgression lines (2DIL18, 2DIL99, 2DIL101, 2DIL103, 2DIL110, and 2DIL112) exhibiting a significant reduction in callus browning compared with GC2 (Fig. 5). Genotype analysis showed that the six selected introgression lines possessed introgression fragments distributed across different chromosomes: 2DIL112 had an introgression fragment near marker RM71 on chromosome 2; 2DIL101 had an introgression fragment near marker RM114 on chromosome 3; 2DIL18 and 2DIL110 had introgression fragments near marker RM335 on chromosome 4; and 2DIL99, 2DIL103, and 2DIL110 had introgression fragments near markers RM328, OSR29, and RM189 on chromosome 9. These lines can be used not only as *indica* rice accession genetic transformation receptors, but can also be programmed as parent materials for the fine-mapping of callus browning.

Discussion

DXCWR contains a wealth of new gene/allele sources of tolerance to abiotic stress

To our knowledge, an efficient tissue-culture and transformation system using mature seeds has been established for *japonica* rice; however, the genetic transformation efficiency of *indica* rice is lower due to callus browning. While most genetic analyses of callus browning are based on cultivated rice, few studies have investigated alleles associated with callus browning that have been lost in the common wild rice. In the previous study, we believe that *BOC1* derived from a common wild rice accession (*Oryza rufipogon* G.) was related to cell senescence and death caused by oxidative stress (Zhang et al. 2020). In this study, DXCWR displayed significantly less callus browning than GC2 (Table 1), and the range variation of callus browning indices in the introgression lines (2DILs) were wide (Table 1; Fig. 3), indicating that DXCWR may harbor favorable alleles for reducing callus browning in cultivated rice.

Accuracy And Reproducibility Of The Phenotypic Assessment

The identification of tissue culture traits is influenced by various factors, such as the genotype, the culture medium, the selection of explants, the physiological status of the donor material, and the interactions between them (Abe and Futsufara, 1986; Hanna and Monika, 2006). Phenotypic evaluation can also suffer from artificial errors due to subjective differences between different people and different survey criteria, even when using the same genotype in the same laboratory (Li et al. 2013). In our study, we investigated the callus browning trait under the supervision of the same person and according to using the same criteria to minimize error. We also set up three replicates per introgression line, and our callus browning trait results were stable and reproducible. In some previous studies, researchers also identified callus status and proliferation ability in subculture (Taguchi-Shiobara et al. 2006; Li et al. 2007; Zhao et al. 2009); however, in the present study, to observe the maximum phenotypic variations for callus browning in the introgression lines and reduce the incubation time to decrease the workload, we investigated callus browning cultured on NB1 medium after 30 days. The selection not to scutellum-derived calli would have increased our working efficiency and speed up the experiments.

Identification Of Qtls For Callus Browning

In our study, a total of 30 QTLs related to callus browning were identified on chromosomes 1, 2, 3, 4, 8, 9, and 12, and the QTL alleles derived from *O. rufipogon* were all associated with reducing callus browning. In a previous study, using 183 BC_1F_3 lines derived from two varieties, Koshihikari and Kasalath, Taguchi-Shiobara et al. (2006) identified two QTLs, *qlc4* and *qlc9*, associated with the induced-callus color, as well as the QTL *qSc4*, which was related to the color of the subcultured callus. These QTLs were all identified in our study. Similarly, using 192 recombinant inbred lines (RILs) derived from YZX (*O. sativa* ssp. *indica*) × 02428 (*O. sativa* ssp. *japonica*) and a high-density bin map, Huang et al. (2021) mapped a QTL on chromosome 9 to a ~ 100-kb genomic interval, which was also identified in the present study. Furthermore, in our previous study, we detected a QTL associated with callus browning at the RM335 locus on chromosome 4 (Zhang et al. 2016). This was achieved using 127 *O. rufipogon* introgression lines, which were constructed using an *O. rufipogon* accession collected from Yuanjiang County, Yunnan Province, China, as the donor and the elite *O. sativa indica* cultivar 93 − 11 as the recipient (Zhang et al. 2016). The QTLs located on the short arm of chromosome 4 and the long arm of chromosome 9 were hotspots for the control of the callus browning phenotype in rice. Furthermore, surprisingly, we identified a new QTL with high phenotypic variance on chromosome 12, which has not been reported before, indicating that it may be unique to DXCWR. These results indicated that the QTLs in our study are worthy of further gene mapping and functional analysis for decreasing callus browning and improving genetic transformation efficiency.

Conclusion

We used 129 introgression lines (2DILs) derived from DXCWR (*O. rufipogon*) in the elite *indica* accession GC2 background to locate the quantitative trait loci related to callus browning trait. The three QTL sites near RM71 on chromosome 2, RM335 on chromosome 4, and RM328, RM189 on chromosome 9 were identified based on the CBR, CBI and SCBI indices. These QTLs accounted for 4%-8% of the phenotypic variations at low callus browning. The QTL near RM247 and RM7003 on chromosome 12 was a new QTL which reduced the callus browning. Moreover, we identified six elite introgression lines with low callus browning. These results provided the materials and genetic resources for the genetic transformation and solving the molecular breeding bottleneck of *indica* rice.

Abbreviations

QTLs Quantitative trait loci

BOC1 BROWNING OF CALLUS1

NiR Ferredoxin-nitrite reductase

SSR Simple sequence repeat

CBR Callus browning rate

CBI Callus browning index

SCBI Standard callus browning index

DXCWR Dongxiang common wild rice

ANOVA Analyses of variance

Declarations

Author Contributions: K.Z. and Y.F. conceived and designed the experiments. Y.W. analyzed the phenotypic data. Y.W., X.Y., and G.X. performed the experiments. X.Y., Y.J, X.L., and J.S. helped to perform the experiments. X.L. helped to perform the QTL analysis. C.S. and Y.F. supervised the research and modified the manuscript. K.Z. analyzed the data and wrote the manuscript. All authors have read and agreed to the published version of the manuscript.

Data availability: All data generated or analyzed during this study are included in this published article.

Funding: This project was supported by the National Natural Science Foundation of China (grant numbers 31571626 and 3217150449) and the National Transgenic Major Project of China (grant number 2016ZX08001-004).

Conflicts of Interest: The authors declare that they have no conflict of interest.

Ethical approval : The current study does not involve any animal or human study.

Informed consent: Not applicable for current study.

References

1. Abe T, Futsuhara Y (1986) Genotypic variability for callus formation and plant-regeneration in rice (*Oryza sativa* L). Theor Appl Genet 72:3–10
2. Adkins SW, Shiraishi T, Mccomb JA (1990) Rice callus physiology - identification of volatile emissions and their effects on culture-growth. Physiol Plant 78:526–531
3. Bolibok H, Rakoczy-Trojanowska M (2006) Genetic mapping of QTLs for tissue-culture response in plants. Euphytica 149:73–83
4. Chu CC, Wang CC, Sun CC, Chen H, Yin KC, Chu CY, Bi FY (1975) Establishment of an efficient medium for anther culture of rice through comparative experiments on nitrogen-sources. Sci China Chem 18:659–668
5. Eshed Y, Zamir D (1995) An introgression line population of *Lycopersicon pennellii* in the cultivated tomato enables the identification and fine mapping of yield-associated QTL. Genetics 141:1147–

1162

6. Gamborg OL, Miller RA, Ojima K (1968) Nutrient requirements of suspension cultures of soybean root cells. Exp Cell Res 50:151–158
7. He Y, Guo XL, Lu R, Niu B, Pasapula V, Hou P, Cai F, Xu Y, Chen F (2009) Changes in morphology and biochemical indices in browning callus derived from Jatropha curcas hypocotyls. Plant Cell Tiss Org 98:11–17
8. Huang CH, Zhang J, Zhou DH, Huang YT, Su L, Yang GL, Luo WL, Chen ZQ, Wang H, Guo T (2021) Identification and candidate gene screening of *qCIR91* a novel QTL associated with anther culturability in rice (*Oryza sativa* L). Theor Appl Genet 134:2097–2111
9. Huo X, Wu S, Zhu ZF, Liu FX, Fu YC, Cai HW, Sun XY, Gu P, Xie DX, Tan LB, Sun CQ (2017) *NOG1* increases grain production in rice. Nat Commun 8:1497
10. Jia HY, Yi DL, Yu J, Xue SL, Xiang Y, Zhang CQ, Zhang ZZ, Zhang LX, Ma ZQ (2007) Mapping QTLs for tissue culture response of mature wheat embryos. Mol Cells 23:323–330
11. Kobayashi Y, Fukui H, Tabata M (1991) Effect of carbon-dioxide and ethylene on berberine production and cell browning in *Thalictrum minus* cell-cultures. Plant Cell Rep 9:496–499
12. Li SJ, Yan S, Wang A -hong, Zou GH, Huang XH, Han B, Qian Q, Tao YZ (2013) Identification of QTLs associated with tissue culture response through sequencing-based genotyping of RILs Derived from 93-11 x Nipponbare in rice (*Oryza sativa*). Plant Cell Rep 32:103–116
13. Li Z, Duan SH, Kong J, Li SQ, Li YS, Zhu YG (2007) A single genetic locus in chromosome 1 controls conditional browning during the induction of calli from mature seeds of *Oryza sativa ssp indica*. Plant Cell Tiss Org 89:237–245
14. Manly KF, Cudmore RJ, Meer JM (2001) Map Manager QTX cross-platform software for genetic mapping. Mamm Genome 12:930–932
15. Murata M, Nishimura M, Murai N, Haruta M, Homma S, Itoh Y (2001) A transgenic apple callus showing reduced polyphenol oxidase activity and lower browning potential. Biosci Biotechnol Biochem 65:383–388
16. Nishimura A, Ashikari M, Lin S, Takashi T, Angeles ER, Yamamoto T, Matsuoka M (2005) Isolation of a rice regeneration quantitative trait loci gene and its application to transformation systems. Proc Natl Acad Sci USA 102:11940–11944
17. Ogawa T, Fukuoka H, Yano H, Ohkawa Y (1999) Relationships between nitrite reductase activity and genotype-dependent callus growth in rice cell cultures. Plant Cell Rep 18:576–581
18. Ozawa K, Kawahigashi H (2006) Positional cloning of the nitrite reductase gene associated with good growth and regeneration ability of calli and establishment of a new selection system for *Agrobacterium*-mediated transformation in rice (*Oryza sativa* L). Plant Sci 170:384–393
19. Sarmadi M, Karimi N, Palazon J, Ghassempour A, Mirjalili MH (2018) The effects of salicylic acid and glucose on biochemical traits and taxane production in a *Taxus baccata* callus culture. Plant Physiol Bioch 132:271–280

20. Sun CQ, Wang XK, Li ZC, Yoshimura A, Iwata N (2001) Comparison of the genetic diversity of common wild rice (*Oryza rufipogon* Griff) and cultivated Rice (*O. sativa* L) using RFLP markers. Theor Appl Genet 102:157–162
21. Sun CQ, Wang XK, Yoshimura A, Doi K (2002) Genetic differentiation for nuclear mitochondrial and chloroplast genomes in common wild rice (*Oryza rufipogon* Griff) and cultivated rice (*Oryza sativa* L). Theor Appl Genet 104:1335–1345
22. Taguchi-Shiobara F, Lin SY, Tanno K, Komatsuda T, Yano M, Sasaki T, Oka S (1997) Mapping quantitative trait loci associated with regeneration ability of seed callus in rice *Oryza sativa* L. Theor Appl Genet 95:828–833
23. Taguchi-Shiobara F, Yamamoto T, Yano M, Oka S (2006) Mapping QTLs that control the performance of rice tissue culture and evaluation of derived near-isogenic lines. Theor Appl Genet 112:968–976
24. Takeuchi Y, Abe T, Sasahara T (2000) RFLP mapping of QTLs influencing shoot regeneration from mature seed-derived calli in rice. Crop Sci 40:245–247
25. Tan LB, Liu FX, Xue W, Wang GJ, Ye S, Zhu ZF, Fu YC, Wang XK, Sun CQ (2007) Development of *Oryza rufipogon* and *O sativa* introgression lines and assessment for yield-related quantitative trait loci. J Integr Plant Biol 49:871–884
26. Tang W, Newton RJ (2004) Increase of polyphenol oxidase and decrease of polyarnines correlate with tissue browning in Virginia pine (*Pinus virginiana* Mill). Plant Sci 167:621–628
27. Van Berloo R (1999) GGT: Software for the display of graphical genotypes. J Hered 90:328–329
28. Wang ZY, Second G, Tanksley SD (1992) Polymorphism and phylogenetic-relationships among species in the genus *Oryza* as determined by analysis of nuclear RFLPs. Theor Appl Genet 83:565–581
29. Zhang K, Su JJ, Xu M, Zhou ZH, Zhu XY, Ma X, Hou JJ, Tan LB, Zhu ZF, Cai HW, Liu FX, Sun HY, Gu P, Li C, Liang YT, Zhao WS, Sun CQ, Fu YC (2020) A common wild rice-derived *BOC1* allele reduces callus browning in *indica* rice transformation. Nat Commun 11:443
30. Zhang K, Yin Z, Xu X, Pu C, Li Q, Wu D, Sun CQ, Fu YC (2016) Quantitative trait loci for mature embryo culturability traits from Yuanjiang common wild rice (*Oryza rufipogon* Griff). Indian J Genet Pl Br 76:167–172
31. Zhao J, Wang SS, Qin JJ, Sun CQ, Liu FX (2020) The lipid transfer protein OsLTPL159 is involved in cold tolerance at the early seedling stage in rice. Plant Biotechnol J 18:756–769
32. Zhao LN, Zhou HJ, Lu LX, Liu L, Li XH, Lin YJ, Yu SB (2009) Identification of quantitative trait loci controlling rice mature seed culturability using chromosomal segment substitution lines. Plant Cell Rep 28:247–256

Tables

Tables 1 to 4 are available in the Supplementary Files section

Figures

Fig. 1

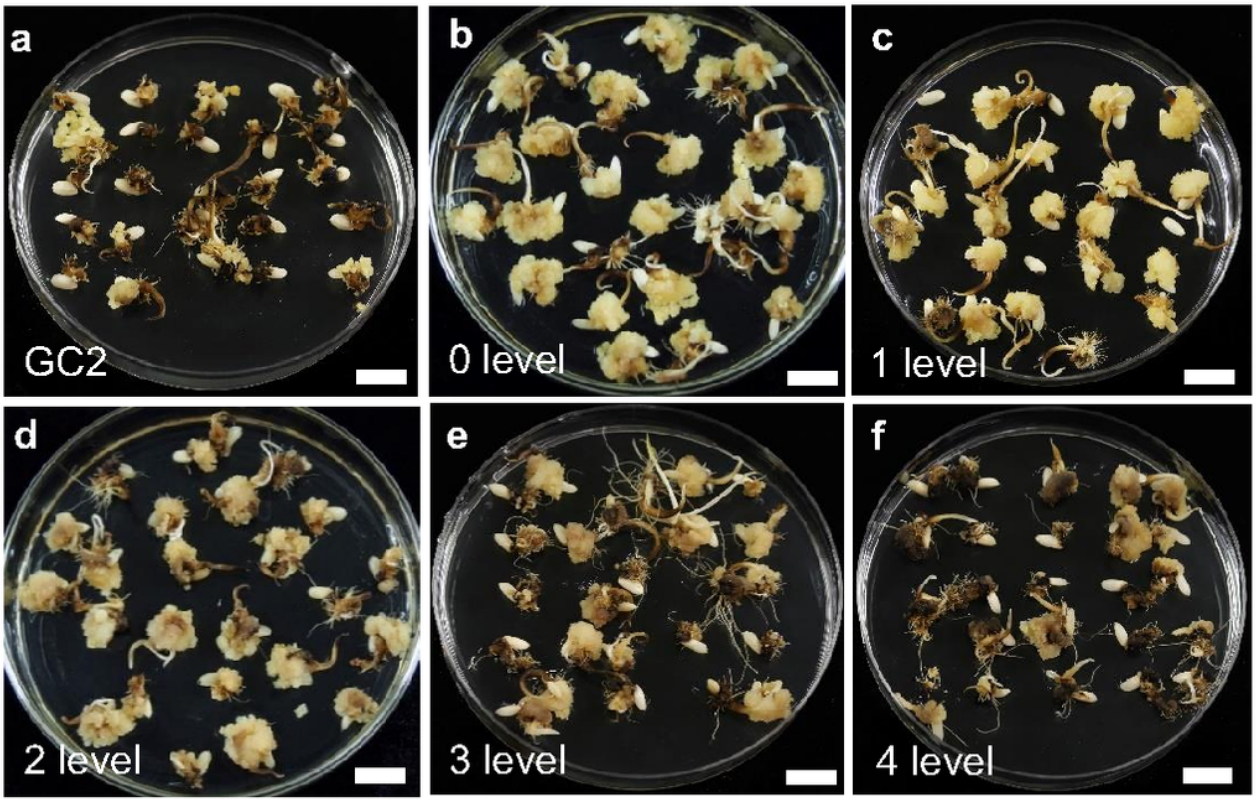

Figure 1

Five categories of callus browning in the *Oryza rufipogon* DXCWR 2DIL introgression lines, with the recurrent parent GC2

Fig. 2

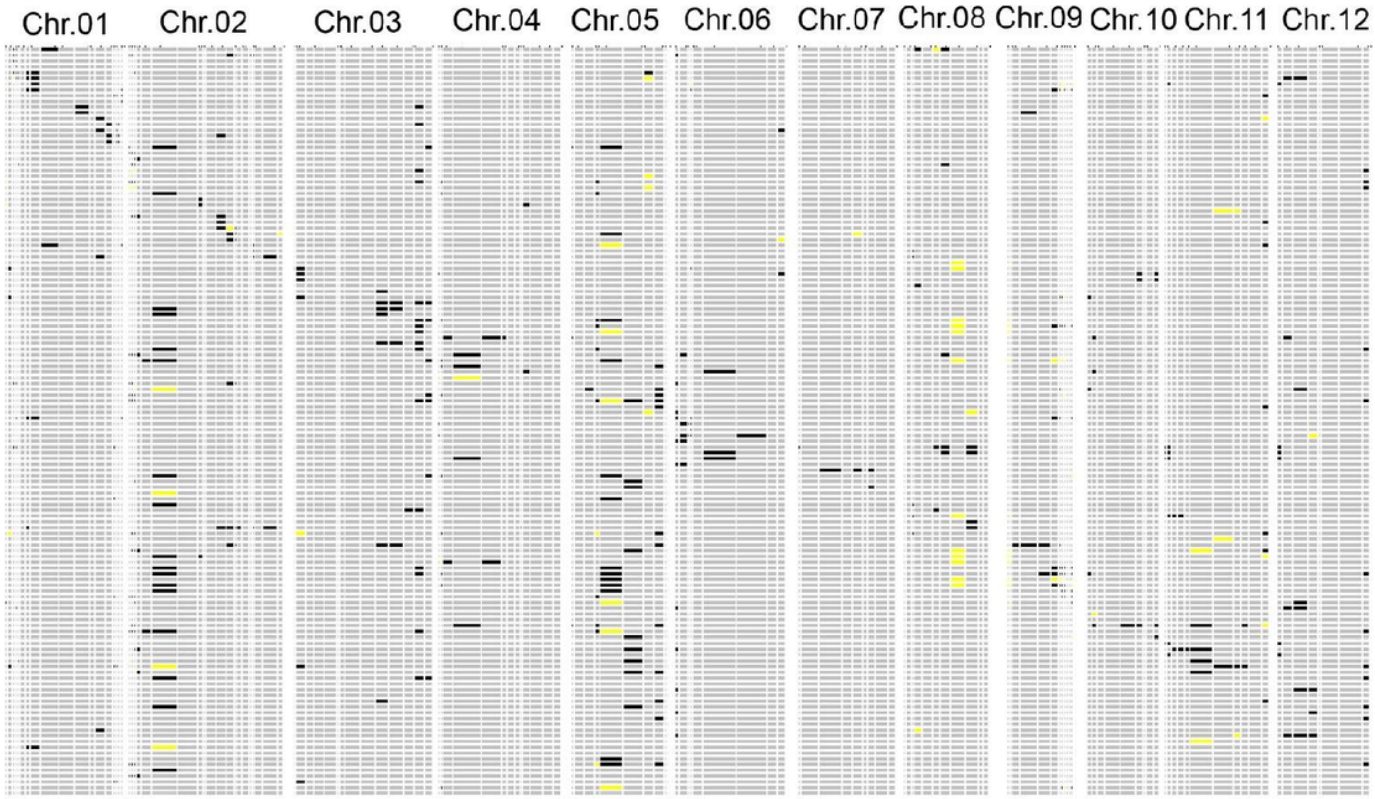

Figure 2

Graphical genotypes of the 129 introgression lines of Dongxiang common wild rice (*Oryza rufipogon*) in the *indica* rice (GC2; *Oryza sativa*) background used in this study.

The black regions show the regions homozygous for Dongxiang common wild rice (*O. rufipogon / O. rufipogon*); the yellow regions indicate the heterozygous regions (*O. rufipogon / O. sativa*); the gray regions indicate the homozygous regions for GC2 (*O. sativa / O. sativa*). The horizontal axis indicates every introgression line, and the vertical axis indicates each introgressed segment of Dongxiang common wild rice.

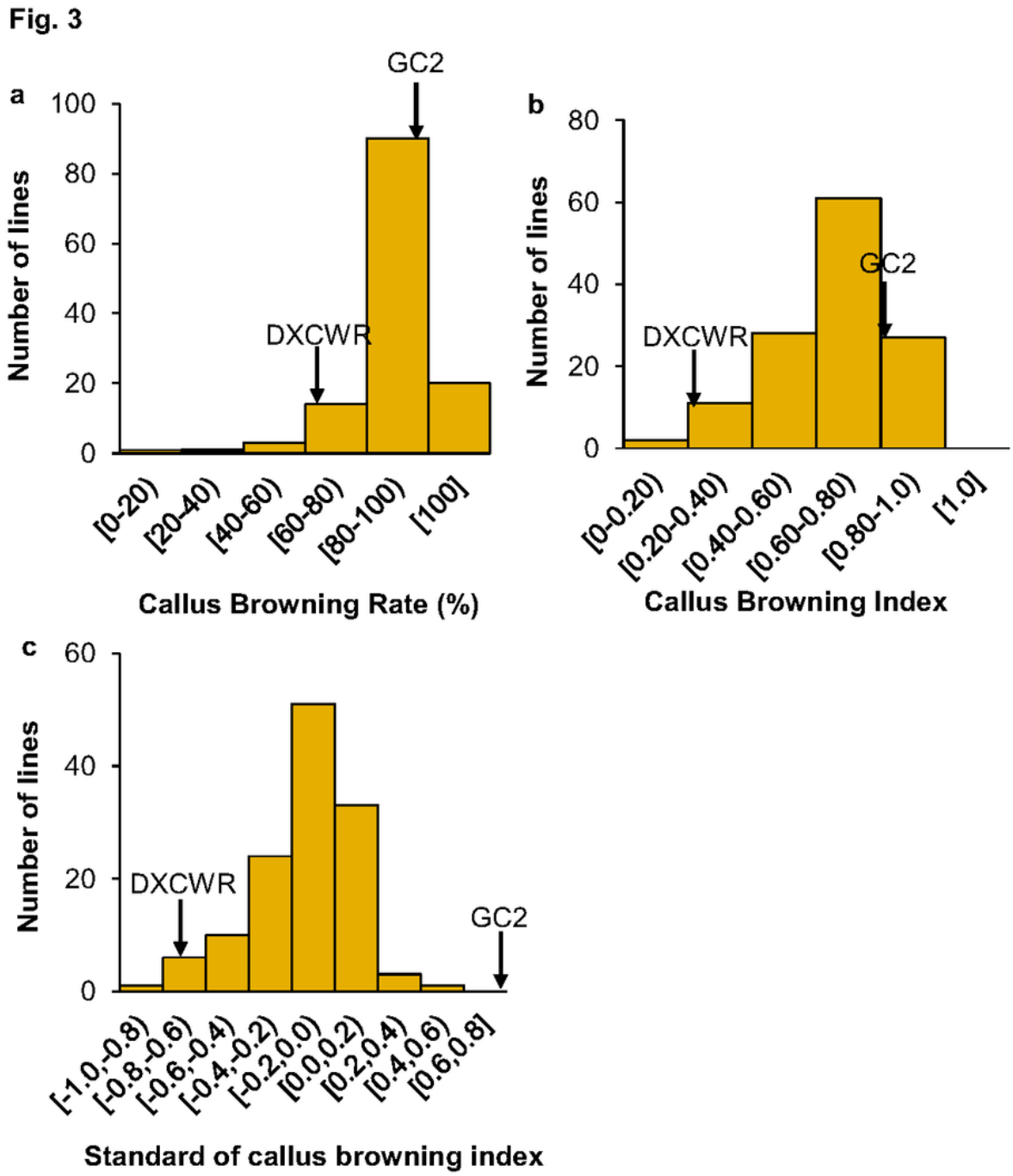

Figure 3

Frequency distribution of the three indices of callus browning in the rice *Oryza rufipogon* DXCWR 2DIL introgression lines

Fig. 4

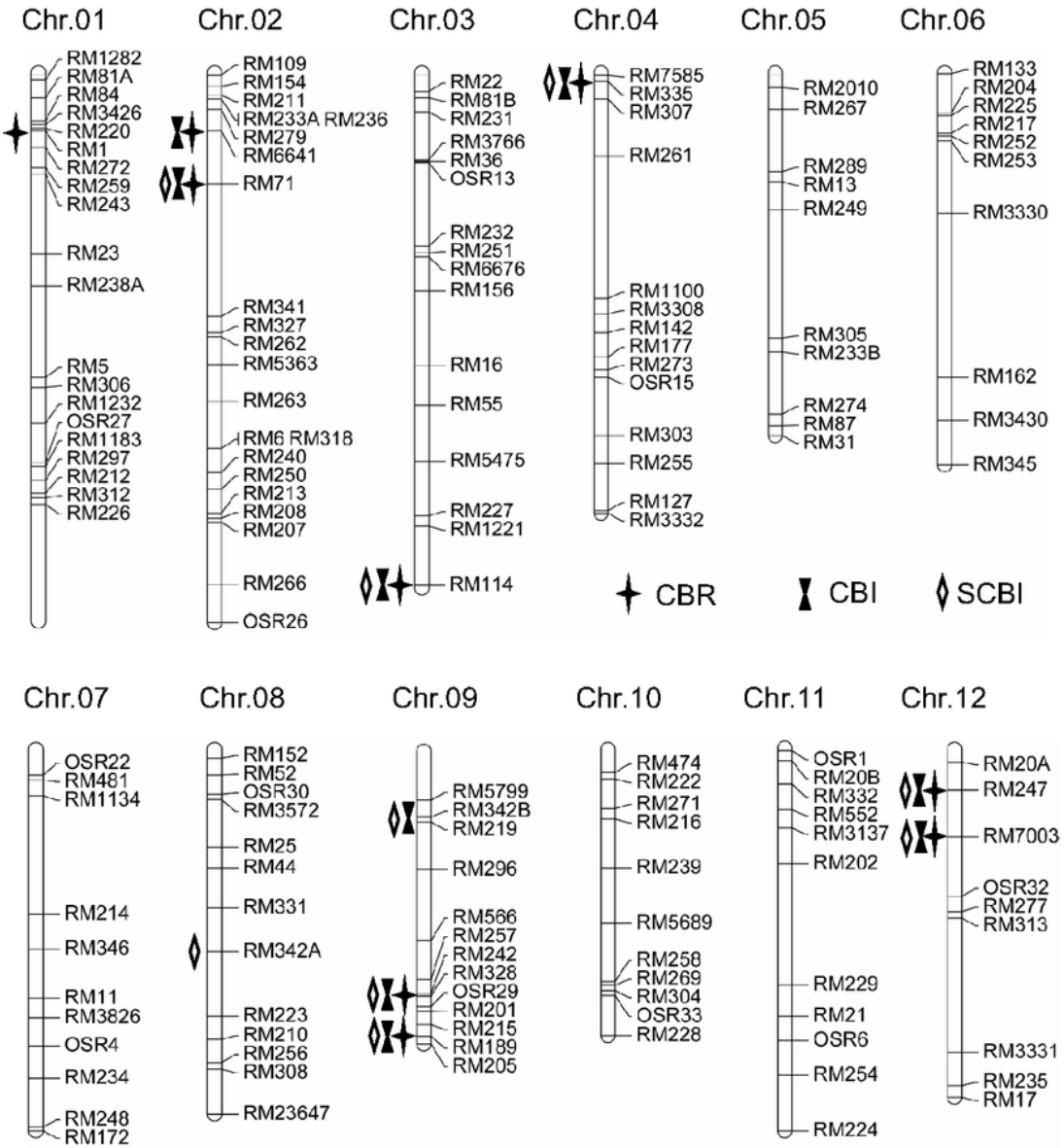

Figure 4

Distribution of the putative QTLs associated with the callus browning trait on the 12 rice chromosomes, detected using a single-point analysis

Fig. 5

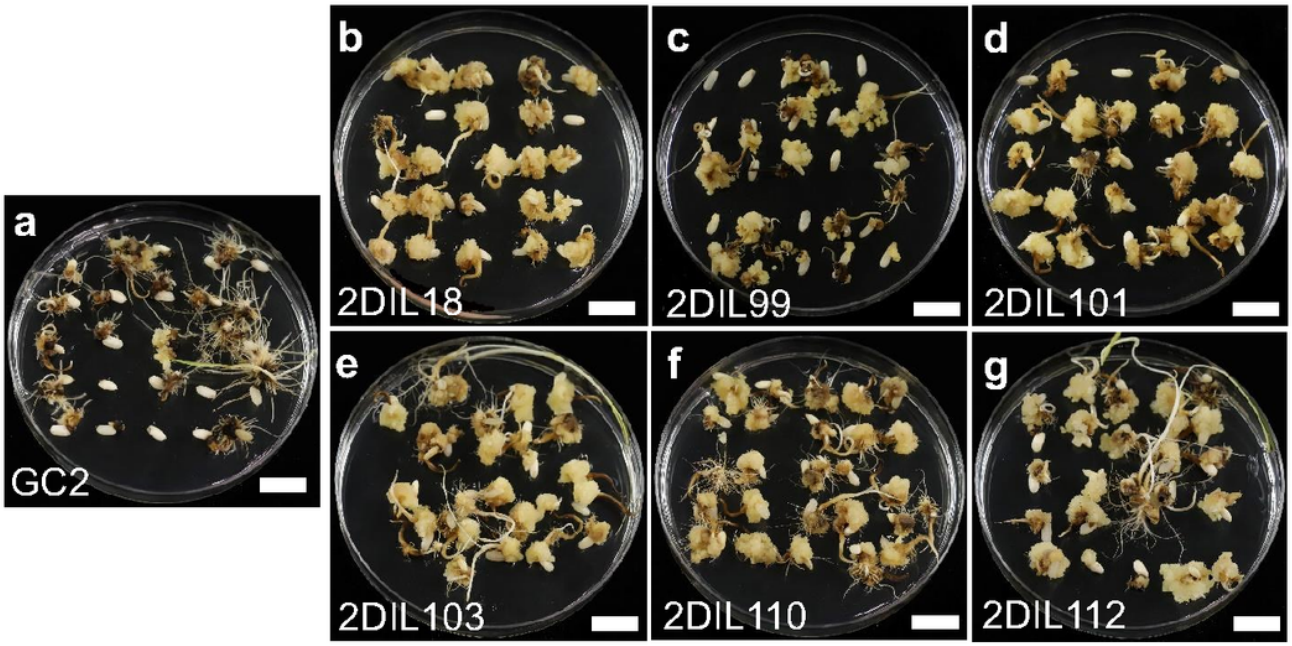

Figure 5

Introgression lines with light callus browning

Supplementary Files

This is a list of supplementary files associated with this preprint. Click to download.

- Table.pdf