Journal of Pharmacognosy and Phytochemistry
Available online at www.phytojournal.com
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Journal of Pharmacognosy and Phytochemistry

E-ISSN: 2278-4136
P-ISSN: 2349-8234
JPP 2018; 7(5): 1571-1574
Received: 10-07-2018
Accepted: 12-08-2018

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Studies on response to drought susceptibility index and grain yield in early backcross generation lines in rice (*Oryza sativa* L.)

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Abstract

Rice under rainfed condition faces frequent moisture stress. Recent studies at IRRI have shown moderate to high heritability of grain yield under stress. Evaluation was done for reproductive stage moisture stress tolerance in severe moisture stress and well irrigated condition to study the effect of the introgressed QTLs in BILs. QTLs were introgressed through backcrossing of Apo (donor) and IR 64 (recipient) lines and Set of 81 backcross inbred lines (BILs) at BC_1F_3 generations developed through MAS (Marker Assisted Selection) bearing one at a time, two and three DTY (yield under drought) QTLs (Quantitative Trait Loci) for yield under stress donated by Apo. Single QTLs DTY2.2 performed best for grain yield and also showed least drought susceptibility index (DSI). Two and three QTL combination in the background of IR 64 viz., [DTY (3.1+8.1)] and [DTY (2.2+3.1+8.1)] QTL combinations recorded lesser DSI (Drought Susceptible Index) values than their parents and check DSI values**.** DTY (2.2 + 8.1) performed better in traits like grain yield than expected three QTL line. QTLs bearing BILs were advantageous over IR 64 under severe moisture stress with respect grain yield and direct us to further studies regarding QTLs effect on drought susceptibility index and its relationship with grain yield.

Keywords: Drought tolerance, QTL, marker assisted selection, rice, drought susceptibility index

Introduction

Rice (*Oryza sativa* L.) is the staple food for more than half of the world's population, especially those living in developing countries such as India, China, Bangladesh, Laos, Vietnam and Indonesia. Green Revolution has increased the rice production by 2.6 times since 1961 but focused mainly on irrigated ecosystems. Drought is ubiquitous constraint and is a source of destabilization of yield in rice. Strictly less than half of the world's rice area is irrigated; the rest of the rice area relies on rainfall for its water requirement. Upland rice, produced by small holder farmers in India, is the lowest-yielding rice production system. The ascending global shortage of water is a great hindrance for rice production as rice requires high amount of water. Recent studies at IRRI have shown moderate to high heritability of grain yield under drought thus opening area for direct selection for grain yield instead of secondary traits (Bernier *et al.*, 2007; Venuprasad *et al.*, 2007; Kumar *et al.*, 2008) [26, 7]. Many QTLs for yield under stress (DTY) have been identified and these QTL possessing lines have been developed through marker assisted backcross selection and were also tested for their drought susceptible index. The relations between the plant yield obtained under conditions of drought and that obtained under conditions of optimal soil moistening were preferred among the field indices of drought tolerance. In wheat, Fisher and Maurer (1978) [2] defined an index namely Susceptibility index based on the relationship of change in relative yield (yield in drought / yield in the absence of drought) of an individual cultivar to the change in mean relative yield, across a range of stress intensities of all cultivars in the comparison. Drought susceptibility indexes (DSI) were calculated in this experiment by determining the changes in grain yield (GY) under two soil moisture (Fischer and Maurer. 1978) [2]. This approach leads to the estimation of genotypic response to stress but it was not independent of yield potential Levels. Selection based on DSI may also lead to the identification of genotypes with high yield in moderate or severe drought stress but not very high yield or yield equivalent to that of current cultivated varieties under normal irrigated situations (Raman *et al.*, 2012) [21]. The correlations between Drought Susceptible Index and Grain Yield confirmed that they are good indicators of drought tolerance in plants (Maciej, *et al.*, 2013).

Material and Methods

The Genetic material used in the study was obtained from the Paddy Breeding Station, and Department of Biotechnology at Centre for Plant Molecular Biology, Tamil Nadu Agricultural

University, Coimbatore. The material for the study consisted of a set of back cross inbred lines of IR64 and ADT45 which were introgressed with QTLs for yield under stress (located on chromosomes 2, 3 and 8), one at a time and combinations of two and three in parental background (Table 1). The QTLs were originally derived from Apo, an indica cultivar. Recombinant inbred lines of IR64 and Apo in F_4 generation with three QTLs for yield under stress in the background donated by Apo were used for backcrossing with IR64 to generate BC_1F_1 and were selfed two generation to obtain BC_1F_3.

Drought susceptibility index (S)

Drought susceptible index (DSI) was calculated for BILs as well as parental lines for their performance of grain yield per plant under severe moisture stress with relation to performance under control conditions. The Drought Susceptibility Index (S) was calculated using the formula suggested by Fischer and Maurer (1978) [2].

$$S = \frac{Y_p - Y_d}{Y_p}$$

Where,
Y_p = Yield with irrigation (Potential yield)
Y_d = Yield with a drought period (Stress yield)

Results and discussion

Drought susceptible index (DSI) was calculated for BILs as well as parental lines for their performance of grain yield per plant under severe moisture stress with relation to performance under control conditions. As expected drought susceptible mega variety IR 64 recorded highest DSI value (0.912) when compared to QTL donor Apo (0.829) which is drought tolerant line where as local check Anna 4 (0.867)was slightly on higher side to Apo. None of the parents and checks had lower index values than QTL possessing BILs. A comparison of minimum values recorded by all single, two and three QTLs of BC_1F_3 of IR 64 is given in table 2. Out of 34 lines possessing single QTL DTY2.2 lines three BILs recorded higher DSI values when compared to IR64, twenty two BILs and twenty seven BILs recorded minimum DSI values when compared QTL donor parent Apo and Anna 4 respectively. BILs with single QTL DTY2.2 on chromosome 2 viz., CB13-900-C-2-23(0.145) and CB13-900-C-2-11 (0.484) recorded the least DSI values compared to other BILs, parents and check and also had higher grain yield of 27g/plant and 22g/plant respectively. Among seven BILs possessing DTY3.1, all of them showed lesser values than IR 64 and Anna 4. Five BILs recorded lesser values than Apo and the least DSI value was recorded by CB13-900-C-3-3(0.643) and all others ranged between 0.765 to 0.886.

Backcross inbred lines containing DTY8.1, CB13-900-C-8-11 (0.957) recorded higher DSI value than IR64, 11 out of 15 had lesser DSI value than Apo and 12 out of 15 had lesser DSI value than Anna 4. CB13-900-C-8-14, CB13-900-C-8-15 and CB13-900-C-8-3 recorded DSI values of 0.562, 0.621 and 0.686 respectively which were the least values among BILs. Two QTL possessing QTLs DTY (2.2+3.1) recorded lesser value compared IR 64 and Anna 4. Except CB13-900-C-23-1 (0.845) all other BILs had lesser DSI value than Apo. Least DSI value of 0.519 was recorded by CB13-900-C-23-3 followed by CB13-900-C-23-3 (0.694) among all the BILs. Among DTY (2.2+8.1) possessing BILs, 2 of them CB13-900-C-28-8 (0.916) and CB13-900-C-28-10 (0.929) had higher values of DSI than IR64. Out of 12, 5 had decreased DSI value than Apo and 3 had lesser than Apo. Minimum value of all BILs and parents was recorded by CB13-900-C-28-5 (0.442) and CB13-900-C-28-12 (0.677). All the BILs with three QTL DTY (2.2+ 3.1+8.1) had lesser DSI value when compared to IR 64, Apo and Anna 4 except CB13-900-C-238-1 (0.833). Minimum DSI value recorded was 0.521 by CB13-900-C-238-3 followed by CB13-900-C-238-2 with 0.576. Lower the DSI, higher the capacity of the genotype to withstand drought. The BILs showing lower values of DSI indicated that handful of lines have been genetically improved for drought resistance as it is reported that drought response index (DSI) is having significant positive correlation with grain yield and harvest index under stress condition (Bidinger *et al.*, 1987a; Pantuwan *et al.*, 2002a, Subashri *et al.*, 2008 and Sellammal, 2009) [22]. For areas where severe stress is a recurrent phenomenon, selection of genotypes with high DSI can be useful. However, selection based on DSI may also lead to the identification of genotypes with high yield in moderate or severe drought stress but not very high yield or yield equivalent to that of current cultivated varieties under normal irrigated situations (Raman *et al.*, 2012) [21].

Table 1: Primer used for marker assisted selection in the experiment

QTL	Chromosome	Position	Primer	Sequence
DTY 2.2	2	8.9 Mb	RM71	‘CTAGAGGCGAAAACGAGATG’ ‘GGGTGGGCGAGGTAATAATG’
DTY3.1	3	30.2 Mb	RM520	‘AGGAGCAAGAAAAGTTCCCC’ ‘GCCAATGTGTGACGCAATAG’
DTY 8.1	8	24.2 Mb	RM256	‘GACAGGGAGTGATTGAAGGC’ ‘GTTGATTTCGCCAAGGGC

Table 2: Drought Susceptibility Index of parents along with BILs.

Genotypes	Y/P(C)	Y/P(s)	DSI	Genotypes	Y/P(C)	Y/P(s)	DSI
IR64 (p)	39.58	3.485	0.912	**DTY3.1**			
APO (p)	48.48	7.5	0.829	CB13-900-C-3-1	33.06	5.93	0.821
ANNA 4 (c)	47.87	5.57	0.867	CB13-900-C-3-2	35.39	8.315	0.765
DTY 2.2				CB13-900-C-3-3	38.71	13.82	0.643
CB13-900-C-2-1	42.25	7.845	0.814	CB13-900-C-3-5	51.53	6.88	0.866
CB13-900-C-2-2	40.75	17.13	0.58	CB13-900-C-3-7	47.51	11.58	0.756
CB13-900-C-2-4	35.75	8.05	0.775	CB13-900-C-3-8	41	7	0.829
CB13-900-C-2-5	41.19	13.09	0.682	CB13-900-C-3-9	46.07	8.68	0.812
CB13-900-C-2-6	47.77	12.03	0.748	**DTY8.1**			

CB13-900-C-2-7	66.06	10.56	0.84
CB13-900-C-2-8	41.27	6.94	0.832
CB13-900-C-2-9	38.86	8.9	0.771
CB13-900-C-2-10	46.1	8.93	0.806
CB13-900-C-2-11	43.17	22.29	0.484
CB13-900-C-2-12	46.48	7.84	0.831
CB13-900-C-2-13	34.17	9.45	0.723
CB13-900-C-2-14	45.2	4.255	0.906
CB13-900-C-2-15	43.53	4.36	0.9
CB13-900-C-2-16	73.66	6.14	0.917
CB13-900-C-2-17	65.45	13.46	0.794
CB13-900-C-2-18	58.05	13.19	0.773
CB13-900-C-2-19	83.18	9.375	0.887
CB13-900-C-2-20	103.03	7.875	0.924
CB13-900-C-2-22	39.23	15.35	0.609
CB13-900-C-2-23	31.98	27.35	0.145
CB13-900-C-2-24	47.2	7.755	0.836
CB13-900-C-2-25	32.88	13.77	0.581
CB13-900-C-2-26	30.39	3.745	0.877
CB13-900-C-2-27	76.45	1	0.987
CB13-900-C-2-29	37.07	11.01	0.703
CB13-900-C-2-30	40.26	11.71	0.709
CB13-900-C-2-31	41.77	14.28	0.658
CB13-900-C-2-32	41.47	12.34	0.702
CB13-900-C-2-33	38.45	9.35	0.757
CB13-900-C-2-34	41.75	6.6	0.842
CB13-900-C-2-35	43.85	10.21	0.767
CB13-900-C-2-36	36.14	8.41	0.767
CB13-900-C-2-37	72.4	9.865	0.864
CB13-900-C-8-1	45.73	5.63	0.877
CB13-900-C-8-3	34.78	10.92	0.686
CB13-900-C-8-4	30.87	5.045	0.837
CB13-900-C-8-5	39.67	13.42	0.662
CB13-900-C-8-6	29.94	10.92	0.635
CB13-900-C-8-7	35.98	10.6	0.705
CB13-900-C-8-8	39.25	9.12	0.768
CB13-900-C-8-9	39.69	8.52	0.785
CB13-900-C-8-11	126.2	5.445	0.957
CB13-900-C-8-14	28.09	12.32	0.562
CB13-900-C-8-15	34.39	13.04	0.621
CB13-900-C-8-16	37.39	8.58	0.77
CB13-900-C-8-17	35.97	8.71	0.758
CB13-900-C-8-18	30.77	9.565	0.689
CB13-900-C-8-19	39.97	3.07	0.923
DTY(2.2+3.1)			
CB13-900-C-23-1	25.57	3.955	0.845
CB13-900-C-23-2	35.06	10.44	0.702
CB13-900-C-23-3	34.94	16.79	0.519
CB13-900-C-23-4	35.56	10.88	0.694
CB13-900-C-23-5	37.81	7.22	0.809
CB13-900-C-23-6	37.11	8.8	0.763
CB13-900-C-23-7	35.05	9.9	0.718
CB13-900-C-23-8	35.11	6.14	0.825

Genotypes	Y/P(C)	Y/P(s)	DSI
DTY (2.2+8.1)			
CB13-900-C-28-1	45.76	9.645	0.789
CB13-900-C-28-2	29.47	5.82	0.802
CB13-900-C-28-5	36.89	20.57	0.442
CB13-900-C-28-7	43.64	7.255	0.834
CB13-900-C-28-8	62.96	5.305	0.916
CB13-900-C-28-9	46.78	10.93	0.766
CB13-900-C-28-10	37.62	2.685	0.929
CB13-900-C-28-11	36.26	8.655	0.761
CB13-900-C-28-12	35.31	11.41	0.677
CB13-900-C-28-13	33.4	9.71	0.709
CB13-900-C-28-14	37.08	4.895	0.868
CB13-900-C-28-15	35.02	5.635	0.839
DTY (2.2+3.1+8.1)			
CB13-900-C-238-1	35.99	6.01	0.833
CB13-900-C-238-2	37.74	16.02	0.576
CB13-900-C-238-3	31.56	15.12	0.521
CB13-900-C-238-4	44.43	6.71	0.849
CB13-900-C-238-5	31.68	7.505	0.763

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