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Effect of Introgressed QTLs of Drought Tolerance in Genetic Background of IR 64 (*Oryza sativa* L.) under Contrasting Water Regiems

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ABSTRACT: *Drought is a major production constraint in rain-fed upland and rainfed lowland ecosystems of rice cultivation. The ascending global shortage of water is a great hindrance for rice production as rice requires high amount of water. Recent studies at IRRI, Philippines have shown moderate to high heritability of grain yield under drought thus opening avenue for direct selection for grain yield instead of secondary traits. Set of backcross inbred lines harboring mega effect QTLs controlling grain yield under stress were developed in the genetic background of IR 64 through Marker Assisted Selection and evaluated for reproductive stage drought tolerance under rain out shelter. Significant differences were revealed by't' test carried out for the mean values among BILs and parents for yield and its contributing traits between severe moisture stress and well irrigated conditions. The BIL harboring QTLs,* DTY3.1 *and* DTY *(3.1+8.1) showed zero differences for days to 50 % flowering and 3 QTL line with* DTY *(2.2+3.1+8.1) showed minimum difference among other QTLs BILs when performances were compared under both water regimes. Over all performances of BILs harboring yield under stress QTLs were found to be superior over susceptible parent IR 64 under both control and severe moisture stress conditions.*

Key words: *Drought resistance, Yield under stress QTLs, Marker Assisted Selection,*

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. It has been suggested that half of the low land rainfed rice area [23% of world's rice area (David, 1991) is prone to drought (Ingram *et al.*, 1990) 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. Recent studies at IRRI have shown moderate to high heritability for 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). By employing direct selection for grain yield under drought, several promising breeding lines for rainfed lowlands and rainfed uplands have been identified recently (Verulkar *et al.*, 2010; Mandal *et al.*, 2010). In the present study conducted at Paddy Breeding Station, Tamil Nadu Agricultural University, Coimbatore, mega variety IR 64 was introgressed with mega effect QTLs controlling drought tolerance from the upland rice variety, Apo from Philippines. Set of 81 backcross inbred lines comprising of BC_1F_3 of IR 64 were evaluated for reproductive stage drought response under severe moisture stress and well irrigated condition.

MATERIALS AND METHODS

The experiment that was carried out in Paddy Breeding Station, Centre for Plant Breeding and genetics, TNAU, Coimbatore during *Kharif* 2013 under rain out shelter to impose stress experiment (withholding irrigation) without rain intervention. The material for the study consisted of a set of back cross inbred lines of IR64 which were introgressed with QTLs for yield under stress (located on

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chromosomes 2, 3 and 8), one at a time and combinations of two and three in parental background. The QTLs were originally derived from Apo, an Indica cultivar. F_1s of the cross IR64 and Apo was selfed till F_4 which were phenotyped under severe stress condition at reproductive stage to identify genotype performing with good seed set and grain yield and screened through MAS to derive QTL (yield under stress) positive lines. Recombinant inbred lines of IR64 and Apo in F_4 generation with three QTLs for yield under stress, donated by Apo were used for backcrossing with IR64 to generate BC_1F_1 and were selfed for 2 generations to obtain BC_1F_3 and were evaluated along with parents. Field experiments were carried out in two replications for both control and drought stress conditions. Recommended crop production and protection practices were followed to raise a healthy crop. Regular irrigation was given until 67 days of crop from date of sowing; later crop was denied irrigation for the rest of its duration. Crop was harvested when the grains reached physiological maturity stage in both the experiments. Z statistics were calculated for equality of two means for QTL classes which had more number of individual BILs by using the formula given below;

$$Z = \frac{\overline{x_1} - \overline{x_2}}{\sqrt{s^2\left(\frac{1}{n_1} + \frac{1}{n_2}\right)}} \sim N(0,1)$$

Where s^2 = pooled variance or combined variance

$$s^2 = \frac{n_1 s_1^2 + n_2 s_2^2}{n_1 + n_2}$$

't'-Test was carried out to comparison of two Sample Means for QTL classes with lesser number of individual BILs. 't' statistics were calculated by using following formula given below

$$S^2 = \frac{n_1 S_1^2 + n_2 S_2^2}{n_1 + n_2 - 2}$$

RESULTS AND DISCUSSION

'z' test was performed for the same set of genotypes with single QTL *DTY*2.2 raised in control and severe stress condition. 't' test was performed for the same set of BILs with single QTL *DTY*3.1, two QTLs combinations *DTY*8.1, *DTY* (2.2+3.1), *DTY* (2.2+8.1) and 3 QTL combination *DTY* (2.2+3.1+8.1) raised in control and severe stress condition and the 'z' and 't' statistic are presented in table 1. Significant differences were noticed for all the characters except for days to 50 % flowering for the BILs with single QTL *DTY* 3.1, *DTY* 8.1 and 2 QTL combination line *DTY* (2.2+3.1) and 3 QTL line *DTY* (2.2+3.1+8.1) which signifies effect of the QTL under stress conditions. Similar results are reported for same QTL that *DTY*3.1 significantly affected days to flowering, and plant height in all the aerobic trials and in lowland severe stress trial. *DTY*2.1 significantly affected plant height in all the aerobic and lowland stress trials but effects on days to flowering were not consistent (Venuprasad *et al.*, 2009). Contrastingly plant height of all class of QTL has significant differences in stress and control conditions. Three QTL line *DTY* (2.2+3.1+8.1) had no significant difference noticed for 1000gw in stress and control conditions. Percent gain in control trial over severe stress conditions for several characters like 1000gw, panicle length; days to 50% flowering and spikelet fertility of *DTY*2.2 were low over stress conditions. But minimum gain over stress for grain yield was recorded by 3 QTL line, *DTY* (2.2+3.1+8.1). Percent gain in control trial over severe stress conditions are presented in table 2. There was up to 80 % yield reduction in severe moisture stress which initiated before flowering as expected based on reports regarding yield reduction in severe moisture stress in earlier works (Pantuwan *et al.*, 2002a; Jongdee *et al.*, 2002; Boonjung and Fukai, 1996; Atlin *et al.*, 2004; Venuprasad *et al.*, 2007). Days to 50% flowering, 1000 grain weight and spikelet fertility suffered minimum losses or zero loss over control conditions by BILs, but overall performances of these QTLs was advantageous over mere susceptible recurrent parent IR 64. Results are available that says Spikelet fertility is positively correlated with 1000gw and were reported by earlier reports as well (Bidinger *et al.*, 1999; Pantuwan *et al.*, 2002; Lafitte *et al.*, 2004; Lanceras *et al.*, 2004). Garrity and O'Toole (1994) reported that, there is negative correla-tion between panicle sterility percentage and single plant yield under drought stress conditions. Here in our study QTLs introgressed thus improve the performances of IR 64 BILs by improving yield contributing traits like 1000 grain weight and spikelet fertility which are major traits which get affected by severe stress. Steele *et al.* (2007) also reported significantly improved grain and fodder yields in the recipient parent, Kalinga III upon introgression with single root QTL located on chromosome 9. Similar results of differential effects of inrtogressing single-root QTLs on Kalinga III have been reported by Shashidhar *et al, (*2005) and in IR 64/Azucena derived NILs by Priyadarshini (2009)

Table 1
'Z' Test Comparison of Single QTL (*DTY*2.2) Lines Raised in Stress and Control Conditions

	DTY2.2		*DTY3.1*		*DTY8.1*		*DTY 2.2+3.1*		*DTY 2.2+8.1*		*DTY2.2+3.1+8.1*	
Traits	*'z'*	*P two-tail*	*'t' Stat*	*P two-tail*	*'t' Stat*	*P two-tail*	*'t' Stat*	*P two-tail*	*'t' Stat*	*P two-tail*	*'t' Stat*	*P two-tail*
VG	2.643	0.008	-2.931	0.026	-2.624	0.020	-1.357	0.217	-5.745	0.000	-1.809	0.145
DFF	2.803	0.005	-0.397	0.705	1.818	0.091	0.000	1.000	2.038	0.066	1.543	0.198
PH	7.612	0.000	4.664	0.003	8.457	0.000	7.139	0.000	12.06	0.000	5.832	0.004
NOP	8.960	0.000	6.954	0.000	5.875	0.000	6.709	0.000	5.047	0.000	2.693	0.054
PL	4.170	0.000	3.209	0.018	3.326	0.005	2.667	0.032	3.788	0.003	3.661	0.022
1000GW	2.473	0.013	1.987	0.094	1.758	0.101	2.712	0.030	2.620	0.024	0.051	0.962
SF	5.850	0.000	3.179	0.019	5.436	0.000	3.492	0.010	4.133	0.002	1.328	0.255
TDM	10.06	0.000	7.121	0.000	6.283	0.000	12.28	0.000	11.19	0.000	7.930	0.001
HI	8.348	0.000	5.404	0.002	5.469	0.000	3.310	0.013	7.041	0.000	2.101	0.103
PY	27.48	0.000	5.922	0.001	19.45	0.000	19.87	0.000	12.10	0.000	4.636	0.010
Y/P	12.74	0.000	12.36	0.000	5.076	0.000	17.54	0.000	10.48	0.000	7.128	0.002

Table 2
Percent Gain of BILs Raised in Control over Stress Conditions

Traits	*DTY2.2*	*DTY3.1*	*DTY8.1*	*DTY 2.2+3.1*	*DTY 2.2+8.1*	*DTY2.2+3.1+8.1*
VG	-23.93	-72.22	-46.34	-20.83	-72.41	-46.15
DFF	2.351	-0.781	1.338	0.000	2.233	2.668
PH	21.71	25.90	24.03	24.27	27.67	20.73
NOP	43.75	46.16	26.48	40.93	40.48	29.26
PL	8.126	11.95	7.956	8.930	10.93	6.083
1000GW	6.368	16.00	6.216	10.74	9.472	0.338
SF	16.50	13.67	17.21	10.48	26.43	8.040
TDM	65.07	62.99	62.70	57.65	57.75	52.33
HI	40.13	43.36	40.88	34.03	50.15	34.52
PY	89.73	88.79	91.28	89.30	93.19	84.04
Y/P	78.48	78.79	78.54	73.17	78.65	71.69

and Mahesh (2007). Babu *et al*. (2003) reported pleiotropic effects of QTLs conferring drought tolerance on grain yield and their contributing traits under moisture stress.This holds the promise of betterment of existing drought susceptible IR 64 for grain yield performances under stress and also spikelet fertility, harvest index which were major reasons for better grain yield in stress condition.

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