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Genetics of marker assisted backcross progenies of the cross HUR-105 X Swarna-SUB1

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Abstract

The Marker Assisted backcross progenies of the cross HUR-105 x Swarna-Sub1 along with their parental lines were evaluated under submergence condition for 14 days at 35 days after sowing. Total 53 BC_2F_1 progenies were subjected to complete submergence out of which twenty lines recovered after desubmergence. The survival percentages of BC_2F_1 plants was observed to be 47.62 per cent. Mean performance for BC_2F_1 generation in submerged condition was found lower for most of the yield traits *viz.*, productive tiller per plants, length of panicle, weight of panicle, spikelet per panicle, test weight, yield per plant, in comparison to all six generations in normal irrigated condition. Amylose content and gel consistency was found comparable to normal irrigated condition for segregating generations. Under submerged condition BC_2F_1 generation showed reduced height and medium to late maturity. The yield of the BC_2F_1 plants under submergence varied from 4.64 g to 14.32 g per plant. On the basis of field test under submergence condition the four lines namely HUR-105-Sub1-6, HUR-105-Sub1-23, HUR-105-Sub1-25 and HUR-105-Sub1-28 were found better in respect to grain yield and other related traits. The level of submergence tolerance in backcross progenies were similar as tolerant parent Swarna Sub-1, revealed that submergence tolerance in HUR 105-Sub-1 is governed by single dominant gene.

Highlights

- Twenty BC_2F_1 plants were recovered after complete submergence of 14 days.
- The backcross progenies (BC_2F_1) were found similar pattern of tolerant as of tolerant parent Swarna Sub-1.
- The chi-square value of B_1 and BC_2F_1 showed the non-significant at 5 per cent level of significance indicating that observed data are in accordance with expected ratio and follow mendelian pattern of inheritance to submergence tolerance.

Keywords: Backcross progenies, submergence, sub1 gene, survival percentage, yield per plant, *oryza sativa*

Rice is the staple food for about half of the world's population and 90% of it is being produced and consumed in Asia. Rice is mainly grown in large areas of Asia, Latin America and Africa that are characterized by a semitropical climate with alternating rainy and dry seasons. The erratic occurrence of biotic and abiotic stresses leads to low and unstable yields in rice. Among the several abiotic stresses affecting rice production, submergence has been identified as the third important constraint for higher rice productivity (Sarkar *et al.* 2006). Flooding is a serious constraint to rice plant growth and survival in rainfed lowland and deep water areas because it results in partial or complete submergence of the plant. About 32.4 per cent of India's total rice area is under rainfed lowland conditions, which are highly fragile ecosystem, always prone to flash floods (Singh *et al.* 2015). Flooding imposes a severe selection pressure on plants principally because excess water in their surroundings deprive them of certain basic needs, like oxygen, carbon dioxide and light for photosynthesis. Flooding leads to a complex abiotic stress as it substantially reduces crop stand, especially if it occurs during early vegetative stage and prolongs for more than a week (Bailey-Serres *et al.* 2010). However, the flood-prone ecosystems have enormous potential for food production to meet the ever increasing demand of rice because of the predominance of good soils and fresh water resources (Ismail *et al.* 2013).

The *indica* rice cultivar namely FR13A was identified from the flood prone areas of coastal belt of Orissa. It is pure line selection of the local lowland rice variety *Dhala Putia*. This cultivar is highly tolerant and survive up to two weeks of complete submergence owing to a major quantitative trait locus designated as Submergence 1 (*Sub1*). Xu and Mackill (1996) identified that this major QTL (*Sub1*) on rice chromosome 9, contributed about 70% of the phenotypic variation for submergence tolerance. This QTL enables the rice plant to survive and more importantly recover after flooding. The discovery and cloning of the *Sub1A* gene facilitated its introgression through MABC to several high yielding varieties within a short span of 2-3 years (Bailey-Serres *et al*. 2010; Collard *et al*. 2013; Mackill *et al*. 2012). The varieties with *Sub1A* gene showed a yield advantage of 1 to 1.3 t/ha over the original varieties following the submergence for up to 18 days (Mackill *et al*. 2012; Neeraja *et al*. 2007; Septiningsih *et al*. 2009; Singh *et al*. 2009; Singh *et al*. 2013). Gene expression studies in *indica* type varieties with Sub1A gene revealed that submergence tolerance is correlated with the presence of Sub1A-1 allele and intolerance is associated with the presence of Sub1A-2 allele or complete absence of this gene. While the Sub1 gene has already been transferred to several popular rice varieties, including Swarna, IR 64, Samba Mahsuri, BR 11 and CR 1009 (Singh *et al.* 2015). It was estimated that Swarna-Sub1 offers an approximate 45% increase in yields over the current popular variety when fields are submerged for 10 days (Dar *et al.* 2013).

The transfer of *Sub1* gene in the cultivars of flash flood prone areas has helped to increase the productivity of rice in such areas. Development of submergence tolerant varieties is generally considered as the most effective way for improving productivity of rice varieties damaged from flash flood. Moreover, the adoption of completely new varieties could take considerable time, whereas the chances of rapid adoption of popular varieties converted through marker assisted backcrossing (MABC) are relatively higher (Septiningsih *et al.* 2009). Genetics research with segregating populations of 15-50 days old seedlings demonstrated (a) there is one dominant gene for submergence tolerance and (b) this gene is present in three out of four of the world's most tolerant rice cultivars. This suggests that a common factor related to tolerance may be responsible for genotypic differences in submergence tolerance of rice (Setter *et al.* 1997). In present investigation, the backcross progenies of cross HUR-105 x Swarna Sub1 were obtained and the yield attributes after fourteen days of submergence were observed in the backcross progeny. There are some traditional cultivars that can survive complete submergence for about 12 days. Since the duration of flash floods rarely exceeds 10 days, yield improvement of these traditional cultivars or the transfer of their submergence tolerance to high-yielding varieties would benefit rainfed lowland rice farmers (Mishra *et al.* 1995).

Materials and Methods

Plant material & experimental plan

The two indica rice varieties HUR-105 (Malviya Dhan-105) and Swarna Sub-1 were taken for the present study in which HUR-105 is most widely grown in North East India owing to its high yielding, semi dwarf, medium duration, with acceptable grain quality but submergence intolerant and Swarna Sub-1 is submergence tolerance variety. The crosses were made in *Kharif*-2009 between HUR-105 × Swarna Sub-1 to raise the F_1. In the next, off season at ICAR-NRRI, Cuttack the F_1 were backcrossed with recurrent parent (P_1) and donor parent (P_2) to generate B_1 and B_2 population, respectively. In *Kharif* -2010 the BC_1 plants were backcrossed to have the BC_2 plants. Six generations, namely, P_1, P_2, F_1, F_2, BC_1 (B_1) and B_2 along with BC_2 were raised in a complete randomized block design in both normal irrigated and submerged condition, with three replications at Agricultural Research Farm, Institute of Agricultural Sciences, Banaras Hindu University, Varanasi, India during *Kharif*-2011. Submergence screening was performed in the controlled condition. BC_2F_1's plants along with P_1, P_2, F_1, F_2, BC_1 (B_1) and B_2 and susceptible check (HUR-105) and resistance check (Swarna Sub-1) were planted. Twenty-one days old seedlings were transplanted in separate plot size 3 × 1 m with spacing 20 × 15 cm apart. The recommended packages of practices were followed to raise healthy crops.

Submergence Screening

The plants were submerged when the plants are 35 days old after transplanting of 21 days old seedlings for 14 days of complete submergence up to 50 cm depth. The plants were de-submerged and the survival of plants was scored after 10 to 21 days on fixed intervals for recovery (Pamplona *et al.* 2007). Scoring of survival plant were done as given procedure as per entry record the number of seedling used (X) and also the number of survivors 21 days after desubmergence (Y). Survivors should be produced at least one green leaf.

Percentage survival = (Y/X) × 100

Observation recorded

The phenotypic traits were assessed on randomly selected plants from each individual entry in the segregating generations and observations were recorded for eleven quantitative traits *viz.*, number of productive tillers per plant (PTP), plant height (PH), days to 50 per cent flowering (DF), days to maturity (DM), length of panicle (LP), weight of panicle (WP), spikelet per panicle (SSP), test weight (TW), grain yield per plant (YPP), amylose content, gel consistency. Ten plants from both parents, 20 plants from F_1, B_1 & B_2 and 40 plants from F_2 generation per replication were randomly selected and tagged for observing the traits under normal irrigated and stress condition.

Statistical analysis

The mean value of each character was determined by summing up all the observations and dividing them by corresponding number of observations.

$$\overline{X} = \frac{\sum_{i=1}^{N} X_i}{N}$$

Where, numerator shows sum of all observations for the character and N = Number of observations.

The frequency of tolerance and susceptible plants in segregating population for submergence was grouped based on survival of plants after de-submergence of plants. The plants survive after 14 days of submergence were recorded as tolerant and then number of plant dies after submergence will account for susceptible. These frequencies were tested using χ^2 test with expected frequencies of tolerant and susceptible plants. The formula for calculation of χ^2 test values is as follow:

$$\chi^2 = \sum (O-E)^2 / E$$

Where,

χ^2 = Chi square value

O = Observed frequency

E = Expected frequency

The value of χ^2 was taken at 5% and / or 1% level of significance for (n–1) degree of freedom (if n number of classes). The non-significant χ^2 value is the indication of goodness of fit of the observed frequencies into expected frequencies. Now, as per goodness of fit to expected frequencies the number of genes could be estimated governing the submergence.

Results and Discussion

Total 53 BC_2F_1 plants of the cross HUR-105 × Swarna-Sub1 along with parental lines were subjected to submergence after 35 days of sowing for 14 days. The survival percentage of plants were estimated after 20 days of desubmergence. Total 20 plants were recovered after desubmergence and subjected to yield trait data.

Comparison of BC_2F_1 progeny with P_1, P_2, F_1, F_2, BC_1 (B_1) and B_2 under normal irrigated condition

The mean performance for various quantitative and qualitative traits of the six generations i.e. P_1, P_2, F_1, F_2, BC_1 (B_1) and B_2 under normal irrigated condition and of 20 BC_2F_1 recovered plants are presented in Table 1. The performance of these twenty BC_2F_1

plants varied from trait to trait. Productive tiller per plants for BC_2F_1 varied from 2 to 10, while F_1 (19.45), F_2 (16.34) and B_1 (18.99) population recorded high number of tillers per plant while B_2 population (13.78) had lesser number of tillers per plant than both parent HUR 105 (14.23) and Sawarna-Sub1 (16.01), plant height for BC_2F_1 from 70 to 79.50 cm, while in F_1 (94.00 cm) and F_2 (94.67 cm) were slightly shorter than recurrent parent HUR 105 (98.89 cm), B_1 population (98.56 cm) had comparable plant height to recurrent parent HUR 105, and B_2 population (88.56 cm) was found taller than donor parent Sawarna-Sub1 (83.67 cm). Days to 50 per cent flowering and days to maturity ranged from 107 to 131 days and 139 to 176 days, respectively for BC_2F_1, all the population F_1 (101.34 and 132.34 days), F_2 (105.00 and 135.67 days), B_1 (104.34 and 135.34 days), B_2 (84.38 and 115.20 days) flowered and matured intermediate as with the parents, panicle length was found more in F_1, F_2, BC_1 (B_1) and B_2 compare to both the parents, while in BC_2F_1 generation, length of panicle found less than above mentioned generations and ranged from 13 to 21 cm. Weight of panicle for BC_2F_1 ranged from 1.24 to 3.10 gm, and found more in F_1 (4.31 gm) and B_1 (4.11 gm) generations than both the parents HUR 105 (4.01 gm) and Sawarna-Sub1 (3.97 gm), meanwhile F_2 (3.58 gm) and B_2 (3.75 gm) showed less than the parents. Spikelet per panicle for BC_2F_1 varied from 68 to 140, which is less than the value obtained for all generation in normal irrigated condition. F_1, F_2, BC_1 (B_1) and B_2 showed more spikelet per panicle than parents. Test weight for BC_2F_1 was found very less compare to all other generations and ranged from 12 to 14.37. For F_1 (19.54 gm), it was found higher than both the parents HUR 105 (17.19 gm) and Sawarna-Sub1 (18.08 gm) and found comparable for B_1 (17.14 gm) with recurrent parent (17.19 gm), yield per plant from 4.64 to 14.32 found in BC_2F_1 was found less than in normal irrigated condition, while yield of F_2 (20.04 gm) was found comparable with non recurrent parent Sawarna-Sub1 (20.56 gm), F_1, B_1, B_2 showed higher yield 22.82, 23.34, 21.71, respectively than both the parents HUR 105 (18.87 gm) and Sawarna-Sub1 (20.04 gm), amylose content and gel consistency ranged from 22 to 28 from 68 to 79 respectively which was found higher than the values found under normal irrigated condition for all the six generations.

Mean performance for BC_2F_1 generation in submerged condition was found lower for most of the yield traits *viz.*, productive tiller per plants, length of panicle, weight of panicle, spikelet per panicle, test weight, yield per plant, in comparison to all six generations in normal irrigated condition. Amylose content and gel consistency was found comparable or little high than obtain in normal irrigated condition for segregating generations. Under submerged condition BC_2F_1 generation showed reduced height and medium to late maturity.

Survival of BC_2F_1 progeny under submergence and its genetics

The survival percentage for donor and recurrent parents were recorded 98 per cent and 20 per cent, respectively. The survival percent of donor parent Swarna Sub-1 is significantly more than that of recurrent parent HUR-105. The results showed that most of the plants in the F_1 were showed tolerant to submergence with 88.47 per cent of plant survival,

Table 1: Mean performance of traits in different generations under normal irrigated condition

Traits	Productive tillers per plant	Plant height	Days to 50% flowering	Days to maturity	Length of panicle	Weight of panicle	Spikelet per panicle	Test weight	Yield per plant	Amylose content	Gel consistency
P_1 ±SEm	14.23	98.89	97.4	130.78	21.67	4.01	141.23	17.19	18.87	23.55	81.00
P_2 ±SEm	16.01	83.67	103.00	143.0	21.23	3.97	150.78	18.08	20. 04	25.79	74.67
F_1 ±SEm	19.45	94.00	101.34	132.34	23.89	4.31	168.22	19.54	22.82	24.57	81.17
F_2 ±SEm	16.34	94.67	105.00	135.67	23.93	3.58	165.78	18.94	20.56	23.56	77.34
B_1 ±SEm	18.99	98.56	104.34	135.34	24.23	4.11	159.23	17.14	23.34	23.89	78.67
B_2 ±SEm	13.78	88.56	104.67	133.34	24.12	3.75	161.78	18.47	21.71	24.48	74.50

** and *: Significant at 1 and 5 per cent level, respectively. ***: Significant at 0.01.

Table 2. Mean performance of yield traits of 20 BC_2F_1 plants under submergence condition

Trait	Productive tillers per plant	Plant height	Days to 50% flowering	Days to maturity	Length of panicle	Weight of panicle	Spikelet per panicle	Test weight	Yield per plant	Amylose content	Gel consistency	Productivity t/ha (calculated)
HUR-105 (recurrent parent)	**4.95**	**82.45**	**112.00**	**141.34**	**18.12**	**2.29**	**117.99**	**11.32**	**8.73**	**22.05**	**79.4**	**few plants survived only**
Swarna Sub-1 (donor parent)	**8.73**	**67.56**	**123.00**	**158.67**	**20.89**	**2.61**	**135.89**	**14.05**	**12.59**	**26.41**	**72.16**	**3.41**
1 HUR 105-Sub1-3	6	75	115	140	20	1.49	95	13.3	11.19	27	72	3.17
2 HUR 105-Sub1-5	6	70	111	143	19	2.31	102	13.33	12.87	25	76	3.64
3 HUR 105-Sub1-6	8	72.8	112	145	21	2.79	135	14.37	14.97	24	75	4.24
4 HUR 105-Sub1-9	7	78	128	160	16	1.24	100	13.39	9.08	28	71	2.57
5 HUR 105-Sub1-10	9	74	115	143	19	3.1	115	13.58	12.25	23	75	3.47
6 HUR 105-Sub1-11	4	76.5	131	167	13	2.19	68	13.42	5.11	27	68	1.44
7 HUR 105-Sub1-13	3	77	130	166	18	2.3	99	13.25	4.64	28	72	1.31
8 HUR 105-Sub1-16	2	73	130	176	20	2.9	110	13.35	2.78	26	72	0.78
9 HUR 105-Sub1-17	8	76	113	144	21	2.5	130	13.5	12.11	22	77	3.43
10 HUR 105-Sub1-18	8	70	125	153	16	1.9	96	12.5	9.64	24	74	2.73
11 HUR 105-Sub1-20	5	71.5	109	140	16.5	1.98	115	13.8	6.5	26	69	1.84
12 HUR 105-Sub1-21	6	76	109	139	17	1.8	92	12	6.1	24	72	1.72
13 HUR 105-Sub1-23	9	76	109	141	20.5	2.75	138	14.2	14.25	26	71	4.03
14 HUR 105-Sub1-24	7	76.3	127	158	17	1.35	126	12.95	10.2	23	76	2.88
15 HUR 105-Sub1-25	10	78	111	143	19	2.4	135	14	14.13	24	76	4.01
16 HUR 105-Sub1-28	9	76	113	143	21	2.5	140	13.98	14.32	24	75	4.06
17 HUR 105-Sub1-31	7	79.5	124	151	16	2.1	100	12.68	7.2	22	79	2.03
18 HUR 105-Sub1-35	9	74	107	139	17	2.05	133	13.4	12.2	27	73	3.45
19 HUR 105-Sub1-39	6	79	113	145	17	1.9	115	12.7	6.13	24	74	1.73
20 HUR 105-Sub1-41	6	76	108	140	16.6	1.98	106	12.78	6.5	23	71	1.84

and in F_2 population it showed only two class as tolerant or susceptible in the ratio of 3T:1S which is mainly due to presence of dominant effects of tolerance gene Sub1. The chi-square value for F_2 generation was 1.5, which is non-significant at 5% level of significance indicating that observed data are in accordance with expected ratio which showed the dominance pattern of Mendelian ratio of inheritance in the F_2 populations. The B_1 populations were classified into tolerance and susceptible in the ratio of 1T:1S and in B_2 population only tolerant plant were obtained. The survival percentage and chi-square value for B_1 populations were 41.76 per cent and 0.33 per cent respectively. In BC_2F_1 populations were again classified into tolerance and susceptible in the ratio of 1:1 and chi-square and survival percentage was 0.05 and 47.62 per cent respectively. The chi-square value of B_1 and BC_2F_1 showed the non-significant at 5 per cent level of significance indicating that observed data are in accordance with expected ratio and follow mendelian pattern of inheritance to submergence tolerance. The survival of 47.62 per cent for BC_2F_1 population, which is close to expected survival percentage i.e. 50 per cent as a backcross progeny ratio. Setter *et al.* (1997) also observed the similar trend in his experiments. These results were also correlated with the results obtained by foreground selection using Sub_1BC_2 marker where out of 53 plants 20 plants showed positive bands which were responsible for tolerance against submergence.

Table 3: Reaction of tolerant and susceptible rice parents and their progenies to submergence

Treatment	Total number of plants transplanted	Plants survived	% survival (Observed)	expected Plant survival	X^2 cal.	X^2 tab (0.05)	D.F.	Mendelian data ratio
HUR 105 (susceptible)	50	10	20	0	--	--	--	--
Swarna Sub1 (tolerant)	50	49	98	50	--	--	--	all
F_1	26	23	88.47	26	0.35	--	--	all
F_2	50	30	60	37.5	1.5	3.841	1	3:1
BC_1	24	10	41.67	12	0.33	3.841	1	1:1
B_2	26	24	92.3	26	0.16	--	--	all
BC_2	42	20	47.62	21	0.05	3.85	1	1:1

Best identified BC_2F_1 progenies on the basis of phenotypic data

Background selection for these 20 BC_2F_1 plants showed that four lines (HUR-105-6, HUR-105-23, HUR-105-25 and HUR-105-28) showed maximum recovery percentage of recurrent parent genome and these results were also reflected in phenotypic performance for various traits. These four line of BC_2F_1 showed good phenotypic performance and on the basis of phenotypic performance for yield and other traits these four lines namely HUR-105-Sub1-6, HUR-105-Sub1-23, HUR-105-Sub1-25 and HUR-105-Sub1-28 were identified as the best progeny. The maturity duration in days for HUR-105-Sub1-6, HUR-105-Sub1-23, HUR-105-Sub1-25 and HUR-105-Sub1-28 were found to be 145, 141, 143 and 143, respectively. However, the maturity duration of 141.34 and 158.6 days under submergence were observed for HUR-105 and Swarna-Sub1, respectively, which is lower than donor parent Swarna-Sub1. The yield per plant (gm) under submerged condition for the selected lines *viz.*, HUR-105-Sub1-6, HUR-105-Sub1-23, HUR-105-Sub1-25 and HUR-105-Sub1-28 were found to be 14.97, 14.25, 14.13 and 14.32, respectively, higher than both the parents HUR-105 and Swarna-Sub1, 8.73 and 12.59, respectively under submergence. Dar *et al.* (2013) observed that Swarna-Sub1 offers 45% increase in yield over the existing popular varieties when fields are submerged for 10 days. Ismail *et al.* (2009) reported high yield and better adaption in the varieties with submergence tolerance gene (*Sub1* gene). Mackill *et al.* (1993) reported some submergence tolerant lines with higher yield and better adaption.

Conclusion

χ^2 test were performed to know the genetics of the submergence tolerance gene and observed value for χ^2 test were found in accordance with the expected value. The segregation pattern in segregating generation showed the dominance pattern of mendelian ratio of inheritance. The backcross generation BC_1 and BC_2 were classified into tolerance and susceptible in the ratio of 1T:1S. Segregation patterns of these genotypes support the hypothesis that submergence tolerance in Swarna Sub-1 is controlled by a single dominant gene. These four promising lines namely HUR-105-Sub1-6, HUR-105-Sub1-23, HUR-105-Sub1-25 and HUR-105-Sub1-28 were found better and could be exploited for improvement of grain yield and other related traits.

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References

Bailey-Serres, J., Fukao, T., Ronald, P., Ismail, A.M., Heuer, S. and Mackill, D.J. 2010. Submergence Tolerant Rice: Sub1's Journey from Landrace to Modern Cultivar. *Rice* **3**: 138–147.

Collard, B.C.Y., Septiningsih, E.M., Das, S.R., Carandang, J., Pamplona, A.M., Sanchez, D.L., Kato, Y., Ye, G., Reddy, J.N., Singh, U.S., Iftekharuddaula, K.M., Venuprasad, R., Vera-Cruz, C.N., Mackill, D.J., Ismail, A.M. 2013. Developing new flood-tolerant varieties at the International Rice Research Institute (IRRI). *Sabrao J. Breed. Genetics* **45**: 42-56.

Ismail, A.M. 2013. Flooding and Submergence Tolerance, In: C. Kole, Ed., *Genomics and Breeding for Climate-Resi- lient Crops*, Springer-Verlag, Berlin, **2**: 269-290.

Ismail, A.M., Ella, E.S., Vergara, G.V., Mackill, D.J. 2009. Mechanisms associated with tolerance to flooding during germination and early seedling growth in rice (*Oryza sativa* L.). *Annals of Botany* **103**: 197-209.

Mackill, D.J., Ismail, A.M., Singh, U.S., Labios, R.V., Paris, T.R. 2012. Development and rapid adoption of submergence-tolerant (*Sub1*) rice varieties. *Advances in Agronomy* **115:** 303-356.

Mackill, D.J., Amante, M.M., Vergara, B.S., Sarkarung, S. 1993. Improved semidwarf rice lines with tolerance to submergence of seedlings. *Crop Science* **33**: 749-753.

Mishra, S.B., Senadhira, D., Manigbas, N.L. 1995. Genetics of submergence tolerance in rice (*Oryza sativa* L.). *Field Crops Research* **46**: 177-181.

Neeraja, C., Maghirang-Rodriguez, R., Pamplona, A.M., Heuer, S., Collard, B., Septiningsih, E., Vergara, G., Sanchez, D., Xu, K., Ismail, A.M., Mackill, D. 2007. A marker-assisted backcross approach for developing submergence-tolerance rice cultivars. *Theor. Appl. Genetics* **115**: 767-776.

Pamplona, A., Ella, E., Singh, S., Vergara, G.V., Ismail, A. and Mackill, D.J. 2007. Screening procedure for tolerance of complete submergence in rice. *Sub1 Rice News* **1**(2).

Sarkar, R.K., Reddy, J.N., Sharma, S.G., Ismail, A.M. 2006. Physiological basis of submergence tolerance in rice and implications for crop improvement. *Current Science* **91**: 899-906.

Septiningsih, E.M., Pamplona, A.M., Sanchez, L.D., Neeraja, C.N., Vergara, G.V., Heuer, S., Ismail, A.M. and Mackill, D.J. 2009. Development of submergence-tolerant rice cultivars: the *Sub1* locus and beyond. *Annals of Botany* **103**(2): 151-160.

Setter, T.L., Ellis, M., Laureles, E.V., Ella, E.S., Senadhira, D., Mishra, S.B., Sarkarung, S. and Datta, D. 1997. Physiology and genetics of submergence tolerance in rice. *Ann. Bot. Supplly* **69**: 67-77.

Singh, A., Mukul, Joshi, M., Mukh Ram, Arya, M. and Singh, P.K. 2015. Screening and evaluation of rice cultivars for submergence tolerance using SSR markers. *The Ecoscan* **9**(1&2): 255-259.

Singh, S., Mackill, D.J., Ismail, A.M. 2009. Responses of *Sub1* rice introgression lines to submergence in the field: yield and grain quality. *Field Crops Research* **113**: 12-23.

Singh, U.S., Dar, M.H., Singh, S., Zaidi, N.W., Bari, M.A., Mackill, D.J., Collard, B.C.Y., Singh, V.N., Singh, J.P., Reddy, J.N., Singh, R.K. and Ismail, A.M. 2013. Field performance, dissemination, impact and tracking of submergence tolerant (*Sub1*) rice varieties in South Asia. *SABRAO J. Breed. Genetics* **45**: 112-131.

Xu, K., Mackill, D.J. 1996. A major locus for submergence tolerance mapped on rice chromosome 9. *Molecular Breeding* **2**: 219-224.

Dar, M.H., De Janvoy, A., Emerick, K., Raitzer, D., Sadoulet, E. 2013. Flood tolerant rice reduces yield variability and raises expected yield, differentially benefitting socially disadvantaged groups. *Scientific Reports* **3**: 3315

Singh, A., Singh, A.K., Shama, P. and Singh, P.K. 2013. Studies on genetic characteristics of upland rice (*Oryza sativa* L.). *International Journal of Agriculture, Environment & Biotechnology* **6**(4): 515-520. http://dx.doi.org/10.5958/j.2230-732x.6.4.025

Singh, A., Saini, R., Singh, J., Arya, M., Mukh Ram, Pallavi, Mukul and Singh, P.K. 2015. *International Journal of Agriculture, Environment & Biotechnology* **8**(1): 143-152. http://dx.doi.org/10.5958/j.2230-732X.2015.00019.4

Singh, R. *et al.* 2016. From QTL to variety-harnessing the benefits of QTLs for drought, flood and salt tolerance in mega rice varieties of India through a multi-institutional network. *Plant Science* **242**: 278-287. http://dx.doi.org/10.1016/j.plantsci.2015.08.008