Asian Jr. of Microbiol. Biotech. Env. Sc. Vol. 24, No. (2) : 2022 : 327-334

ISSN-0972-3005

DOI No.: http://doi.org/10.53550/AJMBES.2022.v24i02.019

EFFECT OF SUBMERGENCE STRESS ON AGRO-MORPHOLOGICAL AND PHYSIOLOGICAL TRAITS IN RICE (*ORYZA SATIVA* L.) GENOTYPES

GOUTAM DAS[1], BANSHIDHAR PRADHAN[1], DEBENDRA NATH BASTIA[1] AND RAMLAKHAN VERMA[2*]

[1]Department of Genetics and Plant Breeding, OUAT, Bhubaneshwar, India.
[2] ICAR-National Rice Research Institute, Cuttack 753 006, India.

(Received 20 December, 2021; Accepted 11 March, 2022)

***Key words**: Submergence stress, Physiological parameters, Morphology, Oryza sativa, Sub1.*

Abstract– Submergence stress due to flash flooding is one of the most adverse abiotic factor of plant growth and productivity, considered a severe threat for sustainable crop production in the changing climate. Rice is semi-aquatic crop, able to make several modulations in their adaptive traits to sustain submergence stress, but intense and prolonged flooding is fatal for its sustainability. In this context, a field experiment was conducted to assess the effect of different submergence regimes (7, 14 and 21 days' submergence) on agro-morphological and physiological traits in rice at ICAR-National Rice Research Institute, Cuttack, India. The experiment was laid out in a Complete Randomized Design with three replications. Control (no submergence) and submergence (7, and 14 days) were imposed on eleven rice genotypes at vegetative stage. Plant height, number of tillers hill^{-1}, spikelets fertility, % survival, chlorophyll content, total soluble sugar content showed decreasing trend under submergence, whereas, accumulation of proline content and ADH activities reported enhanced level under submergence, especially in Sub1 carrying lines. From the current research, it was observed that submergence stress affected plant growth stages substantially, therefore, genotypes showed least vulnerability under seven days' submergence might be utilized in traits improvement program.

INTRODUCTION

Impeding climatic scenario and depleting soil resources threaten rice (*Oryza sativa* L.) crop sustainability throughout the world. Frequent occurrence of submergence, drought and salinity are the most detrimental abiotic stresses for rice production in rainfed ecosystem (Sekar and Pal 2012). Submergence due to flash flooding (FF) is third important abiotic stress in rainfed ecologies, hampers sustainability of crop plants and causes huge yield loss, annually (Iftekharuddaula *et al.*, 2015). In India, ~36% of the rice area are submergence prone, of that ~90% are distributed in eastern states where rice is a major commodity of livelihood (Pradhan *et al.*, 2015). Being semi-aquatic in nature, rice plant are able to modulates in various ways to sustain under submergence (Khush *et al.*, 1997). However, intense and prolonged submergence is fatal for its substantiality (Kumar *et al.*, 2021). Submerged plant canopy faces several challenges like energy shortage, nutrient and oxygen deficiency which affects proper plant growth and survival (Pradhan *et al.*, 2019). Besides, submergence creates very conducive environment for the growth of several biotic stresses like bacterial blight (BB) and sheath blight which makes havoc to the rice sustainability under rainfed shallow-lowland. Changing climate scenario looking very distressing, predicting severe and frequent flooding down the line in coastal regions (Sanchez *et al.*, 2000).

Rice is major staple crop in India, needs further breeding attention step up yield potential in rice by 70% to feed our ever-growing population (Godfray *et al.*, 2010). Frequent outbreak of major biotic stresses and resurgence of minor diseases and pests are others challenges of rice production under RSL (Hasan *et al.*, 2015). In this context, development of climate resilient and multiple disease resistance cultivars found to be substantial to combat rice yield losses (Ronald *et al.*, 1992). Rice plants has inbuilt ability to sustain flooding up to one week (Adkins *et al.*, 1990; Palada *et al.*, 1972,), however, beyond that only tolerant genotypes can survive (Catling *et al.*, 1992). Tolerant plants make several adaptive changes to sustain under hypoxia/anoxia without major damage (Pradhan *et al.*, 2019; Samanta *et al.*, 2020; Sarkar *et al.*, 2014). Tolerant plants are able to manipulate synthesis/secretion of several plant growth regulators like ethylene, gibberellic acid (GA), abscisic acid (ABA) which regulates shoot elongation and leaf senescence, thus, enhances plant survival under flood (Rajpurohit *et al.*, 2011). Tolerant plants are able to switched-on alcohol dehydrogenase (ADH) activity which controls judicious energy consumption and enhances plant survivality under submergence (Kumar *et al.*, 2021).

In rice, genomic blocks regulating adaptive modulations under flooding have been identified, validated and successfully deployed in several suitable genetic background (Singh *et al.*, 2016). The Sub1 gene controlling two weeks' tolerance is mapped on chromosome-9 alongwith other minor QTLs in the Indian rice landrace FR13A (Xu *et al.*, 1996 and 2006). FR13A is utilized as donor of Sub1 in many rice improvement programs (Pradhan *et al.*, 2015 and 2019). Popular rice varieties, Swarna (Neeraja *et al.*, 2007), Ranjit (Chetia *et al.*, 2018), IR64 (Singh *et al.*, 2015), Chehrang (Singh *et al.*, 2015), CO3 (Rahman *et al.*, 2018), CR 1009 (Singh *et al.*, 2015), BRRI dhan52 (Kabir *et al.*, 2017), Aiswarya (Nair *et al.*, 2021) etc. have been pyramided with *Sub1* through MAS/MAB approach. In this context, genomics based improvement of plant resistance/ tolerance has been proven as most promising strategies towards development of sustainable cultivars. Hence, identification of suitable donors having close genetic proximity and duration with recurrent parent is paramount important for precise and fast development of climate resilient cultivars. This study is aimed at assessment of submergence effect on agro-morphological and physiological features in tolerant and intolerant rice genotypes in order to select suitable parent might invigorate the climate resilient rice breeding program.

MATERIALS AND METHODS

Field experiment was carried out with eleven rice genotypes viz., tolerant genotypes- Swarna Sub1, CRMS 31B, CRMS 32B, Chehrang Sub1, CR Dhan 801, and intolerant genotypes- Mrunalini, Hasanta, Upahar, Swarna Shreya, IRBB66 (carrying five resistant genes for bacterial blight, Xa21, xa13, Xa7, xa5 and Xa4) and CRL 22R at ICAR-National Rice Research Institute, Cuttack, India. The experiment was laid out in a Complete Randomized Design (CRD) with three replications. Submergence screening of 11 rice genotypes was performed under standard evaluation system (SES) (IRRI, 2014) at ICAR-National Rice Research Institute, India. Biochemical parameters like Alcohol Dehydrogenase activity (ADH) (Lowry *et al.*, 1951) total soluble sugar content (DuBois *et al.*, 1956, Krishnaveni *et al.*, 1984), total chlorophyll content (Witham *et al.*, 1971) and proline content (PC) (Bates *et al.*, 1973) were estimated/analyzed to assess the biochemical changes and their relation with *Sub1* in tolerant genotypes. Twenty days old seedlings of each genotype were grown in screening tank in three rows with 22 plants per row at 20cm x 15 cm spacing. Subsequently, complete submergence was imposed for 14 days with 1.5-meter water depth. After fourteen days of submergence, water was removed from the tanks and regeneration was recorded after one week (IRRI, 2014). The data were recorded on five plants from each of the entries for the characters namely: days to 50% flowering (DFF), days to maturity (DM), plant height (PH), flag leaf length (FLL), flag leaf width (FLW), number of effective tillers per plant (NETPP), panicle length (PL), panicle weight (PW), number of grains per panicle (NGPP), number of chaffs per panicle (NCPP), Spikelets fertility % (SF%), test weight (TW), grain yield per plant (GYPP) and % survival (% S). Further, the lines were also analyzed for grain and cooking quality parameters such as head rice recovery (HRR) [34], kernel length before cooking (KLBC), kernel breadth before cooking (KBBC), length/breadth ratio (L/B), amylose content (AC) and panel test for palatability trait as described by (Khanna *et al.*, 2015). The statistical analysis was performed using XLSTAT and SPSS packages. Duncan's Multiple Range Test (DMRT) adjudged the treatment means (Gomez *et al.*, 1984). Use of plant material comply with relevant institutional, national, and international guidelines and legislation.

RESULTS AND DISCUSSION

Submergence due to flash flooding during different growth stages in rice affects the growth and productivity. Morphological and physiological parameters of eleven rice genotypes (tolerant and intolerant) recorded at different submergence regimes (0, 7 and 14 days of submergence) showed significant differences at 5% level of probability. Seven days of submergence recorded highest days to fifty percent flowering in all genotypes and it was Upahar amongst genotypes (144.3 days) followed by CRL22R (131.7 days). Similarly, interaction effect of genotype and submergence regime, Upahar recorded highest days of maturity at 14 days of submergence followed by CRL 22R (154.0 days) and Hasanta (152.3 days). (Table 1). In contrast, plant height showed reducing trend with the duration of submergence regimes, it was recorded maximum under 7 days' submergence condition. Genotypes Hasant and Upahar recorded maximum height reduction followed by Mrunalini and CR Dhan 801 (Table 1). Similarly, flag leaf length was another morphological parameter recorded drastic reduction in their length under submergence, it was recorded maximum value under 7 days of submergence in tolerant and intolerant genotypes. CRL 22R, CRMS 31B, Hasanta and Mrunalini has recorded maximum reduction under seven days of submergence stress. Whereas, flag leaf width recorded increasing trend, it was recorded widened under up to 7 days of submergence, then after reduced leaf width in all genotypes irrespective of their tolerance or intolerance nature (Table 1). Besides, number of effective tillers per plant (NETPP) was recorded varying range of impact, most of the genotypes recorded no substantial impact up to 14 days of submergence, but beyond that all genotypes recorded drastic reduction in effective tillers number per plant. It was intolerant genotype Hasanta which recorded maximum reduction (6.41) followed by Upahar (7.47), IRBB66 (7.7) and CRL 22 (8.87). The minimum reduction in tillering ability under 14 days of submergence was recorded in the tolerant genotypes Swarna Sub1, CRMS31B, CRMS 32B, CR dhan 801 (Table 1.0). In addition, panicle length under submergence was found to be least affected, it was reduced up to seven day of submergence and then after recorded increasing length of panicle in all genotypes. The intolerant line CRL 22R recorded maximum recovery in panicle length under 14 days of submergence (24.69 cm) followed by CR Dhan 801, CRMS 31B, CRMS 32B ect. Similarly, per panicle weight was also decreased with the increasing of submergence regimes, maximum reduction was recorded in the genotype IRBB66 (3.79g) followed by Swarna Shreya (4.47g) and Upahar (4.77g) after 14 days of submergence. Number of grain per plant (NGPP) was recorded varying range of response under submergence, it was drastically reduced up to seven days of submergence, highest reduction was recorded in CRMS 31B and CRMS 32B (more than 50 grain per panicle) followed by Swarna Shreya, CRL 22 and IRBB66. Spikelets fertility tested genotypes is another morphological parameter affected most up to 7 days of submergence, but recovered gradually during 14 days and prolonged submergence. It was CRL 22 which recorded minimum spikeltes fertility reduction followed by CRMS 31B and Swarna Sub-1 under 7 days of submergence. Test weight is less affected parameter under submergence, there were no substantial changes were recorded. whereas, grain yield per plant was also suffered substantially under two weeks of submergence, where CRMS31B and CRMS32B recorded maximum yield (more than 13.0 g/plant) penalty under submergence. Grain quality parameters like dimension and milling quality was least affected under submergence (Table 1), differences recorded in these traits in studied genotypes were due to mere genotypic effect of the lines screened. Plant survibility under submergence on the other hand recorded varied range of response, all genotypes carrying Sub1 genes (Swarna Sub1, Chehrang Sub1, CRMS 31B, CRMS 32B, CR Dhan 801) recorded more than 50% recovery after 14 days of submergence, whereas, rest intolerant genotypes were perished.

The eleven rice genotypes evaluated for physiological traits like, total chlorophyll content, total soluble sugar, ADH activities and proline accumulation under 0, 7, 14 and 21 days of flooding regimes recorded substantial differences at 5% level of significance (Table 2). The total chlorophyll (Chl a and Chl b) content was measured in leaf and culm of 60 days old plants after 7, 14 and 21 days of submergence. The result revealed that the maximum chlorophyll content was reported under control condition (ranged from 3.23 to 1.30 mg g-1), highest value is recorded in Hasanta under control (3.23 mg g-1), under 14 days of submergence it was recorded highest in Swarna Sub1 (3.20 mg g-1) and other Sub1 carrying genotypes except CR Dhan 801 which recorded lowest (1.30 mg g-1) among tested genotypes. Furthermore, total soluble sugar (mg g-1 fr. wt.) was recorded maximum in non-submerged leaves in all genotypes which slowly decreased with

Table 1. Combined effect of genotypes and different submergence regimes on agro-morpho and physiological traits of 11 rice genotypes

Geno-type	Submerge-nce regime	DFF	DM	PH (cm)	FLL (cm)	FLW (cm)	NET PP	PL (cm)	PW (g)	NGPP	NCPP	SF%	TW	GYPP (g)	KL (mm)	KW (mm)	L/B Ratio	HR R%	SV %
A	Control	112.0^{bc}	142.3^{bc}	124.2^{b}	27.3^{bcd}	1.144^{bc}	9.667^{d}	23.980^{c}	11.970^{a}	158.667^{de}	28.000^{bcd}	85.006^{ab}	24.803^{ab}	27.837^{e}	4.501^{d}	1.795^{a}	2.510^{e}	64.842^{a}	99.000^{a}
	7 days	130.0^{ab}	136.1^{a}	83.2^{a}	15.7^{a}	3.017^{a}	12.023^{a}	17.593^{a}	55.987^{a}	109.333^{a}	52.708^{a}	59.877^{a}	23.520^{abc}	17.806^{a}	3.597^{a}	1.983^{a}	23.154^{a}	68.538^{a}	76.333^{c}
	14 days	121.0^{c}	152.3^{b}	99.6^{bc}	22.9^{bcd}	1.23^{bc}	6.41^{d}	20.89^{c}	9.82^{a}	130.66^{de}	43.00^{bcd}	75.23^{bc}	22.39^{ab}	22.08^{e}	4.51^{d}	1.78^{a}	2.53^{e}	58.46^{a}	14.33^{cd}
B	Control	111.3^{bc}	144.0^{b}	118.3^{c}	25.1^{d}	1.254^{ab}	11.667^{bcd}	22.980^{c}	10.320^{bc}	144.667^{ef}	33.667^{bc}	81.127^{b}	22.873^{abc}	29.037^{d}	4.181^{e}	1.665^{a}	2.514^{e}	60.722^{bc}	100.000^{a}
	7 days	129.6^{ab}	134.9^{a}	78.7^{a}	14.2^{a}	3.757^{a}	13.023^{a}	16.377^{a}	50.220^{a}	102.333^{a}	54.442^{a}	56.667^{a}	22.633 ^{b}c	18.499^{a}	3.340^{a}	1.897^{a}	21.786^{a}	67.124^{a}	83.333^{b}
	14 days	121.6^{c}	150.6^{b}	96.4^{bcd}	20.7^{d}	1.34^{ab}	8.41^{bcd}	19.89^{c}	8.17^{bc}	116.66^{ef}	48.67^{bc}	70.57^{c}	20.46^{abc}	23.28^{d}	4.19^{e}	1.65^{a}	2.53^{e}	54.34^{bc}	92.00^{ab}
C	Control	108.67^{c}	140.0^{c}	99.3^{f}	22.1^{e}	0.944^{d}	13.533^{ab}	20.180^{d}	11.120^{ab}	135.667^{f}	18.000^{d}	88.298^{a}	23.083^{abc}	36.237^{b}	4.401^{d}	1.425^{bc}	3.094^{d}	65.042^{a}	98.333^{a}
	7 days	126.7^{ab}	126.7^{a}	64.8^{a}	12.1^{a}	4.617^{a}	12.890^{a}	14.777^{a}	47.753^{a}	90.667^{a}	46.977^{a}	60.716^{a}	25.173^{abc}	23.373^{a}	3.407^{a}	1.917^{a}	23.632^{a}	74.338^{a}	94.667^{a}
	14 days	117.0^{d}	150.0^{b}	93.9^{cd}	17.7^{e}	1.04^{d}	10.23^{ab}	17.09^{d}	8.97^{ab}	107.66^{f}	33.00^{d}	76.53^{ab}	20.67^{abc}	30.48^{b}	4.41^{d}	1.41^{bc}	3.12^{d}	58.66^{a}	8.67^{de}
D	Control	127.0^{a}	158.3^{a}	129.3^{a}	31.4^{a}	1.124^{bc}	10.667^{cd}	23.980^{c}	6.920^{efg}	156.333^{de}	37.333^{b}	80.713^{b}	24.503^{ab}	34.437^{c}	4.121^{e}	1.665^{a}	2.478^{e}	58.142^{cd}	95.667^{a}
	7 days	144.3^{a}	148.7^{a}	88.0^{a}	18.4^{a}	3.337^{a}	12.690^{a}	15.910^{a}	49.620^{a}	114.000^{a}	55.941^{a}	57.684^{a}	25.520^{abc}	22.079^{a}	3.300^{a}	1.885^{a}	20.901^{a}	66.738^{a}	82.667^{b}
	14 days	139.0^{a}	168.3^{a}	113.10^{a}	27.09^{a}	1.21^{bc}	7.47^{cd}	20.89^{c}	4.77^{efg}	128.33^{de}	52.33^{b}	71.01^{c}	22.09^{ab}	28.68^{c}	4.13^{e}	1.65^{a}	2.49^{e}	51.76^{cd}	89.00^{b}
E	Control	99.6^{e}	126.3^{f}	103.0^{ef}	28.4^{bc}	1.164^{bc}	14.650^{a}	26.080^{b}	9.920^{bc}	276.667^{a}	51.667^{a}	84.280^{ab}	25.973^{a}	44.437^{a}	4.801^{c}	1.365^{bc}	3.524^{ab}	55.442^{d}	98.667^{a}
	7 days	116.0^{ab}	119.3^{a}	68.4^{a}	16.4^{a}	5.031^{a}	15.707^{a}	18.310^{a}	93.953^{a}	195.000^{a}	69.778^{a}	61.138^{a}	29.833^{a}	28.973^{a}	3.654^{a}	2.011^{a}	20.732^{a}	67.271^{a}	96.333^{a}
	14 days	111.6^{e}	136.3^{d}	94.4^{bcd}	24.0^{bc}	1.25^{bc}	11.00^{a}	22.99^{b}	7.77^{bc}	248.66^{a}	66.67^{a}	78.87^{ab}	23.58^{a}	38.68^{a}	4.81^{c}	1.35^{bc}	3.55^{ab}	49.06^{d}	15.00^{c}
F	Control	101.6^{de}	130.3^{e}	100.9^{f}	29.3^{ab}	1.124^{bc}	13.450^{ab}	27.980^{a}	9.720^{c}	250.333^{b}	53.000^{a}	82.499^{b}	20.403^{c}	23.437^{f}	4.901^{bc}	1.395^{bc}	3.520^{ab}	59.862^{bc}	100.000^{a}
	7 days	119.0^{ab}	120.3^{a}	68.3^{a}	17.0^{a}	4.671^{a}	15.473^{a}	19.510^{a}	81.153^{a}	183.333^{a}	67.790^{a}	58.582^{a}	19.120^{c}	15.006^{a}	3.730^{a}	2.030^{a}	22.201^{a}	70.884^{a}	93.000^{a}
	14 days	113.6^{de}	140.3^{c}	92.2^{cd}	24.9^{ab}	1.21^{bc}	10.20^{ab}	24.89a	7.57^{c}	222.33^{b}	68.00^{a}	76.53^{ab}	17.98^{c}	17.68^{f}	4.91^{bc}	1.38^{bc}	3.55^{ab}	53.48^{bc}	94.00^{ab}
G	Control	110.6^{bc}	140.3^{c}	109.3^{d}	27.3^{bcd}	1.124^{c}	12.667^{abc}	23.780^{c}	7.520^{ef}	145.667^{ef}	26.000^{bcd}	84.861^{ab}	22.083^{bc}	23.237^{f}	4.181^{e}	1.365^{bc}	3.069^{d}	62.542^{ab}	100.000^{a}
	7 days	128.0^{ab}	129.8^{a}	73.333^{a}	15.7^{a}	4.004^{a}	13.957^{a}	15.977^{a}	48.687^{a}	100.000^{a}	51.236^{a}	58.551^{a}	20.173^{bc}	14.633^{a}	3.240^{a}	1.868^{a}	22.784^{a}	73.004^{a}	98.333^{a}
	14 days	122.6^{bc}	150.3^{b}	95.43^{bcd}	22.99^{bcd}	1.24^{c}	9.41^{abc}	20.69^{c}	5.37^{ef}	117.66^{ef}	41.00^{bcd}	74.15^{bc}	19.67^{bc}	17.48^{f}	4.19^{e}	1.35^{bc}	3.09^{d}	56.16^{ab}	11.00^{cde}
H	Control	99.7^{e}	124.0^{f}	106.783^{de}	26.065^{cd}	1.084^{c}	11.667^{bcd}	22.980^{c}	8.060^{de}	165.333^{d}	32.667^{bc}	83.833^{ab}	23.503^{abc}	27.037^{e}	4.981 b	1.465 bc	3.406 bc	55.892^{d}	100.000^{a}
	7 days	116.0^{ab}	117.0^{a}	71.173^{a}	14.8^{a}	3.644^{a}	13.023^{a}	15.623^{a}	56.380^{a}	117.333^{a}	51.537^{a}	59.995^{a}	22.387^{bc}	17.433^{a}	3.807 a	2.043 a	20.796 a	67.904^{a}	93.333^{a}
	14 days	111.6^{e}	134.0^{de}	94.8^{bcd}	21.7^{cd}	1.14^{c}	8.41^{bcd}	19.89^{c}	5.91^{de}	137.33^{d}	47.67^{bc}	74.52^{bc}	21.09^{abc}	21.28^{e}	4.99 b	1.45 bc	3.43 bc	49.51^{d}	95.00^{a}
I	Control	95.6^{f}	120.3^{g}	109.8^{d}	25.7^{d}	1.194^{abc}	13.333^{ab}	22.780^{c}	6.620^{fg}	184.667^{c}	37.667^{b}	83.064^{b}	24.603^{ab}	27.137^{e}	4.501 d	1.395 bc	3.233 cd	59.622^{bc}	99.333^{a}
	7 days	111.3^{b}	117.1^{a}	72.6^{a}	14.6^{a}	4.551^{a}	13.790^{a}	15.010^{a}	61.087^{a}	130.333^{a}	58.031^{a}	59.267^{a}	23.153^{abc}	17.339^{a}	3.464 a	1.940 a	21.922 a	69.058^{a}	85.667^{b}
	14 days	107.6^{f}	130.3^{e}	95.9^{bcd}	21.3^{d}	1.28^{abc}	10.03^{ab}	19.69^{c}	4.47^{fg}	156.66^{c}	52.67^{b}	74.83^{bc}	22.19^{ab}	21.38^{e}	4.51 d	1.38 bc	3.26 cd	53.24^{bc}	13.33^{cde}
J	Control	113.7^{b}	144.0^{b}	117.5^{c}	29.1^{ab}	1.304^{a}	12.067^{bc}	27.780^{a}	9.320^{cd}	186.667^{c}	25.000^{cd}	88.439^{a}	26.603^{a}	33.737^{c}	5.501 a	1.505 b	3.661 ab	59.662^{bc}	97.333^{a}
	7 days	131.7^{ab}	134.5^{a}	79.4^{a}	16.9^{a}	4.324^{a}	14.490^{a}	19.243^{a}	63.553^{a}	129.667^{a}	48.182^{a}	64.358^{a}	26.687^{ab}	22.073^{a}	4.167 a	2.151 a	22.227 a	64.418^{a}	92.333^{a}
	14 days	125.6^{b}	154.0^{b}	103.9^{b}	24.79^{ab}	1.34^{a}	8.87^{bc}	24.69^{a}	7.17^{cd}	158.66^{c}	40.00^{cd}	80.17^{a}	24.18^{a}	27.98^{c}	5.51 a	1.49 b	3.69 ab	53.28^{bc}	15.00^{c}
K	Control	103.6^{d}	134.0^{d}	103.2^{ef}	21.0^{e}	1.084^{c}	11.000^{cd}	20.080^{d}	5.940^{g}	163.667^{d}	32.333^{bc}	83.520^{ab}	21.793^{bc}	23.137^{f}	4.941 bc	1.335 c	3.709 a	60.862^{bc}	100.000^{a}
	7 days	121.7^{ab}	123.1^{a}	67.1^{a}	11.4^{a}	3.977^{a}	11.057^{a}	12.983^{a}	53.633^{a}	113.667^{a}	55.761^{a}	57.868^{a}	19.947^{bc}	14.819^{a}	3.737 a	2.048 a	22.668 a	70.884^{a}	80.000^{bc}
	14 days	114.6^{de}	141.6^{c}	88.0^{d}	16.6^{e}	1.14^{c}	7.70^{cd}	16.99^{d}	3.79^{g}	135.66^{d}	47.33^{bc}	74.14^{bc}	19.38^{bc}	17.38^{f}	4.95 bc	1.32 c	3.74 a	54.48^{bc}	8.33^{e}

Values having common letter(s) in a column do not differ significantly at $p \leq 0.05$ as per DMRT.

increase in sub-mergence period. Under non-submergence (control condition), all genotypes had high level of soluble sugar content which showed considerable reduction up to 7 days of submergence then after recorded increasing trend. Tolerant genotypes had comparatively slowed increase in soluble sugar with prolonged submergence period (beyond 7 days flooding). Result indicates slowed consumption of insoluble sugar during flooding led submergence tolerance in rice.

Similarly, activity of Alcohol dehydrogenase (ADH) was reported increasing trend, recorded maximum (1.60 to 9.67 per minute per g fr. wt.) in the lines carrying Sub1 gene even after 14 day of submergence. ADH activities was found highest in the Swarna Sub1 (9.90 per minute per g fr. wt.) followed by Hasanta (9.67 per minute per gm fr. wt.). In addition, proline maintains normal osmoregulation in plant tissue under submergence. Our results revealed increasing trend of proline accumulation under submergence in all genotypes carrying Sub1. Genotypes carrying Sub1 gene showed maximum proline accumulation even after 14 days of submergence (Table 2).

Submergence stress is detrimental to rice production in Asian countries, accounting ~600 million USD annual monetary loss (Kumar *et al.*, 2021). Turbid water and dense algal growth under flooding hinders light and gaseous transmission, affects the photosynthetic activity and led plants to die due to starvation (Bailey-Serres *et al.*, 2008, Bhaduri *et al.*, 2020). Submergence stress alongwith

Table 2. Combined effect of genotypes and different submergence regimes (0, 7, 14, 21) on physiological traits of 11 rice genotypes

Genotype	Submergence regime	TCC	TSS	ADH	Proline content
Hasanta	Control	3.23a	0.11 ab	2.28 a	0.097 ab
	7 days	3.10 a	0.123 a	9.40 a	0.103 a
	14 days	2.36 bc	0.15a	9.67 a	0.110 a
Mrunalini	Control	2.45 bc	0.13 a	2.14 ab	0.100 a
	7 days	2.30 c	0.13 a	7.75 b	0.090 a
	14 days	2.40 b	0.14 ab	5.91 b	0.103 ab
Swarna Sub-1	Control	2.40 bc	0.12 ab	2.03 ab	0.083 ab
	7 days	2.43 bc	0.11 ab	9.90 a	0.103 a
	14 days	3.20 a	0.13 abc	5.72 b	0.097 abc
Upahar	Control	3.13 a	0.11 ab	2.30 a	0.097 ab
	7 days	2.60 b	0.12 a	2.53 cd	0.100 a
	14 days	1.83 de	0.12 bc	3.65 d	0.090 bcd
CRMS 31B	Control	2.40 bc	0.11 ab	2.26 a	0.090 ab
	7 days	1.80 fg	0.11 ab	2.36 cd	0.090 a
	14 days	2.40 b	0.13 abc	3.82 cd	0.097 abc
CRMS32B	Control	2.63 b	0.13 a	2.10 ab	0.090 ab
	7 days	2.03 def	0.11 ab	1.96 cde	0.100 a
	14 days	2.16 bc	0.08 d	4.70 c	0.090 bcd
CR Dhan 801	Control	2.56 b	0.10 b	2.10 ab	0.080 b
	7 days	2.36 bc	0.11 ab	1.93 de	0.097 a
	14 days	1.30 f	0.13 abc	4.08 cd	0.090 bcd
Chehrang sub-1	Control	2.03 d	0.10 b	2.03 ab	0.093 ab
	7 days	2.03 de	0.11 ab	2.63 c	0.093 a
	14 days	1.60 ef	0.13 abc	2.37 e	0.087 bcd
Swarna Shreya	Control	2.40 bc	0.13 b	2.08 ab	0.090 ab
	7 days	2.07 d	0.11 ab	2.09 cde	0.083 a
	14 days	1.30 f	0.12 c	2.53 e	0.080 cd
CRL 22R	Control	2.00 d	0.13 a	1.83 b	0.080 b
	7 days	1.80efg	0.10 b	2.33 cd	0.070 a
	14 days	2.07 cd	0.07 d	3.67 d	0.080 cd
IRBB66	Control	2.20 cd	0.11 ab	1.77 b	0.080 b
	7 days	1.60 g	0.07 c	1.60 e	0.090 a
	14 days	1.33 f	0.07 d	4.07 cd	0.077 d

Values having common letter(s) in a column do not differ significantly at pd”0.05 as per DMRT.

other biotic factors causing substantial yield and quality loss in rice (Kumar *et al*., 2021). Frequent occurrence of flash flooding and pathogenic dynamics with varying levels of genetic diversity and virulence renders their management extremely challenging (Kumar *et al*., 2021). There are plenty of suitable rice cultivars are available but stress due to submergence threatens rice productivity under RSL and deep water ecosystem (Chetia *et al*., 2018). Enhancing resistance level of the host plant presents a standard practice to mitigate the risks of biotic and abiotic stresses, ecofriendly (Chukwu *et al*., 2019, Das *et al*., 2016, and 2018). Evidence suggests that stacking tolerant/ multiple R genes resulting in quantitative complementation or synergistic response, improves the resistance/tolerant level of plants (Pradhan *et al*., 2019). In recent years, advanced genomic tools (Chukwu *et al*., 2019; Das *et al*., 2016, and 2018, Cobb *et al*., 2019) have made significant contributions to the trait improvement in plant breeding. In this context, identification of suitable donor and recurrent genetic background is paramount importance for improving the resilient traits in rice (Pradhan *et al*., 2015 and 2019).

The present study demonstrates the utility prior screening of parent genotypes to be utilized as donor and recurrent parents in trait improvement programs. The results demonstrated that genotype carrying Sub1 gene had >02 week flooding tolerant (Pradhan *et al*., 2019) than intolerant genotypes (Cheema *et al*., 2008). Selection of genetically similar parent helps in reduction in number of backcrosses in crop improvement program as led accelerated accumulation of desirable genes (minor) in derivatives. Being semi aquatic plant, rice is able to make several modulations to sustain under flood adversity (Kumar *et al*., 2021). Results of the physiological analysis of total soluble sugar content, total chlorophyll content, ADH activities and proline accumulation under submergence revealed considerable impact of Sub1 introgression in the carrier genotypes like Swarna Sub1, CRMS31B, CRMS 32B, CR Dhan 801 and Chehrang Sub1 (Table 2). Delayed degradation of the total chlorophyll content under complete flooding helps plant to sustain longer (Adak *et al*., 2000). SUB1A delays dark-induced senescence through the conservation of carbohydrate reserves and chlorophyll in the photosynthetic tissue (Ghosh *et al*., 1971) which helps in post submergence fast recovery (Xu *et al*., 1996). Our result revealed comparatively slowed degradation in total chlorophyll content in the genotypes carrying Sub1 is fully corroborated with previous findings (Deka *et al*., 2000).

Under submergence, tolerant plants are capable to restricts their growth, thus, conserve ~25-50% more nonstructural carbohydrate which helps in faster post sub-mergence recovery "quiescent strategy" [88]. Slowed release of non-structural sugar or total soluble sugar in submerged canopy indicates its judicious consumption of stored sugar which helps prolonged survibility and post submergence fast recovery (Sarkar *et al*., 2006). We have reported in our study that lines carrying Sub1 gene had showed slowed release of soluble sugar even beyond two weeks of submergence followed 'quiescence' strategies' of submergence tolerant (Kumar *et al*., 2021). Whereas, intolerant lines showed rapid increment in non-structural sugar after one week of submergence. Increased activity of hydrolase enzymes like ADH, á-amylase etc. facilitates energy to submerged canopy through anaerobic respiration (Kumar *et al*., 2021). Results of ADH activities revealed that lines carrying Sub1 including donor had progressive ADH activity during submergence which helped in reduction of aldehyde production, thus increase hypoxia/anoxia tolerance in rice (Rahman *et al*., 2018). Increased accumulation of proline content in stressed plant is also an adaptive mechanism which help to maintain osmoregulation in the submerged condition. The results of this study will helps in selection of desirable parents, specially donor with maximum complementarity with recurrent parents in trait improvement programs for precise and fast development of resilient varieties in rice.

CONCLUSION

All the eleven rice cultivars viz, Swarna Sub1, CRMS 31B, CRMS 32B, Chehrang Sub1, CR Dhan 80, Mrunalini, Hasanta, Upahar, Swarna Shreya, IRBB66 and CRL 22R irrespective of submergence regimes, showed varying range of stress response. Lines carrying Sub1 gene showed better recovery of two-week submergence than intolerant genotypes. From the findings of the study, it may be concluded that among the submergence regimes, seven days' submergence showed the highest interaction effect among the studied traits. So, these stage is crucial because significant reduction was observed in morpho-physiology of different parts due to submergence stress under 7 days of submergence, so, parental lines showed least vulnerability under seven days' submergence might be utilized in traits improvement program.

ACKNOWLEDGEMENTS

We acknowledged the director ICAR-NRRI, Cuttack and VC, OUAT, Bhubaneshwar for all facilities.

REFERENCES

Adak, M. K. and Gupta, D. K. D. 2000. Prolonged waterlogging on photosynthesis and related characters in rice. *Indian Journal of Plant Physiology.* 5: 380–382.

Adkins, S.W., Shiraishi, T. and McComb, J.A. 1990. Submergence tolerance of rice, a new glasshouse method for the experimental submergence of plants. *Plant Physiology.* 80 : 642-646.

Bailey-Serres, J. and Voesenek, L.A.C.J. 2008. Flooding stress: acclimations and genetic diversity. *Annual Review of Plant Biology.* 59 : 313– 339.

Bates, L.S., Waldren, R.P. and Teare, I.D. 1973. Rapid Determination of Free Proline for Water Stress Studies. *Plant and Soil.* 39 : 205-207. http://dx.doi.org/10.1007/BF00018060.

Bhaduri, D., Chakraborty, K., Nayak, A. K., Shahid, M., Tripathi, R., Behera, R., Singh, S. and Srivastava, A.K. 2020. Alteration in plant spacing improves submergence tolerance in Sub1 and non-Sub1 rice (cv. IR64) by better light interception and effective carbohydrate utilisation under stress. *Functional Plant Biology.* 47 : 891-903. doi: 10.1071/FP19364.

Catling, D. 1992. Rice in deep water. International Rice Research Institute, Manila, Philippines.

Cheema, K., Grewal, N., Vikal, Y., Sharma, R., Lore, J.S., Das, A., Bhatia, D., Mahajan, R., Gupta, V., Bharaj, T.S. and Singh, K.A. 2008. A novel bacterial blight resistant gene from Oryza nivara mapped to 38 kb region on chromosome 4L and transferred to *Oryza sativa* L. *Genet. Res. Cambridge.* 90 : 397–307.

Chetia, S.K., Kalita, M., Verma, R.K., Barua, B., Ahmed, T. M., Modi, K., and Singh, N.K. 2018. Flood proofing of Ranjit, a popular variety of North-Eastern India through transfer of Sub1 rice QTL by modified marker-assisted backcross breeding. *Indian J. Genet.* 78 : 166-173, DOI: 10.5958/0975-6906.2018.00021.4.

Chukwu, S.C., Rafii, M.Y., Ramlee, S.I., Ismail, S.I., Hasan, M.M., Oladosu, Y.A., Magaji, U.G., Akos, I. and Olalekan, K.K. 2019. Bacterial leaf blight resistance in rice: a review of conventional breeding to molecular approach. *Molecular Biology Reports.* 46: 1519–1532.

Cobb, J.N., Biswas, P.S. and Platten, J.D. 2019. Back to the future: Revisiting MAS as a tool for modern plant breeding. *Theor. Appl. Genet.* 132: 647–667.

Das, G., Rao, G.J.N. Varier, M. and Prakash, A. 2018. Prasad, D. Improved Tapaswini having four BB resistance genes pyramided with six genes/QTLs, resistance/tolerance to biotic and abiotic stresses in rice. *Sci. Rep.* 8 : 2413. https://doi.org/10.1038/s41598-018-20495-x.

Dash, A.K., Rao, R.N. Rao, G.J.N. Verma, R.L. Katara, J.L., Mukherjee, A.K., Singh, O.N. and Bagchi, T.B. 2016. Phenotypic and Marker-Assisted Genetic Enhancement of Parental Lines of Rajalaxmi, an Elite Rice Hybrid. *Front. Plant Sci.* 7, 1005. doi: 10.3389/fpls.2016.01005.

Deka, M. and Baruah, K. K. 2000. Comparable studies of rainfed upland winter rice (*Oryza sativa*) cultivars for drought tolerance. *Indian Journal of Agricultural Sciences.* 70 : 135–139.

DuBois, M., Gilles, K., Hamilton, J. Rebers, P. Smith, F. 1956. Colorimetric method for determination of sugars and related substances. *Analytical Chemistry.* 28: 350–356.

Ghosh, A. K., Nanda, B. B., Swami, S. G. and Nayak, B. B. 1971. Influence of nitrogen on the physico-chemical characteristics of rice grain. *Oryza.* 891: 87–97.

Godfray, H.C.J., Beddington, J.R., Crute I.R., Haddad, L., Lawrence, D., Muir, J.F. and Toulmin, C. 2010. Food security: The challenge of feeding 9 billion people. *Science.* 327: 812–818.

Gomez, K.A. and Gomez, A.A. 1984. *Statistical Procedures for Agricultural Research* (2 Ed.). John wiley and sons, NewYork, 680p.

Hasan, M.M., Rafii, M.Y., Ismail, M.R., Mahmood, M., Rahim, H.A. and Alam M.A. 2015. Marker assisted backcrossing: a useful method for rice improvement. *Biotechnology and Biotechnological Equipments* 29: 237-254.

Iftekharuddaula, K.M., Ahmed, H.U., Ghosal, S., Moni, Z.R., Amin, A. and Ali, M.S. 2015. Development of a New Submergence Tolerant Rice Variety for Bangladesh Using Marker-Assisted Backcrossing. *Rice Science.* 22: 15-26.

IRRI 2014. International Rice Research Institute: Standard evaluation system for rice (SES) 5. Los Baños: *International Rice Research Institute.*

Kabir, M.E., Iftekharuddaula, K.M., Khan, M.A.I., Mian, M.A.K., Ivy, N. A. Marker assisted introgression of bacterial blight resistant gene into submergence tolerance rice variety BRRI dhan52. *Bangladesh J. Agril. Res.* 42: 403-411.

Khanna, A. 2015. Development and evaluation of near-isogenic lines for major blast resistance gene(s) in Basmati rice. *Theor. Appl. Genet.* 128 : 1243–1259.

Khush, G.S. 1997. Origin, dispersal, cultivation and variation of rice. *Plant Molecular Biology.* 35 : 25–34.

Krishnaveni, S., Balasubramanian, T. and Sadasivam, S. 1984. Sugar distribution in sweet stalk sorghum. *Food Chemistry.* 15 : 229-232.

Kumar, A., Nayak, A.K., Hanjagi, P.S., Kumari, K., Vijaykumar, S., Mohanthy, S., Tripathi, R. and Pannerselvam, P. 2021. Submergence stress in rice: Adaptive mechanism, coping strategies and future research needs. *Environmental and Experimental Botany.* 186: 104448.

Lowry, O.H., Rosenbrough, N.J., Farr, A.L. and Randall, R.J. 1951. Protein measurement with the Folin Phenol Reagent. *Journal of Biological Chemistry.* 193: 265-275.

Nair, M.M. and Shylaraj, K.S. 2017. Introgression of dual abiotic stress tolerance QTLs (Saltol QTL and Sub1 gene) into Rice (*Oryza sativa* L.) variety Aiswarya through marker assisted backcross breeding. *Physiol Mol Biol Plants.* 27: 497-514. doi: 10.1007/s12298-020-00893-0.

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

Palada, M.C. and Vergara, B.S. 1972. Environmental effects on the resistance of rice seedlings to complete submergence. *Crop Science.* 12 : 209-212.

Pradhan, S. K. 2015. Pyramiding of three bacterial blight resistance genes for broad-spectrum resistance in deepwater rice variety. *Jalmagna. Rice.* https://doi.org/10.1186/s12284-015-0051-8.

Pradhan, S.K., Pandit, E., Pawar, S., Baksh, S.Y., Mukherjee, A.K. and Mohanty, S.P. 2019. Development of flash-flood tolerant and durable bacterial blight resistant versions of mega rice variety 'Swarna' through marker-assisted backcross breeding. *Scientific Reports.* 9: 12810 , https://doi.org/10.1038/s41598-019-49176-z .

Pradhan, S.K., Pandit, E. and Pawar, S. 2019. Development of flash-flood tolerant and durable bacterial blight resistant versions of mega rice variety 'Swarna' through marker-assisted backcross breeding. *Sci Rep.* 9: 1-15, 12810, https://doi.org/10.1038/s41598-019-49176-z.

Rahman, H., Dakshinamurthi, V., Ramasamy, S., Manickam, S., Kaliyaperumal, A.K., Raha, S., Paneerselvam N., Ramanathan V., Nallathambi, J., Sabariyappan, R. and Raveendran M. 2018. Introgression of Submergence Tolerance into CO 43, a Popular Rice Variety of India, through Marker-assisted Backcross Breeding. *Czech Journal of Genetics and Plant Breeding.* 54.

Rajpurohit, D., Kumar, R., Kumar, M., Paul, P., Awasthi, A., Osman, B. P., Puri, A., Jhang, T., Singh, K. and Dhaliwal, H.S. 2011. Pyramiding of two bacterial blight resistance and a semi dwarfing gene in Type 3 Basmati using marker-assisted selection. *Euphytica.* 178 : 111–126.

Ronald, P. C., Albano, B., Tabien, R., Abenes, M. L. P., Wu, K. S. and McCouch, S. R. 1992. Genetic and physical analysis of the rice bacterial blight disease resistance locus Xa21. *Mol. Gen. Genet.* 236: 113–120.

Samanta, P., Ganie, S.A., Chakraborty, A. and Dey, N. 2020. Study on regulation of carbohydrate usage in a heterogeneous rice population under submergence. *J. Plant Biochem. Biotechnol.* https://doi.org/10.1007/s13562-020-00577-6.

Sanchez, A. C., Brar, D. S., Huang, N., Li, Z. and Khush, G. S. 2000. Sequence tagged site marker-assisted selection for three bacterial blight resistance genes in rice. *Crop Sci.* 40: 792–797.

Sarkar, R.K., Das K.K., Panda, D., Reddy, J.N., Patnaik, S.S.C., Patra, B.C. and Singh, D.P. 2014. Submergence tolerance in rice: Biophysical Constraints, Physiological basis and Identification of Donors. Central Rice Research Institute, Cuttck, India. p.36.

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

Singh, P. and Sinha, A.K. 2016. A Positive Feedback Loop Governed by SUB1A1 Interaction with MITOGEN-ACTIVATED PROTEIN KINASE3 Imparts Submergence Tolerance in Rice. *Plant Cell.* 2016, 28, 1127-43. doi: 10.1105/tpc.15.01001. Epub, Apr 14. PMID: 27081183, PMCID: PMC4904673.

Singh, R., Singh, Y., Xalaxo, S., Verulkar, S., Yadav, N., Singh, S., Singh N., Prasad, K.S.N., Kondayya, K., Rao, P.V.R., Rani, M.G., Anuradha, T., Suraynarayana, Y., Sharma P.C., Krishnamurthy, S.L., Sharma, S.K., Dwivedi, J.L., Singh, A.K., Singh, P.K., Nilanjay, Singh, N.K., Kumar, R., Chetia, S.K., Ahmad, T.,Rai, M., Perraju, P., Pande, A., Singh, D.N., Mandal, N.P., Reddy, J.N., Singh, O.N., Katara, J.L., Marandi, B., Swain, P., Sarkar, R.K., Singh, D.P., Mohapatra, T., Padmawathi, G., Ram, T., Kathiresan, R.M., Paramsivam, K., Nadarajan, S., Thirumeni, S., Nagarajan, M., Singh, A.K., Vikram, P., Kumar, A,, Septiningshih, E., Singh, U.S., Ismail, A.M., Mackill, D. and Singh, N. K. 2015. 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.* http://dx.doi.org/10.1016/j.plantsci. 08.008.

Xu, K. and Mackill, D. J. 1996. A major locus for submergence tolerance mapped on rice chromosome 9. *Mol. Breed.* 2 : 219–224.

Xu, K., Xu, X., Fukao, T., Canlas, P., Maghirang-Rodriguez, R., Heuer, S., Ismail, A.M., Bailey- Serres, J., Ronald, P.C. and Mackill, D.J. 2006. Sub1A is an ethylene-response-factor-like gene that confers submergence tolerance to rice. *Nature.* 442 : 705-708.