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Effect of submergence stress on yield and yield components of various rice (*Oryza sativa* L.) genotypes with its mapping population

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Abstract

An Experiment was conducted with rice genotypes Swarna Sub1 and Nagina 22 with its mapping population in pot during kharif season 2017 at the pond of department of crop physiology in Narendra Deva University of Agriculture and Technology, Kumarganj, Faizabad (U.P). Submergence treatment was given for 14 days at vegetative stage. The screening of rice genotypes was done on the basis of Shoot elongation, Survival percentage, estimation of catalase, peroxidase and chlorophyll content. The rice genotypes Swarna *Sub1* showed high percent increase in catalase and peroxidase whereas in case of chlorophyll content Swarna *Sub1* shower higher percent reduction in Nagina 22. Therefore, on the basis of their parents some mapping population showed less and higher percent reduction and also showed best tolerance in compare to other population.

Keywords: Shoot elongation, catalase (CAT), peroxidase, rice, submergence, analysis, chlorophyll (SPAD value)

1. Introduction

Rice (*Oryza sativa* L.) is a staple food for more than half of the world's population, but it's production is severely being affected by the impact of abiotic stresses due to frequently changing climate. Rice is grown in almost all the states of India but its cultivation is mainly concentrated in river valleys, deltas and low lying coastal areas. The main rice producing states are West Bengal, Uttar Pradesh, Andhra Pradesh, Punjab, Tamil Nadu, Odisha and Bihar. Uttar Pradesh occupies the second position in rice production in India.

Rice crop faces a number of biotic and abiotic stresses in its life span such as submergence, salinity, drought, cold, etc. Among them, flash flooding in rainfed lowlands causes a major losses to rice farmers. When submergence occurs more than 10-days, it leads to death by anoxia. Especially, flooding is a major determining factor in the survival of rice seedlings in rainfed lowland areas. Because prolonged submergence of seedlings causes plant death due to anoxia (Pragnya *et al.*, 2015).

Submergence is the third most important abiotic stress, hindering rice productivity in eastern India. Though rice is a crop that requires flooded and irrigated condition for cultivation, most of the rice varieties are susceptible to flooding if the water stagnates keeping the plants submerged under water for more than seven days causing leaf or stem elongation, leaf rotting, loss of dry mass and also lodging after the flood water recedes (Goswami *et al.*, 2015) [6].

Submergence is a type of flooding stress and is defined as a condition where the entire plant is fully immersed in water (complete submergence) or at least part of the shoot terminal is maintained above the water surface (partial submergence). About 8.0 mha. of rice area is flood/submergence prone in India that constitutes around 35 % of the total rice area categorized under rainfed lowland and flood prone ecology (Reddy *et al.*, 2013) [12]. Fortunately, a major tolerance source called, Submergence 1 (*Sub1*) on chromosome 9 was identified for managing flash flooding in a highly flood tolerant (FR13A) genotype (Pragnya *et al.*, 2015).

Submergence generally occurs at vegetative stage while drought at reproductive stage in frequent manners in life span of the crop. During submergence, tolerant rice plants exhibit inhibition of shoot elongation, retain chlorophyll in leaf tissues and maintain viability. If rice is submerged under water more than a few days, gradual loss of oxygen leads to withering and death due to the effect on respiration and photosynthesis. Water stress may effect on the plants anatomy, morphology, physiology and biochemistry and influence on almost all of their growth and development aspects According to Sarkar *et al.*, (2006) [15] submergence tolerance is a metabolic adaptation in response to anaerobiosis that enables cells to maintain their integrity

so that the plant survives hypoxia without major damages. Submergence tolerance includes a number of anatomical (formation of higher aerenchyma tissue in nodal region), physiological (more shoot elongation) and biochemical (inhibition of chlorophyll degradation, less utilization of storage carbohydrates, and increased activity of antioxidative enzymes) adaptations. A major QTL loci responsible for submergence tolerance was mapped to chromosome 9, designated as *Submergence 1* (*Sub 1*), reported to be accounting for about 70% of the phenotypic variation under submergence. This locus includes three similar genes that encode ethylene response factor (ERF)/ethylene-responsive element binding (EREB) proteins/Apetala2—type transcription factor domain: *Sub1A*, *Sub1B* and *Sub1C*. The plants are adapted to partial submergence by escape strategy, while they shift towards a non-elongating quiescent strategy by utilizing stored reserves when completely submerged. The capability of a rapid regrowth following de-submergence is another mechanism which plays an important role in the submergence tolerance of plants. Submergence tolerance by escape and quiescence strategies does not only require the corresponding regulation of growth and carbohydrate consumption during submergence period but also entails coordinated recovery of photosynthesis and growth following de submergence (Sarkar *et al.*, 2006; Panda *et al.*, 2008; Luo *et al.*, 2011)^[15, 11].

2. Materials and methods

A pot experiment was conducted with Swarna *Sub1* and Nagina 22 as parent and its mapping population BC₂F₃ (Swarna *Sub1* X Nagina 22). The submergence treatment was given thirty-five days old seedling of rice was exposed to 14 days complete submergence stress in the submergence tank of Deptt. of Crop physiology in Narendra Deva University of Agriculture and Technology, Kumarganj, Faizabad (U.P). Plant height were recorded from base of plant to base of panicle of plants. Estimation of catalase and peroxidase was done according to method of Sinha 1972 and Hammerschmidt *et al.*, 1982. Chlorophyll content was recorded by SPAD machine.

3. Result and discussion

Submergence due to flash flood is the key factor limiting yield of lowland rice which adversely affects grain yield of rice crop (Mohanty *et al.*, 2000)^[10]. On the whole, improved submergence tolerance is an important trait for rice growing in rainfed lowland areas. Therefore, efforts are being directed towards improving submergence tolerance character without affecting grain yield.

Effect of submergence on shoot elongation (cm) and survival percentage

Plant survival as affected by 14 days complete submergence treatment is presented in Table 1 and also in graph. It is clear from the data that the survival of all the genotypes were affected under submergence condition. But survival of Nagina-22 was badly affected and survival percentage is 0. Maximum survival was recorded in Swarna *Sub 1* (85.64 %) followed by NDR-9830102 (66.67%), and NDR-97(10.23%), respectively. The shoot elongation data also have depicted in Table 1. The perusal of data clearly indicates that in submergence, plant height increased due to leaf sheath elongation irrespective of genotypes. Maximum shoot elongation as well as elongation percent was recorded in Swarna *Sub 1* followed by NDR-9830102, mapping

population in submergence condition. The shoot elongation in mapping population BC₂F₃, showed maximum in plant number. 7 (5.40cm), followed by plant number 6 (4.20), plant number 2 & plant number 10 (4.00) and minimum in plant number. 8 (2.16cm). Similarly survival percent showed maximum in plant number. 8 (90%) followed by plant number 4 (85%), plant number 3 (74%) while minimum in plant number. 7(20%).

Table 1: Effect of submergence on shoot elongation (cm) and survival (%) in rice genotypes

Parents	Shoot Elongation (cm)	Survival percentage (%)
NDR-9830102	4.52	66.67
NDR-97	4.20	10.23
Swarna <i>Sub 1</i>	5.23	85.64
Nagina-22	5.42	0.00
Mapping population		
1	3.12	70
2	4.00	60
3	3.86	74
4	2.80	85
5	3.50	75
6	4.20	65
7	5.40	20
8	2.16	90
9	3.45	72
10	4.00	68

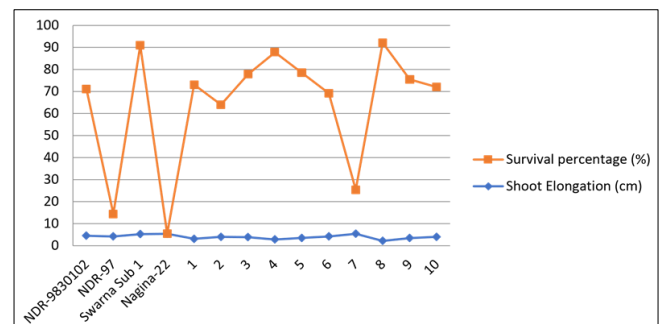


Fig 1: Shoot Elongation (cm) and Survival (%)

Effect of submergence stress on Catalase activity

Catalase (CAT) was assayed by the method described by Sinha *et al.*, (1972). The decrease in H₂O₂ was followed as a decline in absorbance at 570nm using spectrophotometer as against blank. The enzyme activity is expressed as change in absorbance per minute per mg. protein. Rice genotypes *i.e* Swarna *Sub 1* and Nagina 22 showed variability in catalase which was estimated in comparison to their mapping population. High reduction was noted significantly high in Nagina 22 (16.66%) while lowest in Swarna *Sub 1* (13.25%) under submergence stress, while in case of mapping population plant number 1 showed highest % reduction in comparison to plant number. 2,3,4,5,6,7,8,9 and 10 shown in Table 2 and also represented in graph given below. The activity of catalase (CAT) decreased due to submergence stress in both tolerant and susceptible rice cultivars (Sarkar *et al.*, 2001)^[13]. Yet, the enzyme activity was significantly greater in tolerant cultivars and this was highly meaningful for the plants under oxygen-deficient condition because the CAT could directly produce O₂ from H₂O₂ and improve the supply of O₂. The higher levels of H₂O₂ in tolerant cultivars compared to susceptible cultivar under submergence also helped in the maintenance of higher CAT activity and supply probably more molecular O₂. There was not much variation

between tolerant and susceptible cultivars in respect of guaiacol peroxidase (GPX) activity under complete submergence.

Table 2: Effect of submergence stress on catalase activity ($\mu\text{g/g}$ fresh weight mint^{-1}) of rice genotypes

Parent	Control	Submergence	Mean	Percent Increase
NDR-9830102	122.00	302.60	212.30	105.00
NDR-97	119.55	225.44	172.49	28.41
Swarna <i>Sub 1</i>	120.36	355.79	238.07	119.00
Nagina-22	169.10	210.54	189.82	24.50
Mapping population (Swarna <i>Sub 1</i> X Nagina22)				
1	134.50	196.40	165.45	46.02
2	124.00	230.20	177.10	85.64
3	116.00	242.50	179.25	109
4	142.00	227.20	184.60	60
5	120.00	220.12	170.06	83.43
6	127.36	250.10	188.73	96.37
7	130.50	182.40	156.45	39.77
8	118.45	190.45	154.45	60.78
9	123.60	220.00	171.80	77.99
10	125.50	225.20	175.35	79.68

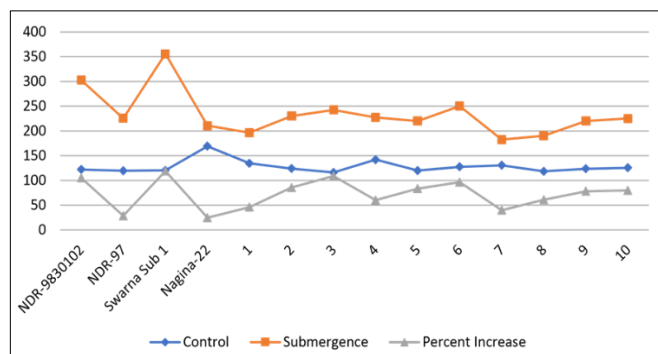


Fig 1: Catalase Activity ($\mu\text{g/g}$ Fresh Weight Mint^{-1})

Effect of submergence stress on peroxidase activity

Peroxidase was assayed according to the method of Hammerschmidt in 1959. The reaction mixture contained 0.1M phosphate buffer, 0.1M pyrogallol and 0.2% H_2O_2 . Enzyme activity is expressed as change in absorbance/minute/mg. protein. estimated value of Swarna *Sub 1* showed lowest (16.27%) reduction while Nagina 22 showed highest (17.56 %) reduction in case of mapping population plant number 2 has highest per cent reduction while plant number 1 has lowest percent reduction in comparison to plant number 3, 4, 5, 6, 7, 8, 9 and 10 in Table 3 and also in graph.

Ascorbate peroxidase (APX) together with the other enzymes contributes to the depletion of H_2O_2 level by utilizing ascorbate (AsA) as the specific electron donor for reducing H_2O_2 to water with the concomitant generation of monodehydroascorbate, which is disproportionate non-enzymatically to Ascorbate and dehydroascorbate (DHA). The DHA is subsequently reduced to Ascorbate by the action of enzyme dehydroascorbate reductase (DHAR), using reduced glutathione (GSH) as the reducing substrate. The resulting oxidized glutathione (GSSG) is then converted back to the reduced form (GSH) by NADPH-dependent enzyme glutathione reductase (GR). The DHA accumulation is generally considered as a negative event for cell metabolism. The probability of DHA accumulation was less in tolerant than in susceptible cultivars due to the higher activity of DHAR. The differences between susceptible and tolerant rice cultivars in the levels of reduced ascorbate during

submergence were non-significant (Das *et al.*, 2004; Panda and Sarkar, 2012a, 2013). Probably, tolerant cultivars recycled the Ascorbate more efficiently and protected the enzymes of ascorbate-glutathione cycle, with the maintenance of its threshold balance. Thus, the level of Ascorbate during submergence was the same between tolerant and susceptible cultivars. The higher activities of antioxidant enzymes APX, DHAR and GR in tolerant cultivars helped in better physiological adaptation to hypoxic condition, so as to maintain the structural integrity of enzyme molecules. When needed upon exposure to high oxygen concentration, these enzymes could detoxify the toxic oxygen species and protect the system effectively.

Table 3: Effect of submergence stress on peroxidase activity ($\mu\text{g/g}$ fresh weight) in rice genotypes

Parent	Control	Submergence	Mean	Percent Increase
NDR-9830102	253.98	419.06	336.52	65.00
NDR-97	242.45	284.50	263.47	17.34
Swarna <i>Sub 1</i>	300.56	510.95	405.75	70.00
Nagina-22	247.30	280.50	263.90	13.42
Mapping population				
1	250.63	356.40	303.51	42.20
2	240.28	402.00	321.14	67.30
3	262.50	386.42	324.46	47.20
4	245.00	399.10	322.05	62.89
5	250.46	420.15	335.30	67.75
6	260.50	350.20	305.35	34.43
7	243.01	412.06	327.53	69.56
8	265.80	370.50	368.15	39.39
9	270.40	406.80	338.60	50.44
10	268.45	386.50	327.47	43.97

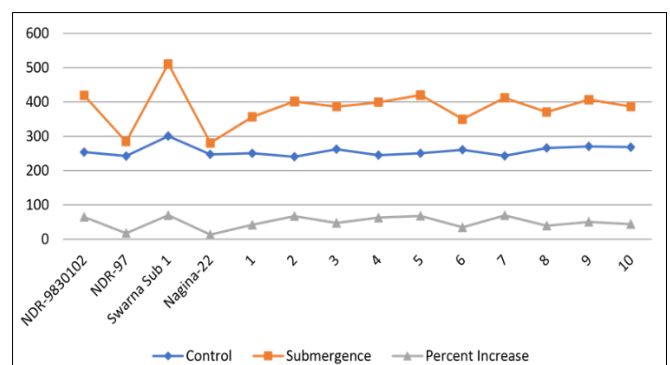


Fig 3: Peroxidase Activity ($\mu\text{g/g}$ Fresh Weight)

Effect of submergence stress on Chlorophyll content

Rice genotypes showed highest percent reduction in Nagina 22 (14.26%) in comparison to Swarna *Sub 1* (5.39%). Plant number 1 showed highest percent reduction (11.93%) and lowest in plant number 7 (5.20%) shown in graph given below. Chlorophyll concentration in leaves decreased under submergence and with increasing submergence duration from 12 to 15 days. The sensitive and tolerant genotypes had similarly high leaf chlorophyll concentrations before submergence but when submerged the tolerant genotypes maintained more chlorophyll than the intolerant genotypes. The Nagina 22 genotypes exhibited 10–20 % greater reduction in leaf chlorophyll concentration than did the tolerant checks and the *Sub1* introgression lines. The pairs of *Sub1* NILs did not differ significantly in leaf chlorophyll concentrations before submergence and under control conditions, yet concentrations were significantly higher in the *Sub1* genotypes following submergence.

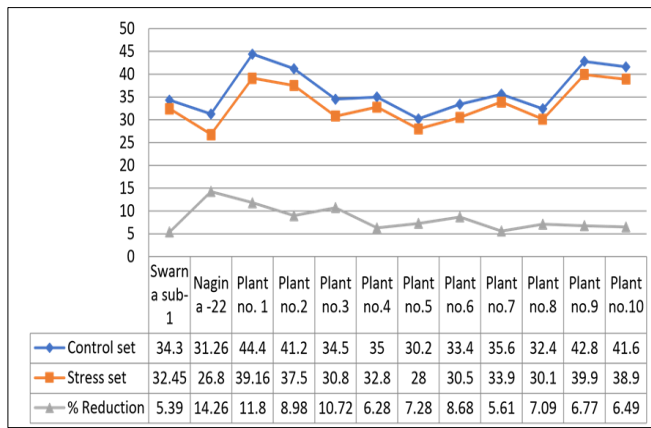


Fig 4: Effect on Chlorophyll Content (SPAD Value)

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