

ISSN Print: 2617-4693 ISSN Online: 2617-4707 IJABR 2024; 8(1): 424-430 www.biochemjournal.com Received: 05-11-2023 Accepted: 08-12-2023

#### Jitendra Kumar

Department of Crop Physiology, Acharya Narendra Deva University of Agriculture and Technology, Ayodhaya, Uttar Pradesh, India

#### Saurabh Singh

Department of Crop Physiology, Acharya Narendra Deva University of Agriculture and Technology, Ayodhaya, Uttar Pradesh, India

#### Rakesh Yadav

Department of Crop Physiology, Chandra Shekar Azad University of Agriculture and Technology, Kanpur, Uttar Pradesh, India

#### Jitender Bhati

Department of Genetics and Plant Breeding, Gochar Mahavidalaya, Rampur Maniharan, Saharanpur, Uttar Pradesh, India

#### Prakriti Chauhan

Department of Seed Science and Technology, Acharya Narendra Deva University of Agriculture and Technology, Ayodhaya, Uttar Pradesh, India

#### Surendra Singh

Department of Genetics and Plant Breeding, Gochar Mahavidalaya, Rampur Maniharan, Saharanpur, Uttar Pradesh, India

Corresponding Author: Saurabh Singh Department of Crop Physiology, Acharya Narendra Deva University of Agriculture and Technology, Ayodhaya, Uttar Pradesh, India

# Screening of rice genotypes (*Oryza sativa* L.) for submergence tolerance by physiological & biochemical approaches

## Jitendra Kumar, Saurabh Singh, Rakesh Yadav, Jitender Bhati, Prakriti Chauhan and Surendra Singh

#### DOI: https://doi.org/10.33545/26174693.2024.v8.i1f.491

#### Abstract

Seventeen genotypes including check SUB-106 and three replication were taken in the investigation and pots were arranges in the complete randomize design (CRD) for the study. After 25 days of sowing plant were submerged in outdoor flooded one meter deep water tank for 14 days and water level maintain the one meter from ground surface of the tank. One set of plants were kept outside as nonsubmerged control set. Before submergence at 25 DAS and after 14 days of submergence period, the data were taken on growth parameters like Dry weight of radical, Dry weight of plumule, Number of plants at 25 and 40 DAS, Dry weight (g) per plant at 25 and 40 DAS and biochemical parameters like  $\alpha$ -amylase enzyme activity in germinated seed at 7 DAG, total chlorophyll content and starch content in the leaves were recorded. Among the seventeen genotypes before submergence and after desubmergence significant genotypic variations were observed and significant reduction/increase in number of plants per pots, dry weight of radical, dry weight of plumule, dry weight per plant and biochemical parameters like  $\alpha$ -amylase enzyme activity in germinated seed at 7 DAG, total chlorophyll content and starch content in the leaves were recorded. The survival percentage, number of plant per pot and dry weight per plant, chlorophyll and starch data basis genotype SUB-102 is found better for submergence tolerant in comparison of check SUB-106 and SUB-113 is also moderate tolerant.

Keywords: *a*-amylase, chlorophyll, de-submergence, growth parameters, rice genotypes, submergence

#### Introduction

Rice (Oryza sativa L.) in family Graminae is a very important cereal food crop worldwide and is the primary stable food for over two billion people. It is the second most important crop of the world after wheat with more than 90% currently grown in Asia (Anonymous, 1992) <sup>[10]</sup>. Rice is highly valuable cash crop that earns substantial foreign exchange for the country. There are 24 species in the genus Oryza, including economically important wild species as well as cultivated species. Two important cultivated species, Oryza sativa L. and Oryza glabberrima L. have AA genomes (Vaughan et al., 2003)<sup>[11]</sup>. Rice (2n=24; estimated genome size =430 Mb) is a model experimental plant for the study of other cereal crops due to its diploid genetics. Flash-flooding and submergence adversely affect at least 16% of the rice lands of the world (~22 m ha). In eastern India, ~13 m ha of rice lands are unfavorably affected by excess water and periodically suffer from flash-floods and complete submergence. Drought and flooding are considered to be two of the most important abiotic stresses that affect rice production globally. A total of approximately 40 million hectares of rice area are affected by different forms of the two stresses occurring at different crop stages. High rainfall during the wet season (WS), overflowing rivers and canals, or high tides often flood farmland and adversely affect productivity in large areas of South and Southeast Asia. Rice is often the only crop capable of surviving under these conditions (Setter and Laureles 1995; Pucciariello and Perata 2013) <sup>[12-13]</sup> and some of the flood-prone environments are still planted with traditional rice (Oryza sativa) landraces that are moderately adapted to flooding. Submergence can be divided into two types, namely stagnant flooding and flash flooding. Stagnant flooding means that water stagnates for a longer duration, commonly more than a month, and occur in deep water and floating rice areas (Finch et al., 2006 and Fatima et al., 2016) [14-15].

Submergence stress can occur in all phases of growth from seedling to flowering plants with 1-2 weeks of flood duration (flash flood) (Mackill et al., 1993; Nugraha et al., 2013) <sup>[16-17]</sup>. Submergence stress can occur in flood-prone areas both in irrigation and rainfed areas. In Asia, rainfed lowland is widely distributed in South Asia such as India, Bangladesh, Nepal, and Southeast Asia (Singh et al., 2001) <sup>[4]</sup>, whereas in Indonesia, the type of lowland that most often suffered by submergence is shallow and middle swampland. The area of shallow and middle swampland in Indonesia covers about 7,512,080 ha, and it has great potential to be used for food crop production (Sarkar, 2006) <sup>[19]</sup>. An alternative strategy to reduce the impact of submergence stress is by planting tolerant rice varieties Shambhoo et al., 2014 <sup>[18]</sup>. The use of tolerant varieties will not increase production costs for farmers and production stability can be maintained (Septiningsih et al., 2009)<sup>[6]</sup>.

Waterlogging and floods cause substantial yield losses in cereal crops worldwide, aggravating poverty and food insecurity in developing countries. Climate change scenarios are predicting increases in future incidences and intensities of floods, especially in the tropics and subtropics. Most dry land cereals, such as maize, wheat, and barley, are sensitive to water logging, causing up to 20% yield losses in irrigated areas, and even greater losses in rainfed ecos systems exceeding 40%.

Water logging hampers root growth and function because of oxygen shortages that restrict root respiration. Climate change scenarios are predicting increases in future incidences and intensities of floods, especially in the tropics and subtropics. Complete submergence during seedling stage due to flooding can affect rice growth and yield in more than 16 million ha of deep-water rice areas throughout the world resulting in substantial economic losses to the farmers. Cultivars such as FR13A, are highly tolerant and survive up to two weeks of complete submergence owing to a major quantitative trait locus designated Submergence 1 (Sub1).

## **Materials and Methods**

The present study was conducted during 2019 in the Kharif Season. (1 Months and 25 days). The investigation on "Screening of Rice Genotypes (Oryza sativa L.) for Submergence Tolerance by physiological & biochemical approaches" was carried out under natural conditions in pots during Kharif season 2019-20. The pot experiment was conducted at net house in the Department of Crop Physiology. Some part of the experiment was carried out in the laboratory of Department of Crop Physiology, Acharya Narendra Deva University of Agriculture and Technology, Narendra Nagar, Kumarganj, Ayodhya (U.P.) during Kharif 2019-20. Complete submergence was applied for 14 days just after 25 DAS in adjoining cemented pond in the net house, Department of Crop Physiology, during Kharif 2019-20. The details of the materials used, experimental procedures and technologies adopted were described.

## Dry weight of radical (mg)

The randomly five selected seedlings for measuring radical length obtained after final count were dried at  $80\pm1$  <sup>0</sup>C for 24 hours in hot air oven, and dry weight of radical in milligrams (mg) was determined by weighing them in an electronic balance and average dry weight of radical was calculated from the replicated data.

## Dry weight of plumule (mg)

The randomly five selected seedlings for measuring plumule length obtained after final count were dried at  $70\pm1$  <sup>0</sup>C for 24 hours in hot air oven, and dry weight of plumule in milligrams (mg) was determined by weighing them in an electronic balance and average dry weight of plumule was calculated from the replicated data.

## Dry weight (g) per plant

Three healthy and uniform plants from each treatment were sampled and oven dried at  $80\pm1$  °C till a constant dry weight was achieved. The weight was recorded with the help of electronic balance. The average dry weight per plant was calculated from the replicated data.

#### Number of plants per pot

Number of plant was recorded by counting at each stage of observation average number of plant per pot was calculated from the replicated data.

## Chlorophyll content

Total chlorophyll content were estimated according to method of Arnon (1949)  $^{[21]}$  and expressed as (mg g  $^{-1}$  fresh wt.) of leaves.

## Total starch content

Starch content in the plant material was estimated by using the method of Mc. Cready *et al.*, (1958) <sup>[22]</sup>.

#### *α*-amylase activity

The amylase activity in embryo axis and endosperm after 48, 72 and 96hrs of germination was assayed according to the method of Bernfeld (1953)<sup>[20]</sup>.

## Results

## Dry weight of radical (mg)

Data regarding to radicle dry weight is presented in Table 1. The radical dry weight of the rice genotypes under laboratory condition were recorded and highly significant were noticed in the genotype SUB-117 (4.12 mg), SUB-117 (4.12 mg), SUB-113 (4.05 mg), SUB-102 (3.91 mg), SUB-116 (3.87 mg), SUB-103 (3.39 mg), SUB-104 (3.27 mg) and SUB-115 (3.15 mg) and followed by SUB-114 (2.89 mg) and SUB-111 (2.50 mg) over the check SUB-106 (2.29 mg), whereas the minimum non-significant value of radicle dry weight were observed in SUB-112 (1.80 mg) and followed by SUB-108 (2.10 mg), respectively over the check. The range of radical dry weight (mg) were between the 1.80 mg (SUB-112) to 4.12 mg (SUB-117). All the genotypes differed significantly with each other except SUB-105 (2.26 mg), SUB-106 (2.29 mg), SUB-107 (2.18 mg), SUB-108 (2.10 mg), SUB-109 (2.26 mg) and SUB-110 (2.16 mg). Hakim et al., (2010)<sup>[1]</sup>, also found significant genotypic variation on the plumule and radical length and plumule and radical dry weight of twelve rice genotypes.

## Dry weight of plumule (mg)

Data related to plumule dry weight are presented in Table 1. The plumule dry weight of the rice genotypes under laboratory condition are recorded and significantly highest plumule dry weight were noticed in the genotypes SUB-103 (23.08 mg), SUB-101 (18.71 mg), SUB-104 (16.68 mg), SUB-102 (15.97 mg), SUB-107 (15.04 mg), SUB-108 (14.74 mg), SUB-113 (14.68 mg) and SUB-115 (14.25 mg) over the check SUB-106 (11.85 mg), respectively and SUB-114 (12.51 mg) and SUB-109 (12.37 mg) were at par over the check. Whereas the minimum non-significant plumule dry weight were observed in SUB-111 (10.10 mg) and followed by SUB-112 (10.33 mg) over the check SUB-106 (11.85 mg), respectively. The range of the plumule dry weight of 17 genotypes were between the 10.10 (SUB-111) to 23.08 (SUB-103). Hakim *et al.*, (2010) <sup>[1]</sup>, also noticed significant genotypic variation on the plumule and radical length and plumule and radical dry weight of twelve rice genotypes.

<b>Table 1:</b> Genotypic variation in the dry weight (mg) of radical
(Root), dry weight (mg) of plumule (Stem) and of rice genotypes

	Dry weight (mg) of	Dry weight (mg) of
Constant	radical (Root)	plumule (Stem) Seedling <sup>-1</sup>
Genotypes	Seedling <sup>-1</sup> at 7 DAG	at 7 DAG
SUB-101	2.45	18.71
SUB-102	3.91	15.97
SUB-103	3.39	23.08
SUB-104	3.27	16.68
SUB-105	2.26	11.89
SUB-106(C)	2.29	11.85
SUB-107	2.18	15.04
SUB-108	2.10	14.74
SUB-109	2.26	12.37
SUB-110	2.16	11.00
SUB-111	2.50	10.10
SUB-112	1.80	10.33
SUB-113	4.05	14.68
SUB-114	2.89	12.51
SUB-115	3.15	14.25
SUB-116	3.87	11.00
SUB-117	4.12	12.04
C.D.at 5%	0.18	0.38
S.Em±	0.06	0.13

## Dry weight (mg) per plant

The data pertaining to dry weight of different genotypes before submergence and just after 14 days complete submergence at 40 DAS are presented in Table 2. The mean effect of submergence on dry weight of seventeen genotypes were showed significant reduction over the control set including check SUB-106. It is evident from the data that highest plant dry weight per plant showed by SUB-108, 175.7 mg at 25 DAS and 289.7 mg at 40 DAS over the seventeen genotypes in control set, while in treated set after de- submergence at 40 DAS, SUB-108 genotype was showed 189.7 mg plant dry weight per plant. On the other hand, minimum plant dry weight per plant was recorded in SUB-105 (87.5 mg) at 25 DAS and 142.8 mg at 40 DAS (control set), over the seventeen genotypes, while in treated set after de-submergence at 40 DAS this genotype was not survived and showed 100 percent mortality.

After 14 days complete submergence highest significant plant dry weight per plant were noticed in the SUB-102 (213.7 mg) at 40 DAS over the 17 genotypes including check SUB-106 (128.3) followed by SUB-108 (189.7 mg), SUB-113 (188.7 mg) and SUB-103 (168.3 mg). Genotypes SUB-102 registered lowest reduction in dry weight (23%) over the 17 genotypes including check SUB-106 (27%) in treated set after de- submergence. The highest significant reduction in plant dry weight per plant was registered in SUB-117 (60%) followed by SUB-107 (55%) and SUB-114 (50%), respectively over the ten genotypes including check.

While genotypes SUB-101, SUB-105, SUB-109, SUB-110, SUB-111, SUB-112 and SUB-116 showed 100 percent mortality after de- submergence at 40 DAS in treated set. In case of genotypic differences without given any treatment at 40 DAS highest increase in dry weight per plant was noticed in the SUB-102 (79%) over all the 17 seventeen genotypes including check SUB-106 (78%). However, minimum increase in dry weight per plant was noted in SUB-116 (41%), over all the 17 genotypes followed by SUB-112 (42%) and SUB-109 (45%), respectively in control set. They would help in better survival than those genotypes having lower dry matter before submergence Chaturvedi et al., (1996)<sup>[2]</sup>, Singh (1997)<sup>[3]</sup>, Singh *et al.*, (2001)<sup>[4]</sup> and Sarkar et al., (2001)<sup>[5]</sup> also reported similar result. Singh et al. (2001)<sup>[4]</sup> noted the percentage survival of the seedlings was positively correlated with the increase in shoot dry weight during submergence.

## Number of Plant Pot<sup>-1</sup> (count)

The data pertaining to number of plant pot<sup>-1</sup> before submergence at 25 and 40 DAS (control set) and when impose the 14 days complete submergence at 25 DAS and taken the data at 40 DAS (treated set) are tabulated in Table 2. Under submergence treatment, all the genotypes registered significant reduction in number of plant pot<sup>-1</sup> over control set. However, highest number of plant were survive in SUB- 102 (11.3 and 56%) over check SUB-106 (11.0 and 55%), but it was not significant difference. Among the 17 genotypes in respect to number of plant per pot and survival percent in genotypes SUB-103 (707 & 39%), 113 (8.0 & 41%), 114 (8.3 & 41%) and 117 (8.3 & 41%) showed ignificant reduction in number of plant pot<sup>-1</sup> over the check SUB-106 (11.0 & 55%), respectively. Rest genotypes *i.e.*, SUB-102 (11.3 & 56%), SUB-104 (10.7 & 53%) were at par with check SUB-106 (11.0 & 55%). On the other hand, SUB-101, SUB-105, SUB-109, SUB-110, SUB-111, SUB-112 and SUB-116 were showed 100 mortality. These results were strongly supported by Septiningsih et al., (2009) [6] they work done in the greenhouse or in the field at the International Rice Research Institute (IRRI), Los Banos, Philippines.

## Survival Percentage (%)

Data regarding percent survival of rice genotypes as affected by 14 days complete submergence treatment have been presented in table 2. It is clear from the data that the survival of all the genotypes were affected significantly under submergence condition over the control set. Plant population subject to submergence treatment significantly reduced at 40 DAS in comparison to 25 DAS control set. The number of plant were surviving under 14 days complete submerged condition were recorded at 40 DAS and survival percent were calculated based on the number of plant at 25 DAS. After submergence, the recovery expressed as survival percentage of respective values just after de-submergence. The maximum survival percentage was noticed in SUB-

102 (56%) over the check SUB-106 (55%). Significant reduction in survival percent were recorded in SUB-103 (39%), SUB-113(41%), SUB-114 (41%), SUB-117 (41%), SUB-108 (45%), SUB-107 (46%) and SUB-115 (46%) over the check SUB-106 (55), respectively. The mean effect of submergence on genotypes was found significant. Among the seventeen genotypes, only ten genotypes found success to survive and survival ranged was 41 percent (SUB-113) to 56% (SUB-102) and seven genotypes (SUB-101, SUB-105, SUB-109, SUB-110, SUB-111, SUB- 112 and SUB-116) showed 100 percent mortality at 40 DAS (treated set). Singh

*et al.*, 2001 <sup>[4]</sup> also reported that water shoot elongation as desirable trait for submergence.

	Number of plant pot <sup>-1</sup> at different growth stags				
Construes	At 25 DAS	At 40 DAS	A+ 40 DAS (AS)	Survival (%) at 40 DAS (AD) over	Mortality % at 40 DAS (AD) over
Genotypes	(Control-WS) (Control-WS)		AI 40 DAS (AS)	control (40 DAS)	control (40 DAS)
SUB-101	20.0	20.0	-	0	100
SUB-102	20.0	20.0	11.3	56	44
SUB-103	19.7	19.7	7.7	39	61
SUB-104	20.0	20.0	10.7	53	47
SUB-105	20.0	20.0	-	0	100
SUB-106 (C)	20.0	20.0	11.0	55	45
SUB-107	20.0	20.0	9.3	46	54
SUB-108	20.0	20.0	9.0	45	55
SUB-109	19.7	19.7	-	0	100
SUB-110	20.0	20.0	-	0	100
SUB-111	20.0	20.0	-	0	100
SUB-112	20.0	20.0	-	0	100
SUB-113	19.5	19.5	8.0	41	59
SUB-114	20.0	20.0	8.3	41	59
SUB-115	20.0	20.0	9.3	46	54
SUB-116	19.0	19.0	-	0	100
SUB-117	20.0	20.0	8.3	41	59
C.D.at 5%	N.S.	N.S.	2.3	2.3	-
S.Em±	0.3	0.3	0.8	0.8	-

WS= without submergence, AD= After De-submergence

#### **Biochemical Parameters**

## Total Chlorophyll Contents (mg g<sup>-1</sup> fresh wt.)

The data regarding to total chlorophyll content of different genotypes before submergence and after de-submergence are presented in Table 3. The mean effect of submergence on total chlorophyll content of seventeen genotypes were showed significant reduction over the control set including check SUB-106. It is evident from the data that highest total chlorophyll content showed by SUB-112 (6.67 mg g<sup>-1</sup> fresh weight) at 25 DAS and 6.99 mg g<sup>-1</sup> fresh weight at 40 DAS in control set over the seventeen genotypes, when at 25 DAS impose the 14 days complete submergence and taken the data at 40 DAS after de-submergence, genotype SUB-112 showed 100 percent mortality. On the other hand, minimum total chlorophyll content was recorded in SUB-101 (3.43 mg g<sup>-1</sup> fresh weight) at 25 DAS and SUB-102 at 40 DAS, showed minimum values 4.15 mg g<sup>-1</sup> fresh weight in control set, over the seventeen genotypes, while after desubmergence at 40 DAS SUB-101 genotype was not survived and showed 100 percent mortality. After desubmergence at 40 DAS minimum total chlorophyll content was recorded in SUB-117 (0.92 mg g<sup>-1</sup> fresh weight) followed by SUB-115 (1.07 mg g<sup>-1</sup> fresh weight) and SUB-114 (1.08 mg g<sup>-1</sup> fresh weight) respectively over ten survived genotypes after de-submergence. When at 25 DAS impose the 14 days complete submergence and taken the data after de- submergence at 40 DAS, all genotypes showed non-significant effect on total chlorophyll content over the check SUB-106 (2.58 mg  $g^{-1}$  fresh weight).

When impose the 14 days complete submergence at 25 DAS and taken the data after de-submergence at 40 DAS, genotypes SUB-102 registered lowest significant reduction (53%) in total chlorophyll content over the 17 genotypes including check SUB-106 (62%). The highest significant reduction in total chlorophyll content was registered in SUB- 107 (84%) followed by SUB-117 (83%) and SUB-114 (78%), respectively over the ten genotypes, while genotypes SUB-101, SUB- 105, SUB-109, SUB-110, SUB-111, SUB-112 and SUB-116 showed 100

percent mortality at 40 DAS in submerged set. In case of genotypic differences without given any treatment at 40 DAS highest total chlorophyll content was noticed in the SUB-107 (6.99 mg g<sup>-1</sup> fresh weight) over all the 17 seventeen genotypes including check SUB-106 (6.72 mg g<sup>-1</sup> fresh weight), however, minimum total chlorophyll content was noted in SUB-102 over all the 17 genotypes followed by SUB-115 (4.16 mg g<sup>-1</sup> fresh weight) and SUB-110 (4.18 mg g<sup>-1</sup> fresh weight), respectively. In case of 25 DAS to 40 DAS (control set) progressive increase in total chlorophyll content was noted and it ranged were 4 percent (SUB-108 & SUB-116) to 31 percent (SUB-101). This increases the chance of survival as intact chlorophyll allows plants to generate more energy through underwater photosynthesis and to resume active growth during recovery from submergence (Vergara and Ismail, 2004)<sup>[7]</sup>.

Table 3: Effect of submergence on total chlorophyll content (mg g<sup>-1</sup> fresh wt.) in leaf of rice genotypes

	Chlorophyll content (mg g <sup>-1</sup> fresh wt.)				
Genotypes	At 25 DAS (Control-WS)	At 40 DAS (Control-WS)	% increased at 40 DAS over 25 DAS	At 40 DAS (AD)	% reduction at 40 DAS (AD) over control at 40 DAS
SUB-101	3.43	4.43	31	-	-
SUB-102	3.52	4.15	19	1.97	53
SUB-103	6.13	6.78	11	2.15	68

International Journal of Advanced Biochemistry Research

SUB-104	4.75	4.89	5	1.43	71
SUB-105	4.54	4.98	12	-	-
SUB-106 (C)	6.42	6.72	5	2.58	62
SUB-107	6.57	6.99	8	1.09	84
SUB-108	5.91	6.15	4	2.15	65
SUB-109	4.92	5.13	5	-	-
SUB-110	4.08	4.18	5	-	-
SUB-111	6.33	6.78	8	-	-
SUB-112	6.67	6.98	6	-	-
SUB-113	5.43	5.67	6	1.98	65
SUB-114	4.25	4.85	15	1.08	78
SUB-115	3.67	4.16	16	1.07	74
SUB-116	5.84	5.98	4	-	-
SUB-117	5.15	5.54	9	0.92	83
C.D.at 5%	2.26	3.40	-	1.42	4.4
S.Em±	0.75	1.13	-	0.50	1.5

WS=Without submergence, AD= After De-submergence

## **Starch Content (mg g<sup>-1</sup> dry wt.)**

The data pertaining to total starch content in control set at 25 and 40 DAS and just after de-submergence at 40 DAS, are presented in Table 4. The mean effect of submergence on all seventeen genotypes was found significant effect. It is evident from the data that highest significant total starch content was noticed in genotype SUB-104 (105.3 & 234.7 mg g<sup>-1</sup> dry wt.) among all the genotypes at 25 and 40 DAS in control set, except after de-submergence at 40 DAS (44.7 mg g<sup>-1</sup> dry wt.), followed by SUB-103 (103.9 & 214.2 mg g<sup>-1</sup> <sup>1</sup> dry wt.) respectively. While minimum total starch content was recorded in SUB- 109 (90.8 mg g<sup>-1</sup> dry wt.) at 25 DAS and at 40 DAS (163.5 mg g<sup>-1</sup> dry wt.) in control set it was significantly differ from check genotypes SUB- 106 (98.2 & 175.9 mg g<sup>-1</sup> dry wt.), respectively, on the other hand, SUB-109 genotype was not survive after de-submergence and showed 100 mortality per cent. After de-submergence, significantly highest total starch content were noticed in the SUB-102 (115.5 mg g<sup>-1</sup> dry wt.) and SUB-108 (102.5 mg g<sup>-1</sup> dry wt.) followed by SUB-113 (97.3 mg g<sup>-1</sup> dry wt.), respectively over the check SUB-106 (94.7 mg g<sup>-1</sup> dry wt.). Genotypes SUB-104 registered highest significant increase (122.8%) in total starch content at 40 DAS under control condition over the 25 DAS, followed by SUB-114 (106.6%) and SUB-110 (106.3), respectively. However, minimum increase in total starch content at 40 DAS in control set was noted in SUB-105 (78.1%) followed by SUB-106 over the control set of 25 DAS. After de-submergence genotypes SUB-103 (58.7 mg g<sup>-1</sup> dry wt.), SUB-104 (44.7 mg g<sup>-1</sup> dry wt.), SUB-107 (23.3 mg g<sup>-1</sup> dry wt.), SUB-114 (36.7 mg g<sup>-1</sup> dry wt.), SUB-115 (41.0 mg g<sup>-1</sup> dry wt.) and SUB- 117 (24.7 mg g<sup>-1</sup> dry wt.), were not found significant, over the check SUB-106 (94.7 mg g<sup>-1</sup> dry wt.). However, minimum increase in total

starch content was noted in SUB-117 (74.7%), at 40 DAS, over control set of 25 DAS. Genotypes SUB-102 registered lowest reduction in total starch content (44%) over the 17 genotypes including check SUB-106 (46%) after desubmergence at 40 DAS. The highest reduction in total starch content was registered in SUB-117 (86%) followed by SUB-107 (83%) and SUB-114 (81%), respectively over the ten genotypes, while genotypes SUB-101, SUB-105, SUB-109, SUB-110, SUB-111, SUB-112

and SUB-116 showed 100 percent mortality, respectively at 40 DAS after de-submergence. This finding is well supported by Das *et al.*, (2009) <sup>[8]</sup>, found that, rate of decreasing levels of total sugar and starch content in rice plants during complete submergence could be considered as an indicator of submergence tolerance as revealed through repeated experiments with two submergence tolerant and two susceptible varieties.

	Starch content (mg g <sup>-1</sup> dry wt.)					
Genotypes	At 25 DAS (Control-WS)	At 40 DAS (Control-WS)	% increased at 40 DAS over 25 DAS	At 40 DAS (AD)	% reduction at 40 DAS (AD) over control at 40 DAS	
SUB-101	103.4	199.4	92.9	-	-	
SUB-102	102.7	200.4	95.2	115.5	44	
SUB-103	103.9	214.2	106.1	58.7	73	
SUB-104	105.3	234.7	122.8	44.7	81	
SUB-105	104.7	186.5	78.1	-	-	
SUB-106 (C)	98.2	175.9	79.1	94.7	46	
SUB-107	100.3	189.5	88.9	32.3	83	
SUB-108	100.9	198.6	96.8	102.5	48	
SUB-109	90.8	163.5	80.1	-	-	
SUB-110	95.9	197.8	106.3	-	-	
SUB-111	102.3	190.2	85.9	-	-	
SUB-112	97.5	185.3	90.0	-	-	
SUB-113	94.2	176.9	87.7	97.3	45	
SUB-114	93.8	193.8	106.6	36.7	81	
SUB-115	100.8	199.5	98.0	41.0	79	
SUB-116	104.4	189.4	81.4	-	-	
SUB-117	103.7	181.2	74.7	24.7	86	

C.D.at 5%	3.8	9.5	-	6.8	1.3
S.Em±	1.3	3.1	-	2.3	0.4

WS= without submergence, AD= After De-submergence

# $\alpha$ -amylase activity in treated seeds (mg/g fresh weight)

It is evident from data that  $\alpha$ -amylase activity in germinated seeds of rice at 7 DAS was estimated and shown in table 5. Data recorded on  $\alpha$ -amylase activity in germinated seeds at 7 DAG, it ranges from 0.246 (SUB-108) to 1.327 SUB-101) mg per g fresh weight. Significant variation were noticed among the genotypes. Genotypes SUB-101 (1.327 mg g<sup>-1</sup>) showed the significant higher values over the check SUB-106 (0.793) followed by SUB-104 (0.941 mg per g fresh weight). Genotypes SUB-102, SUB-103, SUB-105, SUB-107, SUB-108, SUB-109, SUB-110, SUB-111, SUB-112, SUB-113, SUB-114, SUB-116 and SUB-117 showed the non-significant effect over the check SUB-106 (0.793 mg per g fresh weight), respectively. Genotypes SUB-115 (0.831 mg g<sup>-1</sup> fresh weight) showed the higher values of  $\alpha$ amylase activity in germinated seeds over check SUB-106  $(0.793 \text{ mg g}^{-1} \text{ fresh weight})$ , but it was not significant higher over the check genotype. Minimum a-amylase activity was recorded in minimum Sub-108 (0.246 mg g<sup>-1</sup> fresh weight), followed by SUB-109 (0.341 mg g<sup>-1</sup> fresh weight), respectively.

Similar result were reported by Damaris *et al.*, (2019) <sup>[9]</sup> observed that,  $\alpha$ -amylase is the major form of amylase with secondary carbohydrate binding sites, is a crucial enzyme throughout the growth period and life cycle of angiosperm.

Table 5: Effect of submergence on ∝ amylase activity (mg g<sup>-1</sup> fresh wt.) in germinated seed of rice genotypes

Genotypes	a-amylase enzyme activity (mg g <sup>-1</sup> fresh wt.) in germinated seed at 7 (DAG)
SUB-101	1.327
SUB-102	0.428
SUB-103	0.626
SUB-104	0.941
SUB-105	0.426
SUB-106(C)	0.793
SUB-107	0.682
SUB-108	0.246
SUB-109	0.341
SUB-110	0.681
SUB-111	0.570
SUB-112	0.389
SUB-113	0.473
SUB-114	0.536
SUB-115	0.831
SUB-116	0.718
SUB-117	0.743
C.D.at 5%	0.062
S.Em±	0.021

#### Conclusion

- Submergence tolerant genotypes check SUB-106 (c) and another one SUB-102 had higher plant survival, dry weight of plants, chlorophyll and starch content. After submergence as compared to susceptible genotypes. These tolerant genotypes maintained higher dry matter even during submergence which was possible reason for better survival after de- submergence.
- On the basis of survival percentage, number of plant per pot and dry weight per plant, chlorophyll and starch

data basis genotype SUB-102 is found better for submergence tolerant in comparison of check SUB- 106 and SUB-113 is also moderate tolerant.

- After de-submergence significantly decreased in chlorophyll content in the seven genotypes out of ten survived genotypes. The maximum reduction in higher plant survival percentage, dry weight of plants, starch content and chlorophyll content due to submergence were noted in susceptible genotypes SUB-117, SUB-107,SUB-104, SUB-114, SUB-115, SUB-103 and SUB-108 in comparison to tolerant genotypes.
- Tolerant genotypes had high total carbohydrate content during submergence and stored carbohydrate is utilized for regeneration after de-submergence.

#### References

- 1. Hakim MA, Juraimi AS, Begum M, Hanafi R, Ismail, Selamat A. Effect of salt stress on germination and early seedling growth of rice (*Oryza sativa* L.). African Journal of Biotechnology. 2010;9(13):1911-1918.
- 2. Chaturvedi GS, Singh BB, Singh AK, Kumar M, Dwivedi, JL, Singh MP, *et al.* Flood tolerance in crops: Research Accomplishment of Indian Scientists. Journal of Plant Biology. 1996;30(2):229-240.
- Singh HP. Physiological and biochemical basis for tolerance and susceptibility to submergence in rainfed lowland rice. NDUAT, Kumarganj, Faizabad, India. 1997;66:425-434.
- 4. Singh HP, Singh BB, Ram PC. Submergence tolerance of rainfed lowland rice: Search for marker traits. Journal of Plant Physiology. 2001;158:883-889.
- 5. Sarkar RK, Das S, Ravi I. Changes in certain antioxidative enzymes and growth parameters as a result of complete submergence and subsequent reaeration of rice cultivars differing in submergence tolerance. Journal of Agronomy and Crop Science. 2001;187:69-74.
- 6. Septiningsih EM, Pamplona AM, Sanchez DL, Neeraja CN, Vergara G, Heuer VS, *et al.* Development of submergence-tolerant rice cultivars: the Sub1 locus and beyond. Annals of Botany. 2009;103:151-160.
- Vergara A, Ismail D. Natural variation of submergence tolerance among *Arabidopsis thaliana* accessions. New Phytologist. 2004;190:299-310.
- 8. Das BP, Dash P, Roy AT. Role of total sugar and starch content of rice seedlings at different ages in variable submergence tolerance. *Oryza*. 2009;46(4):304-309.
- Damaris Z, Rebecca N, Lin P, Yang, Dongli H. The Rice Alpha-Amylase, Conserved Regulator of Seed Maturation and Germination. International Journal of Molecular Sciences. 2019;20:450.
- Anonymous BM, Gibson P, Edema R, Rena J. Identification of rice genotypes tolerant to submergence at the seedling stage in Uganda. Journal of Plant Breeding and Crop Science. 1992;11(72):425-434.
- 11. Vaughan G, Ismail AM. The mechanism of submergence tolerance. Rice Today. 2003;82:425-434.
- 12. Setter TL, Tuong TP, Ingram KT. Environmental characterization requirements for strategic research in rice grown under adverse conditions of drought,

flooding, and salinity. In: Ingram KT, editor. Rainfed Lowland Rice: Agricultural Research for High-Risk Environments. International Rice Research Institute, Manila, Philippines; c1995. p. 3-18.

- Pucciariello B, Perata S. Study of the Relationship between Carbohydrate Components of Stem and Survival % by Genetic Correlation and Path Analysis for Submergence Tolerance in Rice (*Oryza sativa* L.). Research Journal of Recent Science. 2013;5(8):1-10.
- 14. Finch-Savage K, William E, Leubner-Metzger, Gerhard G. Seed dormancy and the control of germination. New Phytologist. 2006;171:501-523.
- 15. Fatima AL, Ansari, Taoufik K. A Quantitative Assessment of Germination Parameters: the Case of *Crotalaria Persica* and *Tephrosia Apollinea*. Open Ecology Journal. 2016;9:13-21.
- Mackill M, Jr, PA, Patrick, Jr. Effect of zinc on alcohol dehydrogenase activity and nutrient uptake in rice. Agronomy Journal. 1993;80:882-885.
- Nugraha N, Ito O, Jun-Ichi S. Relationship between Shoot Elongation and Dry Matter Weight During Submergence in *Oryza sativa* L. and *O. glaberrima* Steud. Rice Cultivar. Plant Production Science. 2013;11(3):316-323.
- Shambhoo P, Bindu P, Garima Y, Deepak K, Tiwari A, Poonam K, Arun K, *et al.* Effect of alkalinity on germination efficiency index, seedling growth, free proline, and sugar during early seedling growth of rice. African Journal of Agricultural Research. 2014;9(44):3251-3257.
- 19. Sarkar S, Pressey RL, Faith DP, Margules CR, Fuller T, Stoms DM, *et al.* Biodiversity conservation planning tools: present status and challenges for the future. Annu. Rev. Environ. Resour. 2006 Nov 21;31:123-59.
- 20. Bernfeld S. On psychoanalytic training. The psychoanalytic quarterly. 1962 Oct 1;31(4):453-82.
- 21. Arnon DI. Copper enzymes in isolated chloroplasts. Polyphenoloxidase in Beta vulgaris. Plant physiology. 1949 Jan;24(1):1.
- 22. Gee M, McComb EA, McCready RM. A method for the characterization of pectic substances in some fruit and sugar-beet marcs. Journal of Food Science. 1958 Jan;23(1):72-5.