MORPHOPHYSIOLOGICAL AND YIELD RESPONSES OF RICE PLANT (*Oryza sativa* L.) UNDER SALT STRESS AND MITIGATION BY CALCIUM

SOULIN AKIV

DEPARTMENT OF AGRICULTURAL BOTANY
SHER-E-BANGLA AGRICULTURAL UNIVERSITY
DHAKA -1207

JUNE, 2018

MORPHOPHYSIOLOGICAL AND YIELD RESPONSES OF RICE PLANT (*Oryza sativa* L.) UNDER SALT STRESS AND MITIGATION BY CALCIUM

A Thesis
By

SOULIN AKIV

Reg. No. 12-04986

Submitted to the Department of Agricultural Botany
Sher-e-Bangla Agricultural University, Dhaka
in partial fulfillment of the requirements
for the degree of

MASTER OF SCIENCE (M.S.)
IN
AGRICULTURAL BOTANY

SEMESTER: JULY-DECEMBER, 2017

APPROVED BY:

.....	.....
(Professor Dr. Kamal Uddin Ahamed)	**(Dr. Kamrun Nahar, Associate Professor)**
Supervisor	**Co-supervisor**

.....
(Dr. Kamrun Nahar, Associate Professor)
Chairman
Examination Committee

DEPARTMENT OF AGRICULTURAL BOTANY

Sher-e-Bangla Agricultural University

Sher-e-Bangla Nagar, Dhaka-1207

Memo No: SAU/ABOT/……..

CERTIFICATE

This is to certify that the thesis entitled **'MORPHOPHYSIOLOGICAL AND YIELD RESPONSES OF RICE PLANT (*Oryza sativa* L.) UNDER SALT STRESS AND MITIGATION BY CALCIUM'** submitted to the Department of Agricultural Botany, Sher-e-Bangla Agricultural University, Dhaka, in partial fulfillment of the requirements for the degree of **MASTER OF SCIENCE in AGRICULTURAL BOTANY**, embodies the results of a piece of *bona fide* research work carried out by **Soulin Akiv**, Registration No. **12-04986** under my supervision and guidance. No part of the thesis has been submitted for any other degree or diploma.

I further certify that any help or source of information, received during the course of this investigation has been duly acknowledged.

Dated:
Place: Dhaka, Bangladesh

Prof. Dr. Kamal Uddin Ahamed
Department of Agricultural Botany
Sher-e-Bangla Agricultural University
Supervisor

DEPARTMENT OF AGRICULTURAL BOTANY

Sher-e-Bangla Agricultural University

Sher-e-Bangla Nagar, Dhaka-1207

DEDICATED
TO
MY BELOVED PARENTS

ACKNOWLEDGEMENTS

*The author first wants to articulate his enormous wisdom of kindness to the Almighty "**ALLAH**" for His never ending blessing, protection, regulation, perception and assent to successfully completion of research and preparation of thesis.*

*The author would like to express his deepest sense of gratitude to his research supervisor, **Professor Dr. Kamal Uddin Ahamed,** Department of Agricultural Botany, Sher-e-Bangla Agricultural University, Sher-e-Bangla Nagar, Dhaka-1207, for his scholastic guidance, innovative suggestion, constant supervision and inspiration, valuable advice and helpful criticism in carrying out the research work and preparation of this manuscript.*

*The author also grateful to his respected Co-Supervisor **Dr. Kamrun Nahar, Associate Professor,**Department of Agricultural Botany, Sher-e-Bangla Agricultural University, Sher-e-Bangla Nagar, Dhaka-1207, for constant encouragement, cordial suggestions, constructive criticisms and valuable advice during the research period and preparing the thesis.*

The author would like to express his deepest respect and boundless gratitude to his honorable teachers of Department of Agricultural Botany, Sher-e-Bangla Agricultural University, Sher-e-Bangla Nagar, Dhaka-1207, for their valuable teaching, sympathetic co-operation, direct and indirect advice, suggestions and inspirations during the whole study period.

The author would like to express his cordial thanks to the departmental and field staffs for their active help during the experimental period.

The author also wishes to extend sincere gratitude to the Ministry of Science and Technology, Government of the People's Republic of Bangladesh for providing him financial support to conduct the research.

*The author would like to thank **Ripon Kumar Roy**, **Md. Jahedul Alam Forhad, Swarna Sultana Jenni, Sadiya Zaman and Anupama Kanchi Suranjana Saroj** who have helped him so much during the research work and preparation of the thesis.*

The author expresses his unfathomable tributes, sincere gratitude and heartfelt indebtedness from his core of heart to his father ***Rafiqul Islam*** *and mother* ***Nasima Islam*** *whose blessings, inspirations, sacrifices and moral supports opened the gate and paved to way of his higher study.*

December, 2018 ***The Author***

SAU, Dhaka

MORPHOPHYSIOLOGICAL AND YIELD RESPONSES OF RICE PLANT (*Oryza sativa* L.) UNDER SALT STRESS AND MITIGATION BY CALCIUM

ABSTRACT

A pot experiment was conducted in the Net House of Department of Agricultural Botany of Sher-e-Bangla Agricultural University, Dhaka, during the Boro season of (December-June) 2017-2018 to evaluate the response of BRRI dhan67 to calcium supplementation at different salinity levels. The two factors experiment was laid out in Completely Randomized Design (CRD) with three replications. Factor A is different levels of salinity induced by sodium (Na^+) viz. 0, 4, 6 and 8 dSm^{-1} and factor B is different concentration of Ca^{2+} viz. 0, 80, 160 ppm. The total treatment combinations were 12 (4×3). The experimental results showed that salt stress significantly affects morphology, physiology, yield contributing characters and yield of rice. Plant height and tiller number per plant were reduced with increased levels of salinity mostly at 6 and 8 dSm^{-1}. Salinity also adversely affected the leaf, root and shoot dry weight (gm), leaf area (cm^2), leaf chlorophyll content, leaf relative water content, leaf membrane stability, days to flowering, number of panicle $plant^{-1}$, panicle length (cm), number of filled grains $panicle^{-1}$, number of unfilled grains $panicle^{-1}$, 1000-grain weight (g) and also grain yield g $plant^{-1}$ mostly at 8 dSm^{-1}. Exogenous application of Ca^{2+} significantly mitigates the adverse effects of salinity on plant biomass production or morphology, physiology and yield, leaf, root and shoot dry weight (g), leaf area (cm^2), leaf chlorophyll content, leaf relative water content, leaf membrane stability, number of panicle $plant^{-1}$, panicle length (cm), number of filled grains $panicle^{-1}$, number of unfilled grains $panicle^{-1}$, 1000-grain weight (g) and grain yield (g) $plant^{-1}$ were increased and days required for flowering decreased with the application of calcium than the control or without calcium. The yield of rice is gradually decreasing with the increasing levels of salinity. Interestingly, the rate of reduction of yield of rice was decreased with Ca^{2+} in response to different saline conditions and the lowest yield was recorded at the highest salinity (8 dSm^{-1}) along without Ca^{2+}. The present study also showed that the highest yield recorded without salt and 160 ppm Ca^{2+} treatment combination which was statistically dissimilar to control treatment combination in most of the cases. Application of both 80 and 160 ppm of calcium showed mitigating effect against salt stress but 160 ppm of calcium application showed better result than 80 ppm of calcium with or without salt (0, 4, 6, 8 dSm^{-1} level of salt) in every cases. These results are consistent with the findings of regulation of ion uptake in presence or absence of Ca^{2+} at different levels of Na^+ stress. Therefore, this experiment suggests that Ca^{2+} can effectively mitigate the deleterious effect of Na^+ stress in rice cultivation and increase the potential salt stress tolerance in BRRI dhan67.

CONTENTS

CHAPTER	TITLE	PAGE NO.
	ACKNOWLEDGEMENTS	i-ii
	ABSTRACT	iii
	LIST OF CONTENTS	iv-v
	LIST OF TABLES	vi
	LIST OF FIGURES	vii-ix
	LIST OF APPENDICES	x
	LIST OF ABBREVIATIONS	xi-xii
CHAPTER 1	**INTRODUCTION**	1-3
CHAPTER 2	**REVIEW OF LITERATURE**	4-21
2.1	Rice	4-5
2.2	Abiotic stress	5-6
2.3	Salt stress	6-7
2.4	Effect of salt stress on plant	8-13
2.5	Effect of salt stress on rice plants	14-16
2.6	Calcium	16-21
CHAPTER 3	**MATERIALS AND METHODS**	22-28
3.1	Experimental location	22
3.2	Soil	22
3.3	Climate	22
3.4	Description of the rice variety	23
3.5	Layout of the experiment	23
3.6	Treatments	23-24
3.7	Seed collection	24
3.8	Pot Preparation	24
3.9	Fertilizer Application	24
3.10	Sowing of seeds in seedbed	25
3.11	Uprooting and transplanting of seedlings	25
3.12	Intercultural operations	25

CHAPTER	TITLE	PAGE NO.
3.12.1	Weeding and irrigation	25
3.12.2	Plant protection measures	25
3.13	Harvesting	25
3.14	Collection of data	25-28
3.15	Statistical analysis	28
CHAPTER 4	**RESULTS AND DISCUSSION**	29-66
4.1	Plant height(cm)	29-33
4.2	Number of tillers plant^{-1}	33-36
4.3	Number of effective tillers plant^{-1}	37-39
4.4	Number of non-effective tillers plant^{-1}	39-41
4.5	Days to flowering (DAT- Days after transplanting)	41-43
4.6	Panicle length (cm)	43-44
4.7	Leaf Area(cm^2)	45-47
4.8	Relative water content (RWC)	47-49
4.9	Leaf Membrane Stability Index(%)	49-51
4.10	Chlorophyll content (SPAD Value)	51-54
4.11	Number of filled grains panicle^{-1}	54-57
4.12	Number of unfilled grain panicle^{-1}	57-58
4.13	1000- grain weight (g)	59-60
4.14	Grain yield g plant^{-1}	60-63
4.15	Leaf, Root, Shoot Dry Weight plant^{-1} (g)	63-66
CHAPTER 5	**SUMMARY AND CONCLUSION**	67-71
CHAPTER 6	**REFERENCES**	72-90
	APPENDICES	91-95

LIST OF TABLES

TABLE NO.	TITLE	PAGE NO.
4.1	Interaction effect of different salt concentrations and calcium levels on the plant height of rice at different days after transplanting	32
4.2	Interaction effect of different salt concentrations and calcium levels on the tillers plant^{-1} of rice at different days after transplanting	36
4.3	Interaction effect of different salt concentrations and calcium levels on the effective tillers plant^{-1}, ineffective tillers plant^{-1}, days to flowering and panicle length of rice	39
4.4	Interaction effect of different salt concentrations and calcium levels on the leaf area, relative water content (%) and leaf membrane stability of rice	47
4.5	Interaction effect of different salt concentrations and calcium levels on the SPAD value of rice at different days after transplanting	54
4.6	Interaction effect of different salt concentrations and calcium levels on the filled grains panicle^{-1}, unfilled grains panicle^{-1}, 1000 grain weigh and grain yield plant^{-1} of rice	56
4.7	Effect of different salt concentrations and calcium levels on the leaf dry weight plant^{-1}, root dry weight plant^{-1}, shoots dry weight plant^{-1}and total dry weight plant^{-1} of rice	64
4.8	Interaction effect of different salt concentrations and calcium levels on the leaf dry weight plant^{-1}, root dry weight plant^{-1}, shoots dry weight plant^{-1}and total dry weight plant^{-1} of rice	65

LIST OF FIGURES

FIGURE NO.	TITLE	PAGE NO.
1	Global distribution of salt-affected soil (million ha) (Hoang *et al*. 2016)	6
2	Possible responses of plants under salt stress	8
3	Effect of different salt concentrations on the plant height of rice at different days after transplanting	30
4	Effect of different calcium levels on the plant height of rice at different das after transplanting	31
5	Effect of different salt concentrations on the tillers plant-1 of rice at different das after transplanting	34
6	Effect of different calcium levels on the tillers plant-1 of rice at different das after transplanting	35
7	Effect of different salt concentrations on the effective tillers plant^{-1} of rice	37
8	Effect of different calcium levels on the effective tillers plant^{-1} of rice	38
9	Effect of different salt concentrations on the ineffective tillers plant^{-1} of rice	40
10	Effect of different calcium levels on the ineffective tillers plant^{-1} of rice	41
11	Effect of different salt concentrations on the days to flowering of rice	42
12	Effect of different calcium levels on the days to flowering of rice	42
13	Effect of different salt concentrations on the panicle length of rice	43
14	Effect of different calcium levels on the panicle length of rice	44
15	Effect of different salt concentrations on the leaf area of rice	45
16	Effect of different calcium levels on the leaf area of rice	46
17	Effect of different salt concentrations on the relative water content of rice	48
18	Effect of different calcium levels on the relative water content of rice	49
19	Effect of different calcium levels on the leaf membrane stability of rice	50
20	Effect of different calcium levels on the leaf membrane stability of rice	50

LIST OF FIGURES (Cont'd)

21	Effect of different salt concentrations on the SPAD value of rice at different das after transplanting)	52
22	Effect of different calcium levels on the SPAD value of rice at different das after transplanting	53
23	Effect of different salt concentrations on the filled grains panicle^{-1} of rice	55
24	Effect of different calcium levels on the filled grains panicle^{-1} of rice	55
25	Effect of different salt concentrations on the unfilled grains panicle^{-1} of rice	57
26	Effect of different calcium levels on the unfilled grains panicle^{-1} of rice	58
27	Effect of different salt concentrations on the 1000 grain weight of rice	59
28	Effect of different calcium levels on the 1000 grain weight of rice	60
29	Effect of different salt concentrations on the grain yield plant^{-1} of rice	61
30	Effect of different calcium levels on the grain yield plant^{-1} of rice	62

LIST OF APPENDICES

APPENDIX	TITLE	PAGE NO.
I	Experimental location on the map of Agro-ecological Zones of Bangladesh	91
II	Morphological characteristics of the experimental field	92
III	Initial physical and chemical characteristics of the soil	92
IV	Analysis of variance of the data on plant height as influenced by combined effect of salt concentrations and calcium levels of rice	93
V	Analysis of variance of the data on tillers plant-1 as influenced by combined effect of salt concentrations and calcium levels of rice	93
VI	Analysis of variance of the data on SPAD value as influenced by combined effect of salt concentrations and calcium levels of rice	94
VII	Analysis of variance of the data on leaf dry weight plant^{-1}, root dry weight plant^{-1}, shoot dry weight plant^{-1} and total dry weight plant^{-1} as influenced by combined effect of salt concentrations and calcium levels of rice	94
VIII	Analysis of variance of the data on root length, leaf area, leaf membrane stability and relative water content as influenced by combined effect of salt concentrations and calcium levels of rice	94
IX	Analysis of variance of the data on days to flowering, panicles plant^{-1}, effective tillers plant^{-1}, ineffective tillers plant^{-1}and panicle length as influenced by combined effect of salt concentrations and calcium levels of rice	95
X	Analysis of variance of the data on filled grains panicle^{-1}, unfilled grains panicle^{-1}, 1000 grain weight and grain yield plant^{-1} as influenced by combined effect of salt concentrations and calcium levels of rice	95

LIST OF ABBREVIATIONS

%	Percent
@	At the rate of
^{0}C	Degree Celsius
ABA	Abscisic Acid
AEZ	Agro-Ecological Zone
APX	Ascorbate Peroxidase
AsA	Ascorbate
BRRI	Bangladesh Rice Research Institute
Cm	Centimeter
Ca	Calcium
CAT	Catalase
CV%	Percentage of Coefficient of Variance
DAT	Days After Transplanting
DNA	Deoxyribonucleic Acid
DHAR	Dehydroascorbate Reductase
e.g.	As for example
EGTA	Egtazic Acid
et al.	and others
g/gm	Gram
GB	Glycine Betaine
Gly I	Glyoxalase I
Gly II	Glyoxalase II
GPX	Glutathione Peroxidase
GR	Glutathione Reductase
GST	Glutathione S-transferase
GSH	Glutathione
GSSG	Glutathione Disulfide
ha	Hectare
i.e.	that is

	LIST OF ABBREVIATIONS (Cont'd)
kg	Kilogram
kg ha^{-1}	kg per hectare
LSD	Least Significant Difference
L	Liter
M	Meter
MDHAR	Monodehydroascorbate Reductase
MDHA	Monodehydroascorbate
mM	Milli Molar
ml/L	Milliliter per Liter
MG	Methylglyoxal
mg/L	Milligram per Liter
MoP	Muriate of Potash
N	Nitrogen
NR	Nitrate Reductase
nm	Nano Meter
Ng	Nano Gram
P	Phosphorus
pH	Hydrogen ion concentration (Negative Logarithm)
PI	Panicle Initiation
Pn	Net Photosynthetic Rate
Pro	Proline
POX	Peroxidase
PS I/II	Photo System I/II Photochemical
qP	Quenching Randomized Complete
RCBD	Block Design Reactive oxygen
ROS	species
RWC	Relative water content
ppm	Parts Per Million
S	Sulphur
SAU	Sher-e-Bangla Agricultural University

	LIST OF ABBREVIATIONS (Cont'd)
SOD	Superoxide Dismutase
TSP	Triple Super Phosphate
µg/kg	Microgram per kg
Zn	Zinc

Chapter 1

INTRODUCTION

Abiotic stresses are serious threat to agriculture and the natural status of the environment. Among the natural factors, salinity is one of the most harmful abiotic stresses because most of the crops are sensitive to salt stress (Hasanuzzaman *et al*., 2013). Deleterious effects of salinity are attributed to reduced osmotic potential of the growing medium, specific ion toxicity and nutrient deficiency which affect plant growth (Bhattacharjee, 2008).Salinity is a global problem that affects approx. 20% of irrigated land and reduces growth and yield of crops significantly (Qadir *et al*. , 2014). Salinity affected agricultural land is increasing worldwide in a large extent, due to both natural phenomena and agricultural practices such as irrigation systems (Munns and Tester 2008). The crop productivity has been greatly affected by increasing salt concentration in the soil. It has been estimated that salinity affects 7% (approximately) of the world's land area and is responsible for more than 35% decrease in agricultural productivity worldwide (Tanji, 2002). The main threats to plant due to salt stress are two in number: osmotic stress and ionic stress (Flower and Colmer, 2008). Moreover, plant also faces oxidative stress. For general understanding, the response of plants to salt stress can be explained in two main phases: firstly, the shoot ion-independent response occurs within minutes to days, which is thought to be related to Na^+ sensing and signalling (Gilroy *et al*., 2014). Stomatal closure and the inhibition of leaf expansion occurs due to the effect of salinity on water relation (Munns and Termaat, 1986). Secondly, the ion dependent response to salinity which develops over a longer period (days to weeks) and involves the increase of ions in the shoot to toxic concentrations, specially in the older leaves, which causes premature senescence of leaves and resulting in ultimate reduction of yield,even plant death (Munns and Tester, 2008).

Rice (*Oryza sativa* L.) belongs to the grass family (Poaceae) and it is economically and socially dominant over all other crops in Bangladesh. More than three billion people consume rice as staple food accounting for about 50-80% of their daily calorie intake (Khush, 2005). In worldwide, 487.76 milion metric tons of rice was produced during the year of 2018-19(October) (USDA, 2018). USDA estimates Bangladesh to produce around 34.4 million tons of rice in MY 2018-19 (October). Among 114 rice

producers countries of the world Bangladesh ranks fourth (FAO, 2015). The total rice growing area is about 11.38 million hectares, leading to the production of 34.70 million metric tons of rice with an average yield 3.05 t ha^{-1} (DAE, 2015). It is the source of about one third of the total carbohydrate providing considerable amount of recommended Zinc and Niacin. The protein of rice is rich biologically and highly digestive (88%). Being the second most important crop worldwide after wheat it covers almost 90% of area across Asia alone. There are varieties of use of the crop widely ranging from its use as food in cereals, snacks, brewed beverages, flour, rice bran oil (Gopalan *et al*., 2007). One-fifth of the irrigated land around the world is affected by salinity (Koyama *et al*., 2001). Reduction in sodium and chloride uptake into rice while maintaining potassium uptake are characteristics that would aid growth under saline conditions. Salt stress is a major constrain to irrigated rice production in river deltas and former floodplains, particularly in semi-arid and arid climates. Irrigated rice is suitable in controlling and even decreasing soil salinity (Wopereis *et al*., 1998), but rice is highly susceptible to the rhizosphere salinity than other cereals. Vegetative and reproductive stages in rice are highly sensitive.

Response and adaptation of plant to salt stress involve alteration of cellular metabolism and invoking various defense mechanisms (Ghosh *et al*., 2011). Plants can survive under this stressful condition depending on their abilities to perceive the stimulus, generation and transmition of signals, and initiation of various physiological and biochemical changes (Tanou *et al.*, 2009; El-Shabrawi *et al*., 2010). Under stress condition significant increase in reactive oxygen species (ROS), such as singlet oxygen ($^1O_2$), superoxide (O^{2-}), hydrogen peroxide (H_2O_2), and hydroxyl radical (OH^-) occurs (Tanou *et al*., 2009; Pérez-López *et al*., 2010).

It have been found that the basal or foliar application of Ca^{2+} , Mg^{2+}, K^+, proline, glycine-betaine, salicylic acid can mitigate the adverse effects of salinity. As an essential macronutrient, calcium (Ca) plays important roles including stabilizing cell walls and membranes, improving the metabolic processes of other nutrients, regulating enzymatic and hormonal processes, and other essential functions. Calcium also acts as a secondary messenger that mediates many aspects of cell and plant development, as well as the stress-resistance response (White and Broadley, 2003; Jaleel *et al*., 2007a; Jaleel and Azooz, 2009). The Ca^{2+} has an important role in salt stress signaling that controls ion homeostasis pathways (Yokoi *et al*., 2002). Ca^{2+-}

dependent activation of phosphatase results in transcription of the ENA1 gene, which encodes the P-type ATPase (Mendoza *et al*., 1994). The activity of Na^+/H^+ antiporter is affected by the action of a calcium sensor on a protein kinase which is initiated by the components of signal recognition and transduction pathways (Shi *et al*., 2000). In addition, Ca can ameliorate salt-induced Na^+ toxicity by blocking non-selective cation channel (NSCC) which is the major pathway of Na^+ influx in plant (Demidchik and Tester, 2002; Shabala *et al*., 2006). These findings suggest that calcium can mitigate the sodium toxicity of plant. The mechanisms of Ca in regulating the physiological and biochemical processes in response to salinity induced stress in rice plant are still less known. In this context, it is necessary to understand the role of calcium in mediating salinity induced response in rice plant.

Objectives:

- To study the morphophysiology, yield contributing characters and yield of rice plant under salt stress;
- To observe the role of Ca^{2+} in mitigating salinity induced damages in rice plant, and
- To determine the effective dose of Ca^{2+} for mitigating salt stress.

Chapter 2

REVIEW OF LITERATURE

2.1 Rice

Rice (*Oryza sativa* L) is a member of the family Poaceae. The basic chromosome number of rice is n=12. Both diploid and tetraploid species are found. In this respect, *Oryza sativa* L. and *Oryza glaberrima* L both are diploid species (2n= 24) (Brar and Khush, 2003). The Asian cultivated rice (*Oryza sativa* L) is a model crop species as it is the first fully sequenced crop genome. It is considered as a major food crop worldwide. It is consumed as the staple food by more than three billion people accounting for about 50-80% of their daily calorie intake (Khush, 2005). It is a monocarpic crop, therefore blooms once, complete life cycle in a season. Cultivated rice is an annual grass and growing to 0.6- 1.8 m with a round, hollow and joints culms, flat leaves and inflorescence is terminal (Kellogg *et al*., 2013). On the basis of morphology rice cultivars are classified primarily into three types-*indica, japonica*, and *javanica* (Purseglove, 1985). *Indica* rice cultivars are adapted to tropical monsoon climate. *Oryza rufipogon* which was domesticated in China about 8,200-13,500 years ago is the predecessor of both *indica* and *japonica.* (Wei *et al.,* 2012). In Bangladesh almost all the cultivars of rice cultivated belongs to the sub-species *indica* (Alim, 1982). About 76% peoples of Bangladesh live in rural areas and manpower involved in agricultural activities is around 47.5% (Shelley *et al*., 2016). Rice is cultivated all over the country (except southern hilly area) as the tropical climatic condition Bangladesh is suitable for rice cultivation. Rice is grown all year round in Bangladesh with three distinct seasons (Aus, Aman and Boro) and grown in four ecosystems namely irrigated (boro), rainfed (transplanted Aus and Aman), rainfed upland (direct-seeded Aus) and deepwater (broadcast Aman) (Hussain, 2012). The major portion of these three rice growing seasons have covered by modern varieties developed by BRRI (more than 73%) and the rest (6%) hybrid varieties which are marketed by private sectors (Hussain, 2012). From the total global production of rice the people of Bangladesh consumes 6.06% and actual intake is 416 g per capita per day, which makes it the fourth highest rice consuming nation of the world (Hussain, 2012). In the last few decades the rice production increased due to the great effort of rice researchers and farming innovations. In 1970, the rice production was 1.7 t ha^{-1} (FAO, 2014) however; now it is 3.05 t ha^{-1}(DAE, 2015). Despite the success in rice production still now Bangladesh faces many challenges in agricultural sectors, especially in rice production owing to climate change and growing population. At the same time, every year more than 1% cultivable land decrease because of the construction of house, industries, factories, roads and highways (Shelley *et al*., 2016). Moreover, rice is sensitive to abiotic stresses (drought, salinity, high temperature, low temperature etc.) which are occurring frequently in recent due to global climate change (Mohanty, *et al*., 2013). But among all abiotic

stresses salinity is one of the main environmental constraints to production of rice in Bangladesh. Rice (*Oryza sativa* L.) is rated as one of the major food crops in the world, but is also considered extremely salt-sensitive (Maas and Hoffman, 1977). Till now Bangladesh Rice Research Institute (BRRI) and Bangladesh Institute of Nuclear Agriculture (BINA) has developed few salinity tolerant varieties viz. BRRI dhan40, BRRI dhan41, BRRI dhan47, BRRI dhan53, BRRI dhan54, BRRI dhan61, BRRI dhan67, Binadhan-8, Binadhan-10 are salt tolerant (Shelley *et al*., 2016). These few varieties are not sufficient to cope with the adverse environment condition for increasing the rice production to feed the growing population of Bangladesh.

2.2 Abiotic stress

Worldwide agriculture is facing a lot of challenges like producing 70% more food for an additional 9.7 billion people by 2050 while at the same time fighting with poverty and hunger, consuming scarce natural resources more efficiently and adapting to climate change (Wilmoth, 2015). Stress may be defined as an environmental adverse state that can be reduced the plant growth and potential yield of crops. It is usually divided into two groups viz. biotic and abiotic stress (Pandey *et al*., 2017). These two stresses have occurred normally in nature. But abiotic stress causes more damages than biotic stress. (Pande and Arora, 2017). Surveys say that every year about 50% yield loss due to the abiotic stresses such as drought, salinity, high temperature, mineral deficiency and metals toxicity (Haggag *et al*., 2015). However, the productivity of crops is not increasing in parallel with the food demand. The lower productivity in most of the cases is attributed to various abiotic stresses. Lowering crop losses due to various environmental stressors is a major area of concern to cope with the increasing food requirements (Shanker and Venkateswarlu, 2011). Abiotic stresses regulates plant metabolism leading to harmful effects on growth, development and productivity. If the stress becomes very high and/or continues for an extended period it may lead to an intolerable metabolic load on cells, reducing growth, and in severe cases, result in plant death (Hasanuzzaman *et al*., 2012). Harmful chemical entities called reactive oxygen species (ROS), which include hydrogen peroxide (H_2O_2), superoxide radical (O^{2-}), hydroxyl radical (OH^-), etc. are produced in the plant due to abiotic stress (Choudhury *et al*., 2013).

Keunen *et al*. (2013) concluded that plants suffering from abiotic stress are commonly facing an enhanced accumulation of reactive oxygen species (ROS) with damaging as well as signaling effects at organellar and cellular levels. The outcome of an environmental challenge highly depends on the delicate balance between ROS production and scavenging by both metabolic and enzymatic antioxidants. To meet these challenges, genes, transcripts, proteins, and metabolites that control the architecture and/or stress resistance of crop plants in a wide range of environments will need to be identified, in order to facilitate the biotechnological improvement of crop productivity. However, dependent on duration and magnitude of stress plants, try to adjust the environmental

stress through changes in morphological structure, physiological and biochemical activities. Adaptive responses of plants to abiotic stress include closure the stomata which limit the water loss and initiation of a series of physiological processes for maintaining the integrity of photosynthesis, CO_2 fixation apparatus and increase the antioxidant activities in plants (Pandey *et al*., 2017). All biochemical components are present in plants which required for stress tolerance, but the strength of these biochemical components shift species to species depend on the magnitude of stress. This difference of the plant‘s response to stress depends on the signaling insight, transduction and potentiality of defense machinery of plants which respond to these signals (Ben Rejeb *et al*., 2014; Scheres and Van der Putten, 2017). Traditional plant breeding approaches to improve abiotic stress tolerance of crops had limited success due to multigenic nature of stress tolerance.

2.3 Salt stress

Salt stress is considered as one of the most brutal abiotic stress that hamper the normal growth and development of plants (Ahmad *et al.* 2015; Ahanger and Agarwal 2017). Salinity/salt stress refers to the excess amount of salts in the plant, soil and/or water which significantly reduces vigor and yield of the crop. Common salts found in nature are- sodium chloride, sodium sulphate, magnesium sulphate, potassium sulphate etc. (Hasanuzzaman *et al.* 2013). A considerable amount of land around the world is affected by salinity which is increasing day by day. More than 45 million hectares (M ha) of irrigated land which account to 20% of total land have been damaged by salt worldwide and 1.5 M ha are taken out of production each year due to high salinity levels in the soil (Pitman and Läuchli 2002 ; Munns and Tester 2008) .

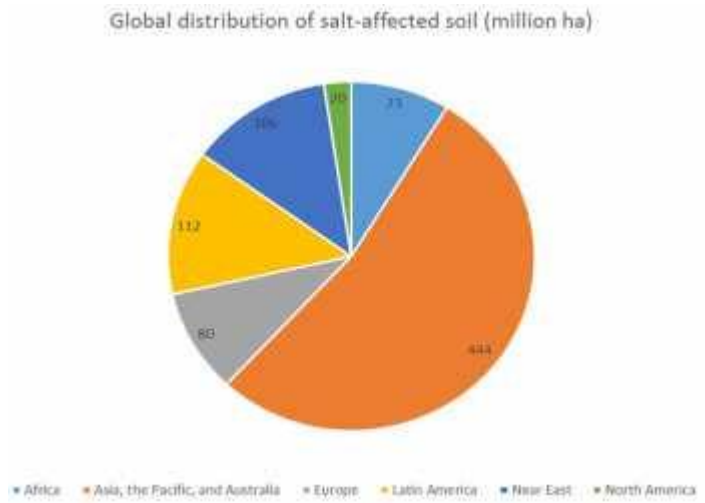


Figure 1: Global distribution of salt-affected soil (million ha) (Hoang *et al*. 2016)

In Bangladesh, around 20% of costal area covers approximately 30% of the net cultivable region (Shelley *et al*., 2016). As in monsoon season the coastal area flooded with saline water and contaminated groundwater which is low in concentration the problem is not severe, but in the dry season, salt concetntration increases because the contaminated groundwater comes out over the soil surface. For this reason land use varies with season to season; thus in winter season coastal area remains fallow. Whether, in wet-season, standing water decrease saline concentration in root zone which allows farmers to cultivate traditional rice varieties. The cropping intensity in saline area of Bangladesh is relatively low, mostly 170% ranging from 62% in Chittagong coastal region to 114% in Patuakhali coastal region (FAO, 2007).

Generally, two main stresses in plants viz. osmotic stress and ionic stress caused by salinity. Osmotic stress occurs when the salt concentration exceeds the tolerance level of plants around rhizosphere in soil, after that salt reaches in older leaves which increase the amount of Na^+ into plants and causes ionic stress, thus interruption the metabolic process and possess cell death (Munns and Tester, 2008; Hasanuzzaman *et al*., 2013a). Moreover, due to salinity many nutrients are unavailable for plant such as nitrogen (N), phosphorous (P), copper (Cu), zinc (Zn) and decrease potassium (K), calcium (Ca), magnesium (Mg) into plants causing significant yield loss (Nahar *et al*., 2016; Shelley *et al*., 2016).

Plants can be categorized into two groups on the basis of their salt tolerance and response to salt stress, such as (a) halophyte, (b) glycophte. Halophytes are those plants that can tolerate high concentration of salinity (400 mM); while glycophytes are tolerant to low concentration of salinity (Hoang *et al*., 2016). In nature, most of the crops are glycophytes and susceptible to salinity, except rye (*Secale cereale*) which is a salt-tolerant cereal crop which can withstand salt stressupto 11 dS m^{-1} electrical conductivity (USDA, 2016). However, rice is a moderate sensitive cereal crop to salinity having threshold electrical conductivity of 3 dS m^{-1} for most cultivated varieties (Mohanty, *et al*., 2013; Hoang *et al*., 2016, USDA, 2016), whereas, normally salinity is denoted in soil when electrical conductivity is above 4 dS m^{-1} (Rengasamy, 2006). For rice, even it was reported that rice yield reduce 10% and 50% at 3.5 dS m^{-1} and 7.2 dS m^{-1} of electrical conductivity respectively (Umali, 1993). Salinity stress tolerance among rice genotypes varies depending on different developmental stages. It has been reported that rice shows salt tolerant during germination, active tillering and toward maturity, but at the time of early vegetative stage and reproductive stage it shows susceptibility (Heenan *et al*., 1988; Zeng *et al*., 2001; Moradi *et al*., 2003; Singh *et al*., 2008).

2.4 Effect of salt stress on plant

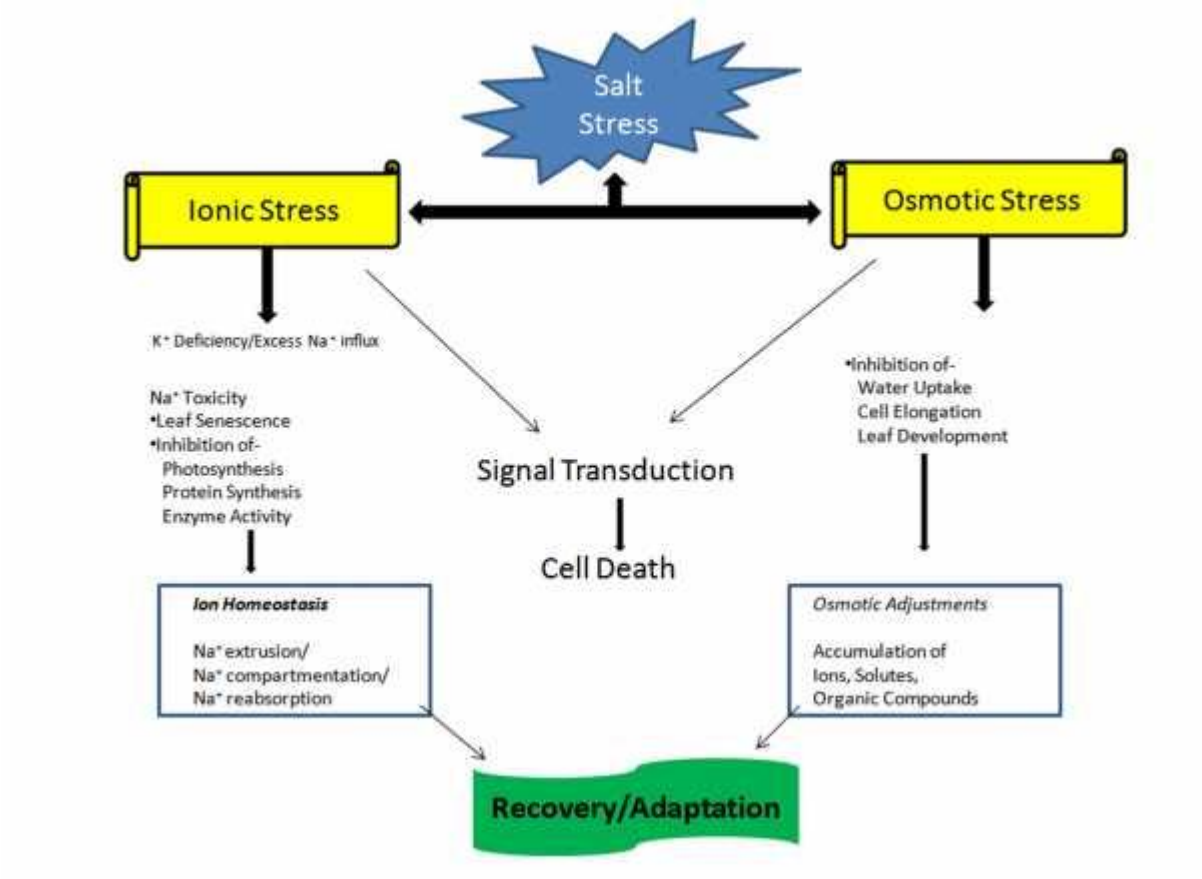


Figure 2: Possible responses of plants under salt stress

2.4.1 Effect on plant water relations

Relative water content (RWC), leaf water potential, stomatal movements, transpiration rate, leaf and canopy temperature are the main characteristics of plant which effect on water relations. Effects of salt on plants water relations have been described in many previous studies. Mainly two components are involved in plant water relations namely water potential and hydraulic conductivity (Negrão *et al*., 2017). Salinity increases osmotic effect on plants due to the reduction of soil water potential surrounding the root zone, which interrupt the ability of water uptake from soil to maintain turgor pressure (Sabir *et al*., 2009). Moreover, under low and moderate salinity stress plants can maintain a potential gradient to uptake water through accumulation of osmolyte solutes. With increased evidence many researchers found that water potential and osmotic potential became more negative with increased salinity, whether, turgor pressure increased (Gulzar *et al*., 2003). In wheat (*Triticum aestivum* L.) plant, Nishida *et al*. (2009) pointed

out that under high salinity concentration leaf water potential and transpiration rate of tomato plant was decreased. Similarly, several authors reported leaf water potential was decreased with the increased of salinity concentration in rice (Siddiqui *et al*., 2014); maize (Prasad *et al*., 2016). Ueda *et al*. (2013) were carried out an experiment with two rice cultivars namely CFX 18 (salt-tolerant) and Juma67 (salt-sensitive). They reported that leaf area, root-shoot fresh and dry weight, leaf water content and leaf water potential significantly decreased in Juma67 than CFX 18 with increased salinity, therefore, CFX 18 considered more salt tolerant. According to Ueda *et al*. (2013) it can be suggested that under higher salinity concentration plants sequester more NaCl in leaf tissue than normally occurs, thus, lower osmotic potentials and reduction in the root hydraulic conductance decreases water flow rate from root to shoot, causing water stress in the leaf tissue

2.4.2 Effect on plant nutrient availability

Salt stress limits the nutrients uptake in plant. It also increases the Na^+ and Cl^- ions in plants; therefore, the concentrations of potassium (K^+), calcium (Ca^{2+}) and nitrate (NO_3^-) are decreased in plants (Tavakkoli *et al*., 2011). Both Na^+ and Cl^- ions inhibit the growth of plants through reduced absorption of other ions and nutrients which are required for plants growth (Ahmed *et al*., 2015). It has also been reported that due to the accumulation of Na^+ and Cl^- in plant tissue potassium (K^+), calcium (Ca^{2+}), magnesium (Mg^{2+}) and manganese (Mn^{2+}) reduced, whereas chloride (Cl^-) ions limits the absorption of nitrate (NO^{3-}), phosphate (PO_4^{3-}) and sulfate (SO_4^{2-}) ions (Termaat and Munns, 1986), which limited the growth of plants. Moreover, Na^+ and Cl^- ions due to salinity stress disturb the specific transport system of these ions into plants (Maathuis, 2006). On the other hand, in a greenhouse experiment Tunçtürk *et al.* (2011) investigated the effect of salinity on micronutrients of twelve canola (*Brassica napus* L.) cultivars and they observed due to 150 Mm NaCl stress iron (Fe), manganese (Mn) and copper (Cu) increased all the parts of plants except some of canola cultivars, whereas zinc (Zn) content in leaves of canola was not significantly affected. Ahmed *et al*. (2013b) reported that accumulation of calcium (Ca), manganese (Mn) and iron (Fe) was increased in wild barley (XZ5) than cultivated barley (CM72) under combined stress of drought and salinity. In contrast, Chakraborty *et al.* (2015) found that under salinity stress (6.76 dS m^{-1}) the accumulation of nitrogen (N), manganese (Mn), iron (Fe) and zinc (Zn) was decreased significantly in susceptible cultivar than tolerant cultivars of *Brassica* spp.

2.4.3 Effect on photosynthetic pigments

Chlorophyll is a major chloroplast component of photosynthesis; therefore, chlorophyll contents have a correlation with photosynthetic rate. Salt stress decreases the chlorophyll contents through accumulation of Na^+ into older leaves. Due to the toxicity of Na^+ into the older leaves start to develop chlorosis and senescence (Yang *et al*., 2011). In some

earlier studies on different plant species showed that salinity stress decreased the chlorophyll contents such as wheat (*Triticum aestivum* L.) (Perveen *et al*., 2010); corn (*Zea mays* L.) (Molazem *et al*., 2010); sunflower (*Heliantus annuus* L.) (Akram and Ashraf, 2011). Ghosh *et al.* (2015) carried out an experiment with mungbean (Vigna radiate L.) crop to investigate effect of salinity stress on plants and they observed that chlorophyll contents of mungbean (*Vigna radiata* L.) leaves decreased with the increased of salinity. Furthermore, the toxicity of Na^+ into oldest leaves impair the biosynthesis of chlorophyll contents or increase the degradation of photosynthetic pigments. Bhusan *et al.* (2016) observed in an experiment that under salinity stress chlorophyll b and total chlorophyll contents of rice cultivars decreased but chlorophyll a was increased significantly. Moreover, experiment on sunflower (*Heliantus annuus* L.) (Akram and Ashraf, 2011) under salinity stress have showed that the important precursors of chlorophyll viz. glutamate and 5-aminolaevulinic acid (ALA) decreased during stress, which denoted that salinity stress affects more significantly on chlorophyll biosynthesis than breakdown. Although, it has been reported that reduction of chlorophyll contents under salinity stress depend on plant species because of chlorophyll contents increase in salt-tolerant cultivars, decrease in salt-sensitive cultivars (Khan *et al*., 2009). For this it has been suggested that increase of chlorophyll contents in salt-tolerant plants are a physiological indicator of salinity stress tolerance in wheat (*Triticum aestivum* L.) (Raza *et al*., 2006); alfalfa (*Medicago sativa*) (Monirifari and Barghi, 2009); rice (*Oryza sativa* L.) (Chunthaburee *et al*., 2016).

2.4.4 Effect on photosynthesis

Many studies have been reported on the reduction of photosynthetic activity under salt stress in a number of plant species (Stepien and Johnson, 2009; Pérez-López *et al*., 2012). Photosynthesis activity impaired in salt stress due to the lower stomatal conductance, hampered in CO_2 fixation process, photochemical capacity inhibited (Ahmed *et al*., 2015). In a previous study Muranaka *et al*. (2002) observed that under 100 mM salt stress the photosynthesis rate was decreased in two wheat (*Triticum aestivum* L.) at two stages. At first photosynthesis was decreased slowly without any visible changes in photochemical and after that in second stage photosynthesis was reduced together with impaired the energy generation efficiency of photo-system two (PSII). However, excessive accumulation of Na^+ under salinity stress in plants affects the electron transport system which reduced photosynthesis activity. Zeid (2009) reported that in maize (*Zea mays* L.) crop salinity stress reduced the photosynthesis activity through impaired the hill reaction. Moreover, at the time of reproductive stage salinity stress reduced the stomatal conductance, net CO_2 assimilations of wheat genotypes leaves (Perveen *et al*., 2010). Furthermore, reduction of photosynthesis in plants due to the salinity stress is related to reduce production of ATP through impair the electron transport system (Curtiss *et al*., 2011). Ionic toxicities of Na^+ and Cl^- reduced the growth and photosynthesis of plants

through impaired the photosynthetic apparatus reported by Tavakkoli *et al.* (2011). With increased evidence it was reported that photosynthesis activity of plant species reduced under salinity stress such as tomato (*Solanum lycopersicum*) (Sholi, 2012); mustard (*Brassica juncea* L.) (Wani *et al*., 2013); cotton (*Gossypium hirsutum* L.) (Zhang *et al*., 2014). Wang *et al.* (2017) observed in their experiment that specific ionic toxicity of Na^+ and Cl^- decreased the photosynthetic rate, CO_2 concentration and also impaired the electron transport system in rice (*Oryza sativa* L.) crop leaves.

2.4.5 Effect on plant yield

Salt stress affects on plant growth at different stages of life cycle. The adverse effect of salinity on rice crop at different stage was observed by Zeng *et al.* (2001) and they were exposed the rice crop to salinity at the time of seeding, first leaf, third leaf, panicle initiation (PI) and booting stages respectively. They point out that salinity stress before PI reduced shoot dry weight and grain yield. Yield of plants are closely associated with grain number and weakly associated with grain size; therefore, Harris *et al.* (2010) carried out a greenhouse experiment with barley (*Hordeum vulgare* L.) and wheat (*Triticum aestivum* L.) to observe the effect of salinity on growth and yield. They observed at high salinity stress grain number reduced at 31%, 22% in barley and wheat crop respectively and also grain size reduced in both crops. Moreover, salinity stress at flowering stage also detrimental as like drought stress. In tomato (*Solanum lycopersicum* L.) plant, salinity reduced the yield due to the tomato plants were subjected to salinity at flowering and fruiting stages (Zhang *et al*., 2017). Plants yield also correlated with spikelet fertility and grain filling. For this, Ahmed *et al*. (2013) observed in a greenhouse experiment on barley (*Hordeum vulgare* L.) cultivars that under single or combined stress of drought and salinity during anthesis period reduced spike length and grain filling per spike in CM72 and XZ16 at 22.6%, 27.7% and 36.8%, 19.9% respectively.

2.4.6 Oxidative damages and ROS production

Reactive oxygen species (ROS) are produced normally in plant for cell metabolism function but when plants expose to environmental stresses lead to excessive reactive oxygen species (ROS) generation which is one of the earliest biochemical responses of eukaryotic cells to biotic and abiotic stresses (Apel and Hirt, 2004). ROS include free radicals ($O^{\bullet 2-}$ and $OH^{\bullet}$) and non-radicals molecules ($^1O_2$ and H_2O_2) (Hasanuzzaman *et al*., 2013b). In plants these ROS generation mainly occur due to the electron leakage from chloroplasts (when CO_2 is limited), mitochondria (overreduction of electron transport chain), peroxisomes (glycolate oxidized to glyoxylic acid during photorespiration), plasma membranes and various metabolic pathway in cellular compartment under stressful conditions (Miller *et al*., 2010; Sharma *et al*., 2012). Salinity played a major role to produce excessive ROS in plants like other abiotic stresses (Nxele *et al*., 2017). These excessive productions of ROS under drought and salinity stress cause oxidative

damage through accelerate the lipid peroxidation, denaturation of protein, mutation of DNA impair cellular homeostasis and the antioxidant activities (Apel and Hirt, 2004; Miller *et al*., 2010), thus inhibit the plants growth and development. However, plants can detoxify this ROS through increasing endogenous enzymatic compounds such as superoxide dismutase (SOD), catalase (CAT), ascorbate peroxidase (APX), monodehydroascobate reductase (MDHAR), dehydroascorbate reductase (DHAR), glutathione reductase (GR), glutathione peroxidase (GPX), glutathione S-transferase (GST) and peroxidase (POX) and as well as non-enzymatic compounds like ascorbate (AsA), glutathione (GSH), carotenoids, α-tocopherols and proline (Hasanuzzamanet *et al*., 2012a). Moreover, under stress condition higher level of methylglyoxal (MG) compound produce in plants which is highly cytotoxic (Hasanuzzaman *et al*., 2014). To detoxify the higher level of MG plant have possessed a defensive mechanism through Gly I and Gly II system; therefore, MG and the glyoxalase are the potential indicators of plants stress tolerance (Nahar *et al*., 2015a).

2.4.7 Antioxidant defense system

Plants have evolved an own antioxidant defensive mechanism in order to decrease the harmful effect of oxidative damage to maintain the metabolic activities under salt stress. Antioxidants are two types in plants defense system enzymatic (SOD, CAT, APX, MDHAR, DHAR, GR, GPX and GST) and non-enzymatic (AsA, GSH, carotenoids, α-tocopherols and proline) (Hasanuzzamanet *et al*., 2012a). These antioxidants play a vital role to detoxify the ROS in specific compartments of cell in a particular condition. In general during stress condition SOD act as a first line defense system which is catalyzed the dismutation of two molecules of superoxide into O_2 and H_2O_2. Further, the H_2O_2 convert into H_2O and MDHA (monodehydroascobate) through APX, where APX used ascorbate as an electron donor (Hasanuzzaman *et al*., 2012a). As like APX, CAT can also convert H_2O_2 into H_2O, but it has lower affinity to H_2O_2 than APX. However, in plant MDHA is instable; therefore a little portion of MDHA restored into AsA by MDHAR and other portion of MDHA are oxidized to DHA (dehydroascorbate) (Lisenbee *et al*., 2005). After that with the help of MDHAR, DHA can be reduced to AsA by GSH (Saruhan *et al*., 2009). At the same time GSSG is oxidized to GSH by GR (Munne´ Bosch *et al*., 2013). Kibria *et al*. (2017) observed in different rice varieties viz. BRRI dhan29, BRRI dhan47, Binadhan-7 and Binadhan-8 in response of salinity stress. In BRRI dhan47, Binadhan-7 and Binadhan-8 activity of CAT and APX increased but decreased in BRRI dhan29, whereas, POX activity decreased in all varieties. This higher level of CAT and APX in BRRI dhan47, Binadhan-7 and Binadhan-8 showed higher level of ROS scavenging ability and protection mechanism under salt stress. Therefore, the most important role of antioxidative system in plant under stress condition is to detoxify the overproduction of ROS to maintain an optimum level for signaling and restore the metabolic activities.

2.4.8 Accumulation of compatible solutes

Under stress condition plants can accumulate different types of organic or inorganic solutes in the cytosol, which are low molecular weight and highly water soluble, even at high concentrations solutes in cytosol are nontoxic for plants (Ahmed *et al*., 2015). These solutes lowered the osmotic potential; thereby maintain cell turgor (Farooq *et al*., 2009). During stress conditions some compatible solutes are played a vital role to osmotic adjustment such as inorganic solute K^+ at the early stage of stress and organic solutes proline (Pro.), glycine betaine (GB) and glucose at the late stress (Nio *et al*., 2011). It has been reported that inorganic solute K^+ decreased the MDA content, improved osmotic potential, water uptake and cell turgor under both drought and salinity stress in wheat (*Triticum aestivum* L.) (Wei *et al*., 2013), rice (*Oryza sativa* L.) (Zain *et al*., 2014) and mustard (*Brassica campestris* L.) (Umar *et al*., 2011). Whereas, among organic solutes proline is a key biochemical solute that accumulate in significant amount when exposed to salinity stress (Munns, 2005). After so many studies it has been anticipated that an increased of proline can protect the plants during abiotic stress through stabilization of subcellular structure to maintain ions homeostasis (Szabados and Savouré, 2010), furthermore it has also suggested to be an important protector (act as osmolyte) against higher accumulation of ROS under stress condition by quenching O_2 and directly scavenging of $OH^{\bullet}$, as a result proline might be able to protect proteins, DNA (Deoxyribonucleic Acid) and membranes from oxidative damages (Smirnoff *et al*., 1989; Matysik *et al*., 2002).

2.4.9 Programmed Cell Death (PCD)

Being sessile, plants are particularly vulnerable to harsh environmental conditions including saline soils and water deficit. To mitigate salinity stress, plants implement a range of strategies. However, if these mechanisms are unable to cope with the increased stress, the plant will implement PCD (Programmed Cell Death) as a last ditch effort to survive (Greenberg, 1996). Programmed cell death is a physiological and genetically controlled process that is evolutionarily conserved across kingdoms and allows multicellular organisms to eliminate excessive or damaged cells which arise during development and in response to abiotic and biotic stress (Fomicheva *et al*., 2012). Alongside cell division and cell migration, PCD enables the organism to strictly control cell numbers and tissue size and to protect itself from unwanted cells that threaten cellular homeostasis (Hegartner, 2000). Liu *et al*. (2007) found that cell death progressed in a well-regulated manner in rice roots during salinity stress. This suggested a possible function of the dead cells in preventing the influx of excess Na^+ ions into the inner parts of roots and into shoots, leading to salt exclusion. It could also be possible that the plant sacrificed cells to prevent uncontrolled death and the release of toxins to protect and keep other cells growing. PCD is also reported to occur in the root tip of rice as a response to salinity stress (Li *et al.* 2007).

2.5 Effect of salt stress on rice plants

Higher level of salinity stress decreased the biomass remarkably in plants. According to Ishak *et al.* (2015) high concentration of salinity (300 mM NaCl) reduced the shoot dry weight of wild-type Nippobare upto 57.14%.

In a greenhouse experiment Hussain *et al*. (2016) used eleven diverse rice genotypes to investigate the effect of salinity stress. For this they exposed the rice plant under 200 mM (NaCl) concentration salinity stress at reproductive stage and observed that the reduction of plant height is higher in BRRI dhan56 (7.67%), followed by BRRI dhan40 (7.18%), BRRI dhan41 (6.52%), BRRI dhan53 (5.55%), IR29 (5.06%), Nona Bokra (4.17%), FL-478 (3.99%) and Binadhan-8 (1.50%) with little reduction in Binadhan-7 (0.91%), whereas Binadhan-10 was not affected.

Mostofa *et al*. (2015) conducted an experiment with rice crop (BR11) at different concentration of salinity (150 mM and 250 mM). They exposed fourteen-day-old rice seedlings in salinity stress and observed relative water content (RWC) decreased at 15-25% and total chlorophyll content decreased at 17-38%.

Kazemi and Eskandari (2011) stated that the three rice cultivars (Anbar, LD and Hamar) were significantly ($P \leq 0.05$) affected by salt stress, where germination, plumule and radicle length and weight were decreased with increasing in salt concentration. The extent of these reductions was related with the variations in rice cultivar under different salt stress condition. By increasing NaCl concentration, seed germination delayed and decreased in all cultivars. Regarding the relationship between speed of germination and seed vigor, salt stress decreased seed vigor of rice cultivars LD a superior cultivar under all salt stress which can be suggested for cultivation under salinity condition.

Rahman *et al*. (2016a) carried out an experiment with rice cultivar BRRI dhan47 to investigate the response rice seedlings under salinity stress. They exposed the thirteen-day-old rice seedling to 200 mM NaCl salinity stress. They evaluated that due to salinity stress fresh weight, dry weight, relative water content, chlorophyll a, chlorophyll b and carotenoid decreased at 31.23%, 21.74%, 19.24%, 33%, 38.36% and 37.5% respectively compared with control plant. They also found salinity stress decrease the uptake of minerals in rice plants such as Ca (7.32% in root and 29.26% in shoot), Mg (48% in root and 13.63% in shoot), Mn (27.78% in root and 37% in shoot) and Zn (31.15% in root and 31.58% in shoot).

Anbumalarmathi and Preeti (2013) reported that the response of eight *indica* rice varieties against six salinity levels (0, 4, 8, 12, 16 and 20 dS m^{-1}) was studied at germination and early seedling growth stage. Germination was completely arrested in six varieties at 20 dS m^{-1} salt concentration. Rice varieties ADT43, IR50, and MDU5 showed greater salt toleran BRRI dhan47 and Binadhan-10 were treated with five concentrations of NaCl,

viz., 0, 4, 8, 12, and 16 dSm^{-1}. Result indicated that plant height, number of effective tiller $hill^{-1}$, number of in effective tiller $hill^{-1}$, number of field grain $panicle^{-1}$, number of unfilled grain $panicle^{-1}$, panicle length and grain yield $hill^{-1}$ were influenced at different levels of salinity. The number of effective tiller $hill^{-1}$, panicle length, number of filled grain $panicle^{-1}$ and grain yield $hill^{-1}$ were significantly decreased with the increased levels of salinity. It was found that the K content in shoot was decreased with the increased levels of salinity. The highest K content (1.77 %) in shoot was found in Binadhan-10 at 0 dSm^{-1}. The highest Na content (1.69 %) in shoot was found in BRRI dhan47 at 12 dSm^{-1}. Between these two varieties Binadhan-10 showed better performance at salinity stress up to a certain level except plant height (Sultana *et al.*, 2014).

To investigate the effect of salinity on growth as well as physiological and biochemical characteristics Kibria *et al.* (2017) conducted an experiment with four rice varieties including one salt-sensitive variety BRRI dhan28 and three salt-tolerant varieties viz. BRRI dhan47, Binadhan-8 and Binadhan-10 and comprised four different salt concentration (0, 20, 40 and 60 mM NaCl). They exposed all rice varieties to salinity stress at five weeks after transplanting and observed that chlorophyll content of BRRI dhan28, BRRI dhan47, Binadhan-8 reduced by 38%, 32% and 42% at 60mM NaCl respectively compared with control and among all varieties Binadhan-10 showed the higher K^+/Na^+ ratio compared than other varieties.

Hakim *et al*. (2014) conducted a glasshouse experiment to observe the effect of salinity on growth and yield of rice. They comprised four levels of treatment viz. 0, 4, 8, 12 dS m^{-1}) and they treated rice genotypes with four levels of salinity at reproductive stage and observed the yield reduction varied between 34-96% with the salinity concentration increased. they found that BRRI dhan29 did not survive at 8 dS m^{-1} salt concentration.

Hussain *et al*. (2016) reported that higher level of salinity (200 mM) decreased the yield of rice varieties. In their study they tasted 11 rice varieties yield under salinity stress and observed reduction level is higher in IR29 (49.64%).

In order to investigate the effect of salinity Nounjan *et al*. (2012) performed an experiment with one rice cultivar namely KDML 105 which is salt- sensitive. For this they used 100 mM NaCl as salinity treatment and thirty three-day-old rice seedlings exposed to stress. After completing the treatment period they observed that proline content, SOD, POX, APX and CAT activities increased in rice plants.

To observe the biochemical responses of rice seedlings under salinity stress Mostofa *et al.* (2015) performed a study. For this treatments were comprised at 150 mM and 250 mM salt concentrations (NaCl). Twelve-day-old seedlings exposed to salt stress for 72 hours. They observed due to salt stress the MDA, H_2O_2, Pro and LOX activity increased. Moreover, salt stress stimulated the activities of SOD, MDHAR, DHAR, GR, GPX, APX

and GSH (Glutathione) but decreased the AsA, AsA/DHA ratio and GSH/GSSG (Glutathione Disulfide) ratio.

Dry weight of root, shoot and yield significantly decreased with the increase of salinity levels, while MR232 and MR211 were less affected. Na^+ ions accumulations increased in the root and shoot with the increase of salinity, while the lowest accumulation was in MR211. Na^+/ K^+ ratio sharply increased in the root with increasing the salinity. Whereas, Ca^{++}/Na^+ and Mg^{++}/Ca^{++} ratio showed decreasing trend with increasing salinity level. The maximum amount of nitrogen and phosphorous accumulation was observed in the shoot of MR211, while Na^+ in BRRI dhan29, K+ in Pokkali. The highest accumulation of Na^+ and K^+ observed in the root of MR219. The maximum Ca^{++} and Mg^+ were found in MR33 and MR211, respectively. Considering all, genotypes MR211 andMR232 were found to be relatively tolerant to salt than the other genotypes (Hakim *et al.,* 2014).

Rahman *et al*. (2016b) conducted an experiment with a rice cultivar namely BRRI dhan47 under salinity stress which is salt-tolerant. They grown the rice seedlings with commercial hydroponics nutrient solution (Hyponex, Japan) in a growth chamber and renewed the nutrient solution after three days. After twelve days they exposed the rice seedlings to 150 mM NaCl stress and then after three and six days later of treatment they assayed the antioxidant contents and observed enzymatic activities of SOD, MDHAR, DHAR, GR, GPX and Gly II (Glyoxalase II) increased in both three and six days, whereas, APX and Gly I (Glyoxalase I) activities increased only in six days treatment. Also Pro. content overtly increased under salinity stress in rice seedlings. Moreover, non-enzymatic antioxidants the AsA and the AsA/DHA ratio decreased but DHA content increased, whereas GSH and GSSG increased and GSH/GSSG decreased in rice seedlings.

2.6 Calcium

Calcium plays an important role in plant growth and development. It is implicated in the movement of cellular organelles such as the spindle apparatus and secretory vesicles, and may play a key role in integrating plant cell metabolism (Jaleel *et al*., 2007a). Calcium is required by the cells of fibrous tissue to bind the polysaccharides that form the middle lamella in the cell plates that arise between daughter cells. The membrane needs adequate levels of calcium to function normally. Most of the interest in calcium in plants has centered on its role in the cytoplasm in controlling developmental process. Free calcium in the apoplast may also influence plant growth (Lawlor, 2002; Jaleel *et al*., 2007b).

Calcium is involved in a wide array of cellular processes, such as cell wall stabilisation, cell extension, secretory processes, maintenance of cell membrane integrity and control

of membranes' permeability and selectivity, cation-anion balance and osmoregulation. Calcium nutrition plays an important role in the maintenance of a high growth rate under saline conditions. (Marschner, 1995). According to Husain *et al.* (2004), the major role of Calcium in increasing the salt tolerance of plants is related to its inhibitory effect on the xylem loading of Na and thus decreases the Na concentration in shoot.

Elevated levels of external Ca^{2+} can increase both growth and Na^{+} exclusion of plant roots exposed to NaCl stress. In addition, roots supplied with elevated levels of external Ca^{2+} (5-10 mM) are often to maintain their K^{+}-concentration; whereas, roots supplied with low Ca^{2+} (0.2-0.5 mM) frequently cannot (Lauchli, 1990). Thus adequate Ca^{2+} is required in the external medium to maintain the selectivity and integrity of cell membrane (Aslam *et al*., 2000). Supplemental Ca^{2+} may also have effects on intracellular membranes of root cells exposed to NaCl stress and may decrease NaCl induced vacuolar alkalization in root tissues by a Ca^{2+} effect on Na^{+} efflux at the plasma membrane (Martinez & Lauchli, 1993). Besides, supplemental Ca^{2+} is shown to prevent NaCl induced breakdown of the pH tonoplast in Mungbean roots exposed to 100 mM NaCl (Colmer *et al*., 1994).

Non-selective cation channels (NSCC) are the major pathway of Na^{+} influx in plant and Ca can ameliorate salt-induced Na^{+} toxicity by blocking those (Shabala *et al*., 2006). Additionally, supplemental Ca inhibits K^{+} efflux resulted from Na^{+} induced plasma membrane depolarization (Shabala and Pottosin, 2014). Moreover, several studies have also revealed that exogenous application of Ca in plant growth medium helps to develop abiotic-stress tolerance by maintaining ion homeostasis (Wu and Wang, 2012), enhancing the antioxidant defense system and other physiological and biochemical attributes (Srivastava *et al*., 2014; Ahmad *et al*., 2015).

Chaum *et al.* (2012) reported that Calcium (Ca) is a signaling molecule that plays an active role in regulating various mechanisms involved in recognition and response to abiotic stresses in plants. However, not much has been done to evaluate its role in regulating physiological and biochemical process in response to salt-induced stress. Two rice genotypes viz. Pokkali, salt tolerant and IR29 salt susceptible, grown on liquid Murashige and Skoog medium (MS) supplied by 1.98 mM $CaCl_2$ (control) were compared to 2 (3.96 mM), 4 (7.92 mM) and 8 (15.84 mM) folds exogenous $CaCl_2$ pretreatment subsequently exposed to 200 mM NaCl salt stress. Thus, the present investigation evaluated the potential of exogenous calcium chloride ($CaCl_2$) supply in improving the growth performance and photosynthetic ability in salt stressed rice. In IR29 salt susceptible rice, leaf area of salt-stressed seedling was significantly recovered

by exogenous application of 7.92 mM $CaCl_2$, which was greater by 1.38-folds over that in 1.98 mM $CaCl_2$ application. Exogenous $CaCl_2$ (7.92 mM) enhanced proline accumulation in both Pokkali (3.26 μmol g^{-1}) and IR29 (4.37 μmol g^{-1}) genotypes, and reduced relative electrolyte leakage thereby indicating its positive role in membrane stability. Treatment of 7.92 mM $CaCl_2$ significantly enhanced the photosynthetic abilities, including maximum quantum yield of PSII (Photo System II)(Fv/Fm), photon yield of PSII photochemical quenching (qP) and net photosynthetic rate (Pn), in two genotypes of salt-stressed rice seedlings, especially in salt susceptible IR29 genotypes. The study concludes that an exogenous application of 7.92mM $CaCl_2$ significantly enhanced the photosynthetic abilities and overall growth performances in the photoautotrophic growth of salt-stressed rice seedlings. Exogenous calcium in the culture media may absorb by root tissues, transfer to whole plant and function as salt defense mechanisms including calcium signaling in the abscisic acid (ABA) regulation system and calcium sensing in stomatal closure when plant subjected to salt stress.

Soualem *et al*. (2014) studied the effect of calcium sulfate ($CaSO_4$) supply under salt stress was studied in two populations of *Atriplex halimus* from two locations (coastal western Algeria (Oran) and continental semi-arid zone contrasted for salinity gradients. The plants were grown in pots and subjected to salt stress (0, 300 or 500 mM NaCl) with a supply of (5 or 10mM) of $CaSO_4$. Growth, mineral, proline and soluble sugars contents were measured. The results showed a reduction in growth with increasing NaCl concentration. The impact of salinity was more pronounced on the inland population than the coastal one. The leaves Na^+ content increased with increasing salt stress and led to reduced plant growth. In response to the intensity of salt stress and $CaSO_4$ supply, plants accumulated more soluble sugars, proline and K^+. This accumulation was more pronounced at high concentrations of NaCl and $CaSO_4$ in both populations. Our results emphasized that supply of $CaSO_4$ reduced the inhibitory effects of NaCl.

The protective effect of calcium against salt stress in plants, by counteracting some of the deleterious effects of sodium, is well documented (Gul and Khan, 2008). Externally supplied Ca^{2+} reduces the toxic effects of NaCl, presumably by facilitating a high K^+/Na^+ selectivity (Abdel Latef, 2011).

Sodium reduces binding of Ca^{2+} to the plasma membrane, inhibits influx while increasing efflux of Ca^{2+}, and depletes the internal stores of Ca^{2+} from endomembranes. Ameliorative effects of supplemental Ca^{2+} on salt stress are exerted through preventing Na^{+}-related changes in the cell Ca^{2+} homeostasis. (Rengel, 1992).

Jafari (2009) studied the interactive effects of salinity, calcium and potassium on physio-morphological traits of sorghum (*Sorghum biclolor* L.) in a greenhouse experiment. Treatments included 4 levels of NaCl (0, 80, 160, and 240 mM NaCl), 2 levels of $CaCl_2$ (0 and 20 mM), and 2 levels of KCl (0 and 20 mM). Number of leaves was significantly affected by NaCl, while elevated calcium promoted total number of leaves, particularly at high levels of NaCl. The interaction effect of Ca^{2+} and K^{+} improve leaf generation only at 160 mM NaCl. Salt stress, increased mortality of leaves exposed to salinity. Application of Ca^{2+} and K^{+} didn't make up the adverse effects of salinity, in comparison to control.

Tzortzakis (2010) reported that, Salinity either of soil or of irrigation water causes disturbance in plant growth and nutrient balance and reduces crop yields. The effects of NaCl salinity and/or calcium or potassium level on the plant growth and severity of gray mold (*Botrytis cinerea* [De Bary] Whetzel) were investigated in endive (*Cichorium endivia* L., cv. Green Curled) grown with the nutrient film technique under greenhouse conditions during early spring. Plants were supplied with nutrient solutions containing 40 mmol L^{-1} of sodium chloride (NaCl) and/or 10 mmol L^{-1} potassium sulphate (K_2SO_4). Additionally, plants treated with foliar spray of 15 mmol L^{-1} calcium nitrate [$(CaNO_3)_2$] or distilled water. Salinity or K and Ca enrichment mainly affected the upper part of endive plants and reduced leaf area. However, when salinity combined with either K or Ca enrichment, the negative impact of salinity on plant growth was reversed. Salinized and/or K and Ca enriched, plants did not differ in plant biomass, leaf/root ratio, leaf fresh weight, leaf number, and root length. Salinity did not have any impacts on photosynthetic rate, stomatal conductance, and intercellular CO_2 concentration. Indeed, photosynthetic rate and stomatal conductance increased with Ca foliar application and decreased with K while the opposite effects were observed for the intercellular CO_2 concentration. Total nutrient uptake was reduced 2-fold in salt-treated plants compared to controls. No symptoms of tip-burn or blackheart were recorded throughout the experimental study. Endive grown in the nutrient film technique had tolerance to NaCl salinity, and this method could be used to exploit saline water in soilless culture. These findings also suggest that a proper management of the salt concentration of the nutrient solution plus external elemental enrichment may provide an efficient tool to improve the quality of leafy vegetables with little effect on yield.

A green house experiment was conducted by Mohamed and Basalah (2015) to assess the morphological measurements and photosynthetic pigments of Cowpea (*Vigna unguiculata* L.) at 50 days after sowing. Results proved that shoot and root length, leaves number, leaf area and fresh and dry weights of shoot and root were significantly

decreased with increasing salt concentrations, except for the low concentration 20 mM NaCl where it non significantly increased compared with the control plants. Addition of calcium chloride led to a significant increase in all growth characters compared to salt stressed plants. Highly significant increase in photosynthetic pigments contents in cowpea plants were obtained at 20 mM NaCl and then it significantly decreased in the higher concentrations of salt. When the salt stressed plants treated with calcium chloride, photosynthetic pigments contents were increased significantly.

Calcium supplementation in the salt-stressed rice seedlings improved ion homeostasis by inhibition of Na^+ influx and K^+ leakage. Exogenous Ca also improved ROS and MG detoxification by improving the antioxidant defense and glyoxalase systems, respectively. On the other hand, applying EGTA (egtazic acid) along with salt and Ca again negatively affected the seedlings as EGTA negated Ca activity. It confirms that, the positive responses in salt-stressed rice seedlings to exogenous Ca were for Ca mediated improvement of ion homeostasis, antioxidant defense and glyoxalase system. (Rahman *et al.* 2016a).

Khan (2013) reported that salinity reduced the growth of wheat plants. When K and N were applied as foliar spray on the wheat plant, it reduced the effect of salinity and increased the plant growth such as plant height, leaf number plant fresh and dry weight and physiological attributes such as chlorophyll content of wheat plants. Similarly, grains yield is also decreased by salinity but foliar application of K and N mitigated the salinity effect on grains yield.

Parvin *et al*. (2015), exogenous application of Ca^{2+} significantly mitigates the adverse effects of salinity on plant biomass production or morphology, physiology and fruit production. The plant height, leaf number/plant, branch number/plant, dry weight of shoot/plant, leaf chlorophyll content, fruit weight/plant were increased with the application of calcium in saline condition compared to without calcium. The fruit weight of tomato is gradually decreased with the increased levels of salinity. In case of treatment combinations, the reduction rate of fruit weight of tomato was decreased with increased levels of Ca^{2+} in response to different saline conditions and the lowest fruit weight was recorded at the highest salinity level (8 dS m^{-1}) along with without Ca^{2+}.

The effect of potassium (K) and calcium (Ca) on growth and antioxidant defence system of salt-stressed Indian mustard plants was studied by Yousuf *et al.* (2015). The response of the plants was studied ten days after treatment. Salt stress inhibited growth parameters including biomass, chlorophyll content, protein content and NR (Nitrate Reductase) activity. Membrane damage was induced by the salt treatment with a concurrent increase in antioxidant defense system and proline content. Individual application of K and Ca mitigated the negative influence of the stress with the maximum alleviating potential exhibited by the combined application of these nutrients.

Manivannan *et al.* (2007) worked on the ameliorating effect of calcium chloride on sodium chloride-stressed plants of *Vigna radiata* L. Wilczek. Plants were treated with solutions of 100 mM NaCl, 100 mM NaCl with 5 mM $CaCl_2$, or 5 mM $CaCl_2$. Groundwater was used for irrigation as the control. Plants were harvested randomly 30 and 50 days after sowing. NaCl and $CaCl_2$-stressed plants showed reduced growth as indicated by decreased root length, stem length, total leaf area and dry weight. Proline and glycinebetaine content and the activity of the antioxidant enzymes superoxide dismutase, ascorbate peroxidase and catalase were increased under treatment with NaCl alone and $CaCl_2$ alone. When $CaCl_2$ was combined with NaCl, $CaCl_2$ altered the overall plant metabolism to ameliorate the deleterious effects of NaCl stress and increased the vegetative growth of the plants.

Anbu and Sivasankaramoorthy (2014) worked on a pot culture was carried out with *Oryza sativa* L. vari-Co-39, to investigate the effects of supplementary calcium chloride on plants grown at NaCl (50mM) concentration. Treatments were: (1) Control: nutrient solution alone (C); (2) nutrient solution plus 50mM sodium chloride (NaCl); (3) nutrient solution plus 10mM calcium chloride ($CaCl_2$); (4) nutrient solution plus 15mM calcium chloride ($CaCl_2$); (5) nutrient solution and 50 mM NaCl plus supplementary 10 mM $CaCl_2$ (NaCl + $CaCl_2$); and (6) 50 mM NaCl plus additional mixture of 15 mM $CaCl_2$ in nutrient solution (NaCl + $CaCl_2$). The plants grown under salt stress produced low dry weight and relative water content than those grown in standard nutrient solution and in $CaCl_2$ alone. Supplemental calcium chloride added to nutrient solution containing salt significantly improved growth and relative water content. Membrane permeability increased with high NaCl application and these increases in root membrane permeability were decreased with supplementary Ca. The concentration of chloride (Cl) increases highly for all treatments. Sodium (Na) concentration in plant tissues increased in both shoots and roots at high NaCl treatment. Application of supplementary Ca lowered Na concentration. Concentrations of Ca. K and N were at deficient ranges in the plants grown at high NaCl levels and these deficiencies were corrected by supplementary Ca. The ameliorating effect of Ca on growth and physiological variables could reduce the negative effect of salinity of *Oryza sativa* L. plants.

Chapter 3

MATERIALS AND METHODS

This chapter presents a brief description about experimental period, site description, climatic condition, crop or planting materials, treatments, experimental design and layout, crop growing procedure, fertilizer application, uprooting of seedlings, intercultural operations, data collection and statistical analysis.

3.1 Experimental location

To study the morpho-physiological and yield responses this experiment was conducted at Experimental shed of the Department of Agricultural Botany, Sher-e-Bangla Agricultural University, Dhaka during the period from December-April, 2017-18. The location of the experimental site has been shown in Appendix I.

3.2 Soil

The soil of the experimental area belonged to the Modhupur tract (AEZ No. 28). It was a medium high land with non-calcarious dark grey soil. The pH value of the soil was 5.6. The soil was clay loam in texture with common fine medium distinct dark yellowish brown mottles. The collected soil was pulverized and inert materials, visible insect pest and propagules were removed. The soil was dried in the sun, crushed carefully and thoroughly mixed. The physical and chemical properties of the experimental soil have been shown in Appendix II.

3.3 Climate

The experimental area was under the subtropical climate and was characterized by high temperature, high humidity and heavy precipitation with occasional gusty winds during the period from April to September, but scanty rainfall associated with moderately low temperature prevailed during the period from October to March.

3.4 Description of the rice variety

BRRI dhan67 is a salt tolerant variety of rice which was, used as the test crop in this experiment. This variety was released in 2014 by Bangladesh Rice Research Institute,

Gazipur. It can tolerate salinity 12-14 dS/m (upto 3 weeks) during seedlings stage and 8 dS/m during maturity stage. Grain is medium fine. Its plant height is 100 cm and yield is 3.8-7.4 t ha^{-1}. Life cycle of this variety ranges from 140 to 150 days.

3.5 Layout of the experiment

The experiment was set in Completely Randomized Design (CRD) having two factors with 3 replications. The treatment combination of the experiment was assigned at random into 12 pots of each at 3 replications.

3.6 Treatments

The experiment consisted of two factors:

Factor A: Different doses of sodium (Na^+) with irrigation water (4):	Factor B: Different doses of Ca^{2+} as $Ca(OH)_2$ with irrigation water (3):
S_0 = 0 dS m^{-1} S_1 = 4 dS m^{-1} S_2 = 6 dS m^{-1} S_3 = 8 dS m^{-1}	Ca_0 = No Calcium is applied with irrigation water. Ca_1=80 ppm Calcium applied with irrigation water. Ca_2 =160 ppm Calcium applied with irrigation water.

Total 12 treatment combinations were as follows:

S_0Ca_0 = without Salt + without Calcium
S_0Ca_1 = without Salt + 80 ppm Calcium
S_0Ca_2 = without Salt + 160 ppm Calcium
S_1Ca_0 = 4 dS m^{-1} Salt + without Calcium
S_1Ca_1 = 4 dS m^{-1} Salt + 80 ppm Calcium
S_1Ca_2 = 4 dS m^{-1} Salt + 160 ppm Calcium

S_2Ca_0 = 6 dS m^{-1} Salt + without Calcium

S_2Ca_1 = 6 dS m^{-1} Salt + 80 ppm Calcium

S_2Ca_2 = 6 dS m^{-1} Salt + 160 ppm Calcium

S_3Ca_0 = 8 dS m^{-1} Salt + without Calcium

S_3Ca_1 = 8 dS m^{-1} Salt + 80 ppm Calcium

S_3Ca_2 = 8 dS m^{-1} Salt + 160 ppm Calcium

Salinity was maintained throughout the life cycle by checking soil electrical conductivity on a daily basis. Calcium was applied 2 times, after seedling establishment and before anthesis.

Conduction of the experiment

3.7 Seed collection

Seeds of BRRI dhan67 were collected from Bangladesh Rice Research Institute, Joydebpur, Gazipur.

3.8 Pot Preparation

The collected soil was sun dried, crushed sand sieved. The soil and fertilizers were mixed well before placing the soils in the pots. Each pot was filled up with 12 kg soil. Pots were placed at the net house of Sher-e-Bangla Agricultural University. The pots were pre-labeled for each variety and treatment. Finally, water was added to bring soil water level to field capacity.

3.9 Fertilizer Application

The nitrogenous, phosphatic, potassic and sulphur fertilizer were applied in the experimental pots @ 250 kg ha^{-1}, 110 kg ha^{-1}, 140 kg ha^{-1}, 50 kg ha^{-1} in the form of urea, triple super phosphate, muriate of potash and gypsum, respectively. One-third of urea and the whole amount of other fertilizers were incorporated with soil at final pot preparation before sowing. Rest of the nitrogen were applied in two equal splits one at 30 days after transplanting (DAT) and second at 45 days after transplanting (DAT).

3.10 Sowing of seeds in seedbed

Previously collected seeds were soaked for 48 hours and then washed thoroughly in fresh water and incubated for sprouting, the sprouted seeds sown in the wet seedbed.

3.11 Uprooting and transplanting of seedlings

Two Seedling of fourty five days old was uprooted carefully from the seedbed and transplanted in the respective pots at the rate of two seedling pot^{-1} on January 15, 2018.

3.12 Intercultural operations

3.12.1 Weeding and irrigation

Sometimes there were some small aquatic weeds observed in pots that were uprooted by hand pulling. About 3-4 cm depth of water was maintained in the pot until the crop attained maturity.

3.12.2 Plant protection measures

Before heading green leafhopper infestations were observed in the crop and they were successfully controlled by applying Durshban two times on 55 DAT and 62 DAT at 20ml/10L of water. Rice stem borer also attacked and it was controlled by the application of Furadan 5G at 2.5 g/pot.

3.13 Harvesting

The crop was harvested at maturity on 22th April 2018. The harvested crop of each individual pot was bundled separately. Grain, straw and root yields were recorded as g $plant^{-1}$.

3.14 Collection of data

Data collections were done on the following parameters-

- Plant height at 30 days interval up to harvest
- Number of tillers $plant^{-1}$ at 30 days interval up to harvest
- Number of effective tillers $plant^{-1}$

- Number of ineffective tillers plant^{-1}
- Days to flowering
- Number of panicle plant^{-1}
- Panicle Length (cm)
- Chlorophyll content (SPAD Value)
- Relative water content (RWC)
- Leaf Membrane Stability Index
- Leaf Area (cm^2)
- Number of filled grains panicle^{-1}
- Number of unfilled grains panicle^{-1}
- 1000-grain weight (g)
- Grain yield g plant^{-1}
- Leaf dry weight (g)
- Root dry weight (g)
- Stem dry weight (g)

Plant height (cm)

Plant height (cm) was measured from the root base to the tip of the longest leaf at the time of 30, 60, 90 DAT and at harvest.

Number of tillers plant^{-1}

Total tiller number was taken from 30 DAT at 30 days interval up to 90 DAT.

Chlorophyll content (SPAD)

Three leaves were randomly selected from each pot. The top and bottom of each leaves were measured with atLEAF as atLEAF value. Then it was averaged and total chlorophyll content was measured by the conversion of atLEAF value into SPAD units.

Relative water content (RWC)

Three leaves were randomly selected from each pot and cut with scissors. Relative water content (RWC) was measured according to Barrs and Weatherley (1962). Leaf laminas were weighed (fresh weight, FW) and then immediately floated on distilled

water in a petridish for 4 h in the dark. Turgid weights (TW) were obtained after drying excess surface water with paper towels. Dry weights (DW) were measured after drying at 80∘C for 48 h. Then calculation was done using the following formula:

RWC (%) = $(FW - DW / TW - DW) \times 100$.

Leaf membrane stability index

Leaf trips (0.2 g) of uniform size were taken in two sets of test tubes containing 10 ml of distilled water. Test tubes in one set were kept at 40 ^{o}C in a water bath for 30 minutes and electrical conductivity of the water containing the samples was measured (C_1). Test tubes in the second set were incubated at 100 ^{o}C in the boiling water bath for 15 minutes and electrical conductivity (C_2) was measured. MSI was calculated by following formula:

MSI = $[1-(C_1/C_2)] \times 100$

Leaf Area plant^{-1} (cm^2)

Leaf area was measured at heading stage and was expressed in cm^2. Leaf area meter was used in this case.

Number of effective tillers and panicles plant^{-1}

Number of Effective tillers and panicles plant^{-1} were counted at maturity.

Panicle length

Panicle length was recorded from the basal nodes of the rachis to apex of each panicle.

Number of grain panicle^{-1}, filled grains panicle^{-1}, and unfilled grains panicle^{-1}.

In case of more than 5 effective tillers plant^{-1}, average number of grains panicle^{-1} was calculated by counting the number of filled grains and unfilled grains of 5 panicles plant^{-1} which was selected randomly. In case of less than 5 effective tiller plant^{-1}, average number of filled grain was calculated by counting the number of filled grains and unfilled grains of all the panicles plant^{-1}.

Thousand grain weight (g)

Thousand grains weight plant^{-1} was calculated by weighing 100 grains of each treatment and then multiplied by 10.

Grain yield plant^{-1} (g)

The grain yield of the plant which had effective tiller was recorded by weighing after proper drying the grain.

Leaf, Stem and root dry weight (g)

After harvesting, all the leaves, Stems and roots of plant were collected and sliced into very thin pieces and were put into envelop and placed in oven maintaining at 70 ^{o}C for 72 hours. The samples were then transferred into desiccators and allowed to cool down at room temperature. The final weight of the sample was taken in gram.

3.15 Statistical analysis

All the data collected on different parameters were statistically analyzed following the analysis of variance (ANOVA) technique using MSTAT-C computer package program and the mean differences were adjudged by least significant difference (LSD) test at 5% level of significance (Gomez and Gomez, 1984).

Chapter 4

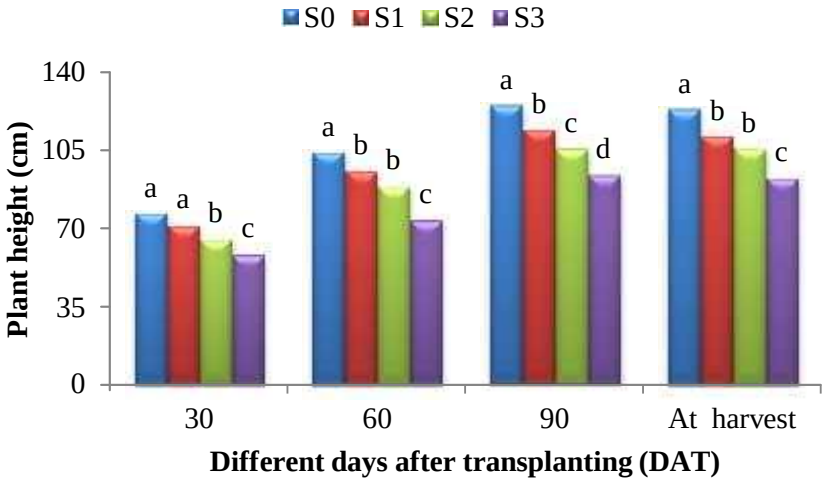
Results And Discussion

An experiment was conducted during the Boro season of (December-june) 2017-2018 to evaluate the response of BRRI dhan67 to calcium supplementation at different salinity levels. It was conducted in the net house of the Department of Agricultural Botany and in the Laboratory of Agricultural Botany of Sher-e-Bangla Agricultural University (SAU), Dhaka. The different morphological, physiological and yield parameters were studied including plant height, number of tillers plant^{-1}, number of effective tillers plant^{-1}, number of ineffective tillers plant^{-1}, chlorophyll content (SPAD Value), relative water content (RWC), leaf Membrane Stability Index, leaf Area (cm^2), days to flowering, number of panicle plant^{-1}, panicle Length (cm), number of filled grains panicle^{-1}, number of unfilled grains panicle^{-1}, 1000-grain weight (g), Grain yield g plant^{-1}, leaf dry matter (g), root dry matter (g), stem dry matter (g) of the selected rice cultivar (BRRI dhan67) .The results are presented in figures and tables in this chapter and their possible interpretations are done as follows-

4.1 Plant Height

Effects of salinity

The plant height of BRRI dhan67 decreased as the level of salinity increased (Figure 3 and Appendix IV) at different days after transplanting (DAT). The plant height was highest (76.49, 103.7, 124.7 and 122.7 cm respectively) at 30 DAT, 60 DAT, 90 DAT and at harvest in 0 dSm^{-1}, at 30 DAT the highest plant height in S_0 treatment which was statistically similar with S_1 (70.87 cm) .The shortest plant height (57.92, 73.23, 93.41 and 91.97 cm respectively) was obtained by 8 dSm^{-1} (S_3) at 30 DAT, 60 DAT, 90 DAT and at harvest (57.92, 73.23, 93.41 and 91.97 cm respectively). Different levels of salinity have significant influence on plant height. Salinity level S_3 showed lower height of BRRI dhan 67 in every stages of plant.



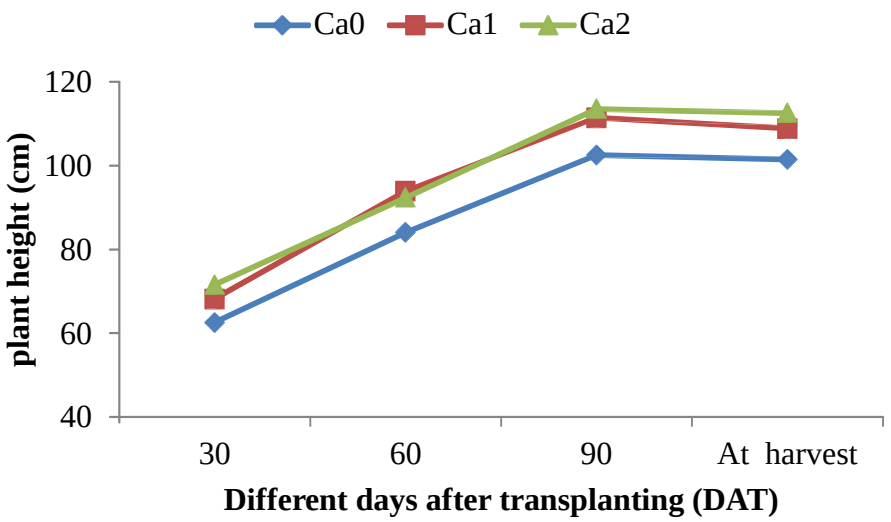
S_0 =0 dSm^{-1}, S_1 = 4 dSm^{-1}, S_2 =6 $dSm^{-1}$and S_3 =8 dSm^{-1}

Figure 3. Effect of different salt concentrations on the plant height of rice at different days after transplanting (LSD $_{(0.05)}$ =5.76, 8.10, 8.03 and 9.09 at 30, 60, 90 DAT and at harvest, respectively)

Hasanuzzaman *et al.* (2009) observed that plant height is negatively influenced by the increase of salinity levels in the rice varieties.

Effects of Calcium (Ca)

The plant height of BRRI dhan67 differed significantly due to the different doses of Ca over all the levels of salinity (Figure 4).The highest plant height at 30 DAT, 90 DAT and at harvest (71.63, 113.6 and 112.6 cm respectively) was given by Ca_2 and 60 DAT (93.95 cm) was given by Ca_1 and the shortest plant height at 30 DAT, 60 DAT, 90 DAT and at harvest (62.60, 84.11, 102.6 and 101.5 cm respectively) was obtained by Ca_0 (Figure 4 & Appendix V)At 30 DAT, the highest plant height (71.63cm) was observed from the Ca_2 treatment which was statistically similar with Ca_1 (68.26 cm),at 60 DAT the highest plant height (93.95 cm) was observed from the Ca_1 treatment which was statistically similar with Ca_2 (92.44 cm) whereas, the lowest plant heights given by Ca_0 at 30 DAT, 60 DAT, 90 DAT and at harvest were statistically dissimilar from plant heights obtained by Ca_1 and Ca_2 at every stage.



Ca_0 = 0 ppm of Ca, Ca_1 = 80 ppm of Ca, Ca_2 = 160 ppm of Ca

Figure 4. Effect of different calcium levels on the plant height of rice at different das after transplanting (LSD $_{(0.05)}$ =4.99, 7.01, 6.96 and 7.87 at 30, 60, 90 DAT and at harvest, respectively)

Rahman *et al*. (2016a) observed that Ca application improved plan height of rice.

Combined effect of salinity and Calcium

The effect of application of different doses of Ca on plant height of BRRI dhan67 at different salinity levels was found significant. (Table 4.1) At 30 DAT the highest plant height (80.07 cm) was recorded in S_0Ca_2 while at 60 DAT the highest plant height (106.4 cm) was recorded in S_0Ca_2 , at 90 DAT the highest plant height (128.0 cm) was recorded in S_0Ca_2 and at harvest the highest plant height (126.6 cm) was recorded in S_0Ca_2 .At 30 and 60 DAT, 90 DAT and at harvest the lowest plant height (53.68, 63.37, 82.11 and 82.00 cm respectively) was recorded in S_3Ca_0 (8dSm^{-1} treated with 0 ppm of Ca). At 30 DAT, the highest plant height (80.07 cm) was observed from the S_0Ca_2 treatment which was statistically similar with S_0Ca_0, S_0Ca_1, S_1Ca_1, S_1Ca_2 are observed (72.10, 77.31, 71.61 and 75.05 cm respectively).

Table 4.1. Interaction effect of different salt concentrations and calcium levels on the plant height of rice at different days after transplanting

Treatment combinations	Plant height (cm) at different days after transplanting DAT)			
	30	60	90	At harvest
S_0Ca_0	72.10 a-d	101.2 ab	121.1 ab	118.6 a-c
S_0Ca_1	77.31 ab	103.5 ab	125.0 a	122.9 ab
S_0Ca_2	80.07 a	106.4 a	128.0 a	126.6 a
S_1Ca_0	65.94 c-e	90.14 b-d	107.9 bc	105.8 c-e
S_1Ca_1	71.61 a-d	99.50 ab	115.4 ab	111.7 a-d
S_1Ca_2	75.05 a-c	96.61 ab	117.7 ab	114.7 a-d
S_2Ca_0	58.69 ef	81.76 cd	99.02 c	99.55 de
S_2Ca_1	66.43 c-e	93.24 a-c	107.7 bc	107.1 cde
S_2Ca_2	69.03 b-d	90.00 b-d	108.5 bc	109.1 b-e
S_3Ca_0	53.68 f	63.37 e	82.11 d	82.00 f
S_3Ca_1	57.70 ef	79.54 cd	97.77 c	93.77 ef
S_3Ca_2	62.37 d-f	76.78 de	100.3 c	100.1 de
$LSD_{(0.05)}$	**9.98**	**14.02**	**13.91**	**15.7**
Significance level	*	*	*	*
CV (%)	**8.73**	**9.18**	**7.52**	**8.64**

S_0 =0 dSm^{-1}, S_1 = 4 dSm^{-1}, S_2 =6 $dSm^{-1}$and S_3 =8 dSm^{-1}

Ca_0 = 0 ppm of Ca, Ca_1 = 80 ppm of Ca, Ca_2 = 160 ppm of Ca**,** *-Significant at 5% level, NS-Non Significant

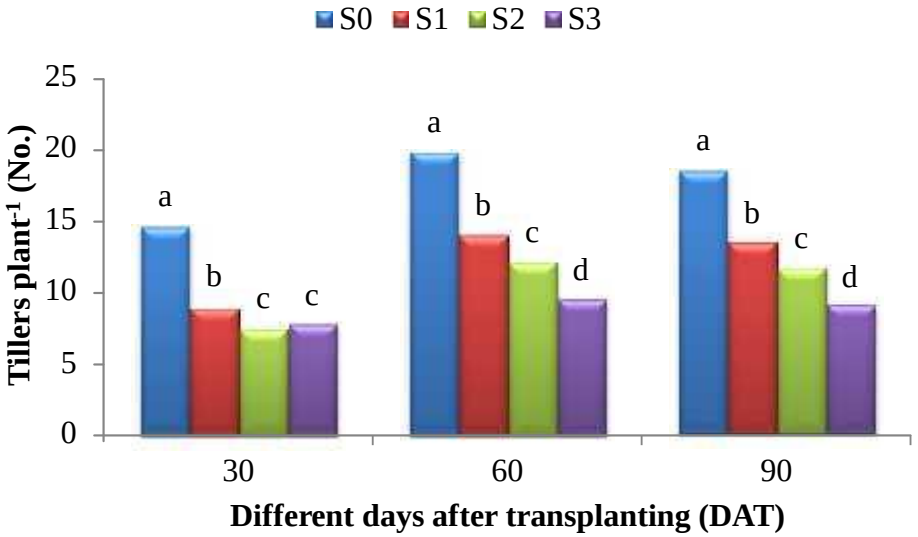
At 60 DAT, the highest plant height obtained by S_0Ca_2 (106.4 cm) which was statistically similar with S_0Ca_0, S_0Ca_1, S_1Ca_1, S_1Ca_2, S_2Ca_1 (101.2, 103.5, 99.50, 96.61and 93.24 cm respectively) in 90 DAT the highest plant height obtained by S_0Ca_2 (128.0 cm) which was statistically with S_0Ca_0, S_0Ca_1, S_1Ca_1, S_1Ca_2 is observed (121.1, 125.0, 115.4 and 117.7 cm respectively), and at harvest the highest plant height 126.6 (S_0Ca_2) cm it was statistically similar with S_0Ca_0, S_0Ca_1, S_1Ca_1, S_1Ca_2 are observed (118.6, 122.9, 111.7, 114.7 cm respectively). At 30 DAT the lowest plant height is 53.68 cm (S_3Ca_0) which was statistically similar with S_2Ca_0, S_3Ca_1, S_3Ca_2 is observed (58.69, 57.70, 62.37 cm respectively), at 60 DAT the lowest plant height

obtained by S_3Ca_0 (63.37 cm) statistically similar with S_3Ca_2 is observed (76.78 cm). From this experiment it was observed that calcium increased the plant height as compared with control where the best result was found from 160ppm concentration of calcium. These results are in agreement with that of Rahman *et al.* (2016a) who found that the supplementation of Ca increased plant height, where this growth component of rice adversely affected by increasing salinity. The increasing levels of Ca application improved plant height, tiller numbers, shoot dry weight of both salt tolerant and susceptible cultivars and this beneficial effect of Ca application under saline conditions may be attributed to its influence on net photosynthesis (Cha-um *et al*. 2012).

4.2Number of tillers plant^{-1}

Effects of salinity

Number of tiller plant^{-1} of BRRI dhan67 decreased as the level of salinity increased due to the mean effect of different sort of Ca applications (Figure 5 Appendix V) under different salinity at different days after transplanting (DAT). At 30 DAT, 60 DAT and 90 DAT the highest number of tillers plant^{-1} (14.56, 19.78 and 18.44 respectively) was recorded in S_0 (0 dSm^{-1}). The lowest number of tillers hill^{-1} at 60 DAT and 90 DAT (9.556, 9.111respectively) was obtained from S_3 (8 dSm^{-1}) (Appendix V).The lowest number of tillers hill^{-1} at 30 DAT was observed in Treatment S_2 (7.444) was statistically similar with S_3 (Figure 5).This result is similar with Govindaraju and Balakrishnan (2002) who indicated that plant height, number of productive tillers, chlorophyll content and photosynthetic ability of rice decreased with increase of salinity.

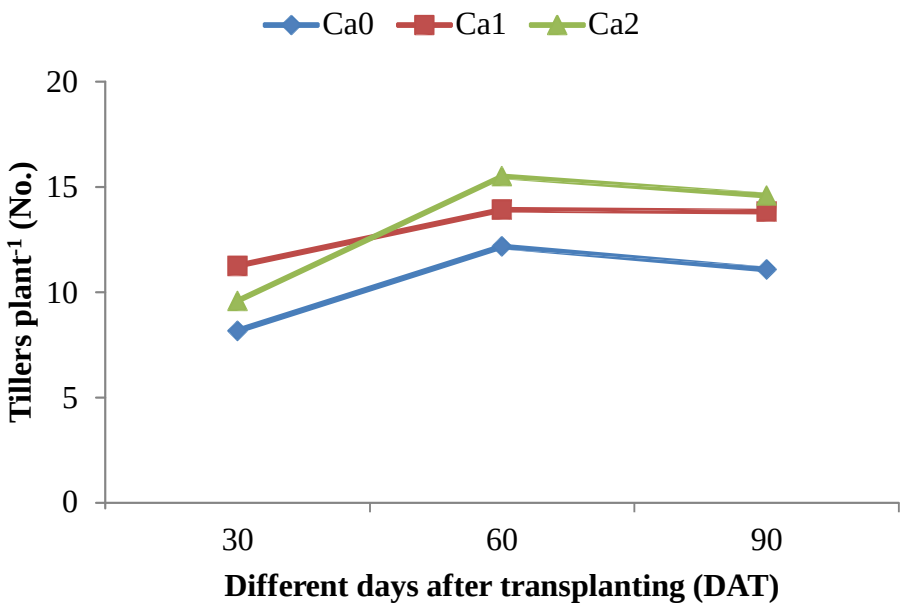


S_0 =0 dSm^{-1}, S_1 = 4 dSm^{-1}, S_2 =6 $dSm^{-1}$and S_3 =8 dSm^{-1}

Figure 5. Effect of different salt concentrations on the tillers plant^{-1} of rice at different das after transplanting (LSD $_{(0.05)}$ =0.76, 1.10 and 1.28 at 30, 60 and 90 DAT, respectively)

Effects of calcium (Ca)

Number of tiller plant^{-1} of BRRI dhan67 decreased as the level of salinity increased due to the mean effect of different sort of Ca applications (Figure 6 and Appendix V) at different days after transplanting (DAT).At 60 DAT and 90 DAT the highest number of tillers plant^{-1} (15.50 and 14.58 respectively) was recorded in Ca_2 (160 ppm) and at 30 DAT the highest number of tillers plant^{-1} was recorded in Ca_1 (80 ppm).At 90 DAT the highest number of tillers plant^{-1} was observed from the treatment Ca_2 which was statistically similar with Ca_1 (14.58, 13.83 respectively). Bohra and Doerffling (1993) observed that the increasing levels of K application improved the tiller numbers both salt tolerant and susceptible cultivars. In this experiment it is found that calcium increased the number of tiller significantly.



Ca_0 = 0 ppm of Ca, Ca_1 = 80 ppm of Ca, Ca_2 = 160 ppm of Ca

Figure 6. Effect of different calcium levels on the tillers plant^{-1} of rice at different das after transplanting (LSD $_{(0.05)}$ =0.66, 0.95 and 1.11 at 30, 60 and 90 DAT, respectively)

Combined effect of salinity and calcium

The effect of different doses of Ca on number of tillers plant^{-1} of BRRI dhan67 at different stages. The highest number of tillers plant^{-1} at 30 DAT, 60 DAT and 90 DAT (15.00, 21.67 and 20.00 respectively) was recorded in S_0Ca_2 (160 ppm Ca). The lowest number of tillers plant^{-1} at 60 DAT and 90 DAT was obtained from S_3Ca_0 (7.67 and 6.67 respectively), at 30 DAT S_2Ca_2 (6.00) gives the lowest number of tillers plant^{-1} which is statistically similar with S_3Ca_0. It was observed that S_1Ca_1, S_1Ca_2 gave higher number of tillers plant^{-1} than S_1Ca_0, S_2Ca_1, S_2Ca_2 gave higher number of tillers plant^{-1} than S_2Ca_0 and S_3Ca_1, S_3Ca_2 gave higher number of tillers plant^{-1} than S_3Ca_0 at 30, 60 and 90 DAT which indicates mitigation of the stress caused by salt by application of calcium. Compared to sole saline treatments, plants provided with calcium gives better results in each case.

Table 4.2. Interaction effect of different salt concentrations and calcium levels on the tillers plant^{-1} of rice at different days after transplanting

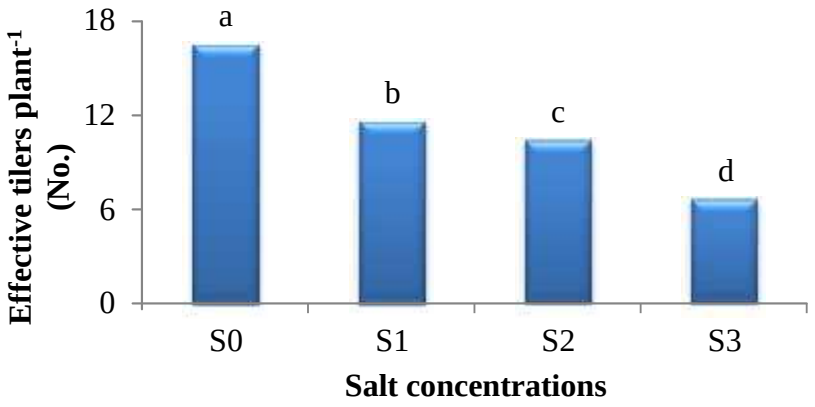
Treatment combinations	Tillers plant^{-1} (No.) at different days after transplanting (DAT)		
	30	60	90
S_0Ca_0	11.00 c	18.33 b	16.33 b
S_0Ca_1	17.67 a	19.33 b	19.00 a
S_0Ca_2	15.00 b	21.67 a	20.00 a
S_1Ca_0	7.67 f	12.67 de	12.00 de
S_1Ca_1	10.00 cd	14.00 cd	14.33 bc
S_1Ca_2	9.00 de	15.33 c	14.00 cd
S_2Ca_0	7.67 f	10.00 f	9.33 f
S_2Ca_1	8.67 ef	12.67 de	12.33 c-e
S_2Ca_2	6.00 g	13.67 cd	13.33 cd
S_3Ca_0	6.33 g	7.67 g	6.67 g
S_3Ca_1	8.67 ef	9.67 f	9.67 f
S_3Ca_2	8.33 ef	11.33 ef	11.00 ef
LSD $_{(0.05)}$	**1.32**	**1.90**	**2.21**
Significance Level	*	*	*
CV (%)	**8.07**	**8.11**	**9.92**

S_0 =0 dSm^{-1}, S_1 = 4 dSm^{-1}, S_2 =6 dSm^{-1}and S_3 =8 dSm^{-1}

Ca_0 = 0 ppm of Ca, Ca_1 = 80 ppm of Ca, Ca_2 = 160 ppm of Ca, *-Significant at 5% level, NS-Non Significant

4.3 Number of effective tillers plant^{-1}

Effects of salinity



S_0 =0 dSm^{-1}, S_1 = 4 dSm^{-1}, S_2 =6 dSm^{-1}and S_3 =8 dSm^{-1}

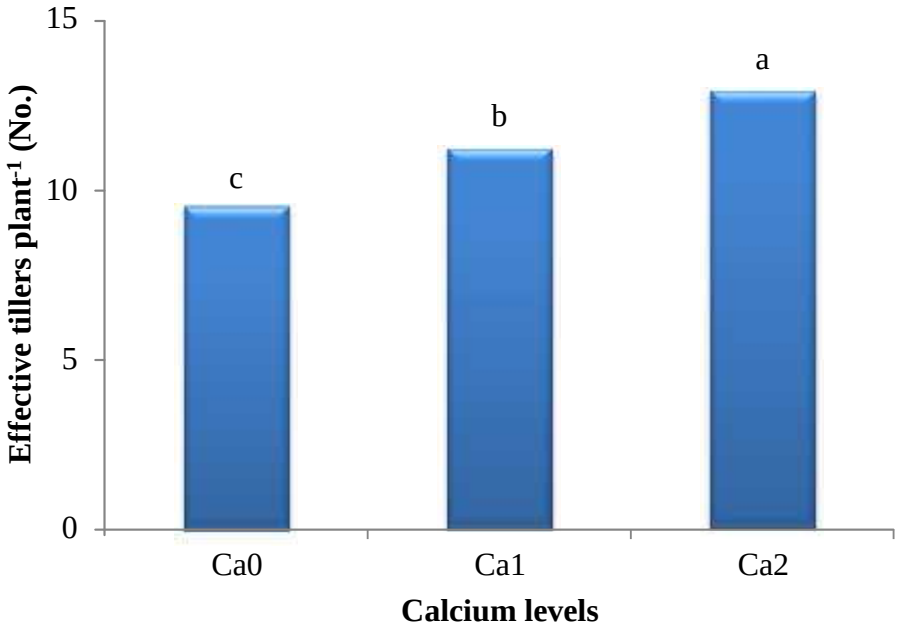
Figure 7. Effect of different salt concentrations on the effective tillers plant^{-1} of rice (LSD $_{(0.05)}$ = 1.05)

It was observed that, as the salinity level increased, the number of effective tillers plant^{-1} decreased significantly (Figure 7 and Appendix IX). Highest number of effective tillers plant^{-1} was recorded in S_0 (16.44) and lowest number of effective tillers plant^{-1} was found from S_3 (6.56) (Appendix VI). Similar result was found that tiller production gradually decreased with increased levels of salinity by Zeng and Shannon (2000).

Effects of calcium (Ca)

Application of calcium in different amount showed variation among them for number of effective tillers plant^{-1}. The highest number of effective tillers plant^{-1} (12.92) was recorded in Ca_2 (160 ppm Ca) which is statistically dissimilar with Ca_1 (11.25) and lowest was in Ca_0 (9.50) (Figure 8 and Appendix VI). Tillers number increase with the increase of Calcium

.



Ca_0 = 0 ppm of Ca, Ca_1 = 80 ppm of Ca, Ca_2 = 160 ppm of Ca

Figure 8. Effect of different calcium levels on the effective tillers plant^{-1} of rice (LSD $_{(0.05)}$ = 0.91)

Combined effect of salinity and calcium

The effect of different doses of Ca on number of effective tillers plant^{-1} of BRRI dhan67 at different salinity levels was found non-significant. The highest number of effective tillers plant^{-1} (26.00) was recorded in S_0Ca_2 (0 dSm^{-1} of Salt treated with 160ppm of Ca). The lowest number of effective tillers plant^{-1} was recorded in S_3Ca_0 (4.67) in 8 dSm^{-1} salinity. Other treatments also gave satisfactory result as it was observed that S_1Ca_1 (11.00) and S_1Ca_2 (13.00) gave higher number of effective tillers plant^{-1} than S_1Ca_0 (10.67), S_2Ca_1 (10.67) and S_2Ca_2 (11.67) gave higher number of effective tillers plant^{-1} than S_2Ca_0 (8.67), S_3Ca_1 (7.00) and S_3Ca_2 (8.00) gave higher number of effective tillers plant^{-1} than S_3Ca_0 (4.67) which indicates mitigation of the stress caused by salt by application of calcium.

Table 4.3. Interaction effect of different salt concentrations and calcium levels on the days to flowering, panicles plant^{-1}, effective tillers plant^{-1}, ineffective tillers plant^{-1} and panicle length of rice

Treatment combinations	Days to flowering (DAT)	Effective tillers plant^{-1} (No.)	Ineffective tillers plant^{-1} (No.)	Panicle length (cm)
S_0Ca_0	61.33 e	14.00 c	2.67 b	25.83 bc
S_0Ca_1	54.00 f	16.33 b	2.33 bc	27.69 ab
S_0Ca_2	51.33 g	19.00 a	1.33 e	29.03 a
S_1Ca_0	63.00 c-e	10.67 e	2.33 bc	23.59 c-e
S_1Ca_1	62.00 de	11.00 e	2.00 cd	24.87 b-d
S_1Ca_2	63.33 c-e	13.00 cd	1.67 de	25.90 bc
S_2Ca_0	63.33 c-e	8.667 f	2.33 bc	19.32 fg
S_2Ca_1	65.00 a-c	10.67 e	1.67 de	21.57 ef
S_2Ca_2	63.67 b-e	11.67 de	1.63 de	22.38 d-f
S_3Ca_0	66.00 ab	4.67 g	3.27 a	17.95 g
S_3Ca_1	66.67 a	7.00 f	2.67 b	19.67 fg
S_3Ca_2	64.33 a-d	8.00 f	2.33 bc	20.43 fg
LSD $_{(0.05)}$	**2.565**	**1.82**	**0.44**	**3.12**
Significance Level	**NS**	*****	*****	*****
CV (%)	**2.44**	**9.55**	**11.93**	**7.95**

S_0 =0 dSm^{-1}, S_1 = 4 dSm^{-1}, S_2 =6 dSm^{-1}and S_3 =8 dSm^{-1}

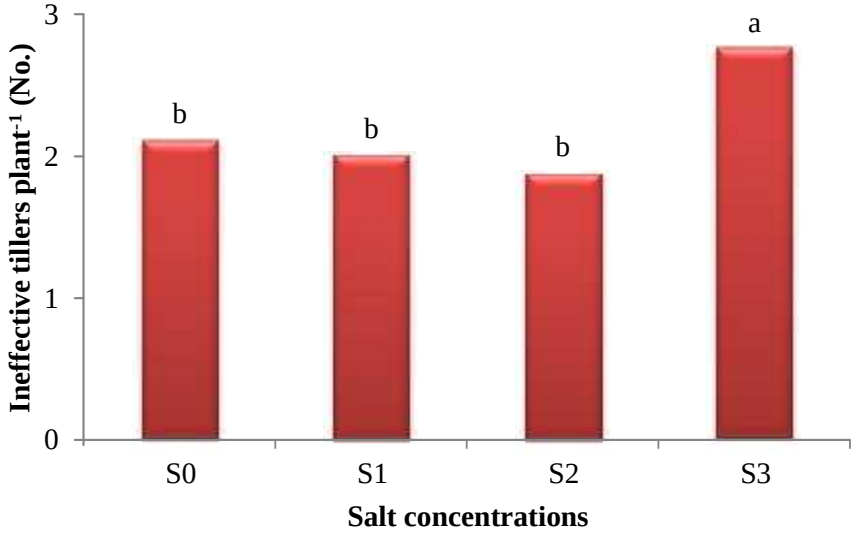
Ca_0 = 0 ppm of Ca, Ca_1 = 80 ppm of Ca, Ca_2 = 160 ppm of Ca, *-Significant at 5% level, NS-Non Significant

4.4 Number of ineffective tillers plant^{-1}

Effects of salinity

It was observed that, as the salinity level increased, the number of ineffective tillers plant^{-1} decreased firstly then increased significantly (Figure 9 and Appendix IX).

Highest number of ineffective tillers plant^{-1} was recorded in S_3 (2.76) which was statistically different from S_0, S_1 and S_2 (2.11, 2.00 and 1.88) and lowest number of ineffective tillers plant^{-1} was found from S_0 (1.88) statistically same with S_1 and S_2 (2.00 and 1.88) (Appendix VI).

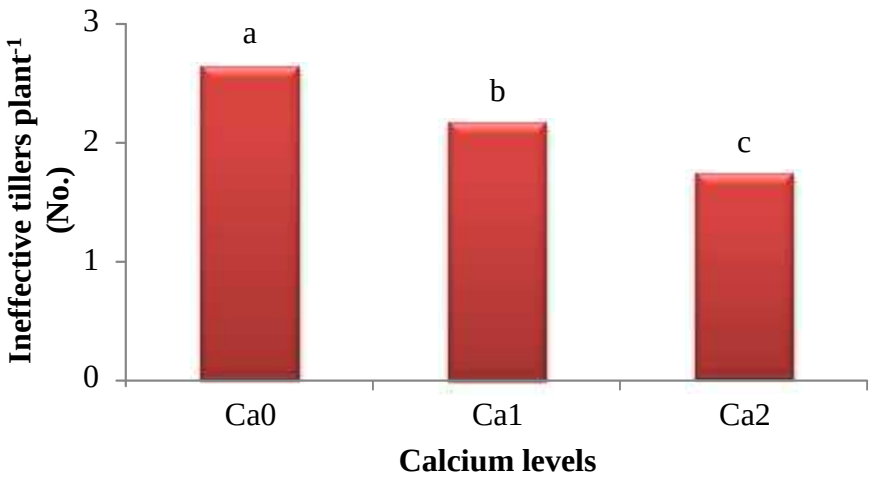


S_0 =0 dSm^{-1}, S_1 = 4 dSm^{-1}, S_2 =6 dSm^{-1}and S_3 =8 dSm^{-1}

Figure 9. Effect of different salt concentrations on the ineffective tillers plant^{-1} of rice (LSD $_{(0.05)}$ = 0.25)

Effects of calcium (Ca)

Application of calcium in different amount showed variation among them for number of ineffective tillers plant^{-1}. The highest number of ineffective tillers plant^{-1} (2.65) was recorded in Ca_0 (0 ppm of calcium) and the lowest in Ca_2 (1.74). (Figure 10 and Appendix IX). Aslam *et al*. (2001) conducted a field experiment to evaluate the response of rice crop to calcium fertilization in saline-sodic soil during 2001. The results of that experiment indicated that increasing rates of Calcium fertilizer increased the number of tillers plant^{-1}, Panicle length (cm), paddy weight and paddy as well as straw yield significantly.



Ca_0 = 0 ppm of Ca, Ca_1 = 80 ppm of Ca, Ca_2 = 160 ppm of Ca

Figure 10. Effect of different calcium levels on the ineffective tillers plant^{-1} of rice (LSD $_{(0.05)}$ = 0.22)

Combined effect of salinity and calcium

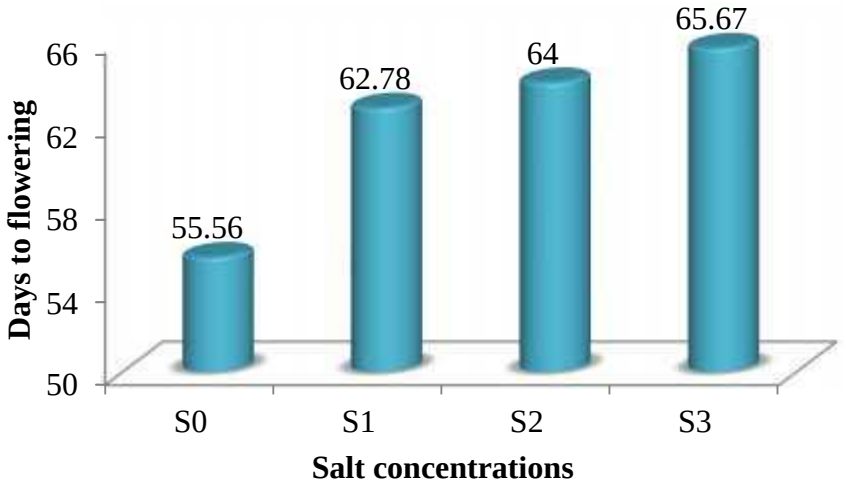
The effect of different levels of Ca on number of ineffective tillers plant^{-1} of BRRI dhan67 at different salinity levels was found significant. The highest number of ineffective tillers plant^{-1} (3.27) was recorded in S_3Ca_0 (8 dSm^{-1} salt) and lowest number of ineffective tillers plant^{-1} was found from S_0Ca_2 (1.33) and both are statistically significant. In this experiment it was observed that S_1Ca_1 (2.00) and S_1Ca_2 (1.67) gave lower number of ineffective tillers plant^{-1} than S_1Ca_0 (2.33), S_2Ca_1 (1.67) and S_2Ca_2 (1.63) gave lower number of ineffective tillers plant^{-1} than S_2Ca_0 (2.33), S_3Ca_1 (2.67) and S_3Ca_2 (2.33) gave lower number of ineffective tillers plant^{-1} than S_3Ca_0 (3.27).

4.5 Days to flowering (DAT- Days after transplanting)

Effect of salinity

It was observed that, as the salinity level increased, the number of days to flower increased significantly (Figure 9 and Appendix IX). Highest number of days required was recorded in S_3 (65.67 DAT) which was statistically different from S_0, S_1 and S_2 (55.56, 62.78 and 64.00 DAT) and lowest number of days required to flower was found from S_0 (55.56) .S_1 and S_2 are statistically similar (62.78 and 64.00 DAT)

(Appendix X). Rao *et al.* (2008) found the same result that flowering is delayed with the increase of salinity and it reduces grain quality and quantity ultimately.

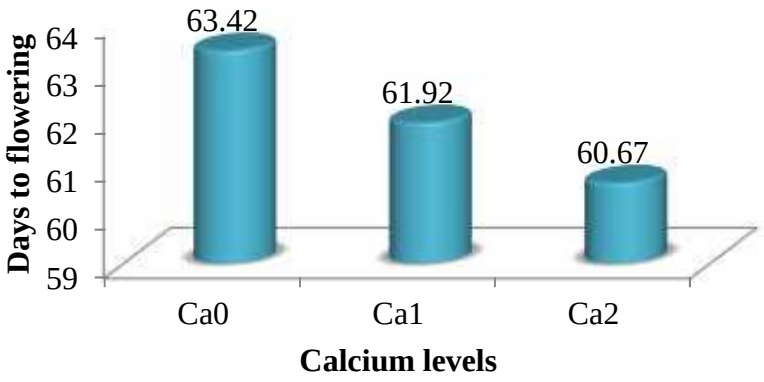


S_0 =0 dSm^{-1}, S_1 = 4 dSm^{-1}, S_2 =6 $dSm^{-1}$and S_3 =8 dSm^{-1}

Figure 11. Effect of different salt concentrations on the days to flowering of rice (LSD $_{(0.05)}$ = 1.48)

Effects of calcium (Ca)

Application of calcium in different amount showed variation among them for number of days required for flowering. The highest number of days required to flower (63.42 DAT) was recorded in Ca_0 (0 ppm of calcium) and the lowest in Ca_2 (60.67 DAT) which is statistically similar with Ca_1 (61.92 DAT). (Figure 12 and Appendix IX).



Ca_0 = 0 ppm of Ca, Ca_1 = 80 ppm of Ca, Ca_2 = 160 ppm of Ca

Figure 12. Effect of different calcium levels on the days to flowering of rice (LSD $_{(0.05)}$ = 1.28)

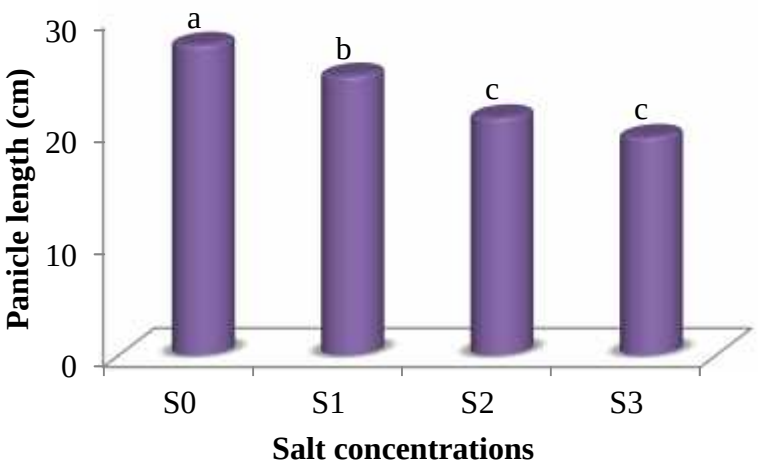
Combined effect of salinity and calcium

The effect of different doses of Ca on number of days required for first flowering of BRRI dhan67 at different salinity levels was found significant. The highest number of days after transplanting required to heading or flowering (66.67 DAT) was recorded in S_3Ca_1 (8 dSm^{-1} of Salt treated with 80 ppm of Ca). The lowest number of days required to flower was recorded in S_0Ca_2 (51.33 DAT) in 0 dSm^{-1} salinity treated with 160 ppm of calcium. In this experiment it was observed that days required to flowering was reduced by calcium supplementation mostly at higher level of salinity (8 dSm^{-1}) as we can see plants under the treatment S_3Ca_2 (64.33 DAT) required shorter duration of time to flower than S_3Ca_0 (66.00 DAT).

4.6 Panicle length (cm)

Effects of salinity

It was observed that, as the salinity level increased, the length of panicle decreased significantly (Figure 13 and Appendix IX). Highest length of panicle was recorded in S_0 (27.52cm) and lowest length of panicle was recorded in S_3 (19.35 cm) which was statistically same with S_2 (21.09 cm) (Appendix VI).

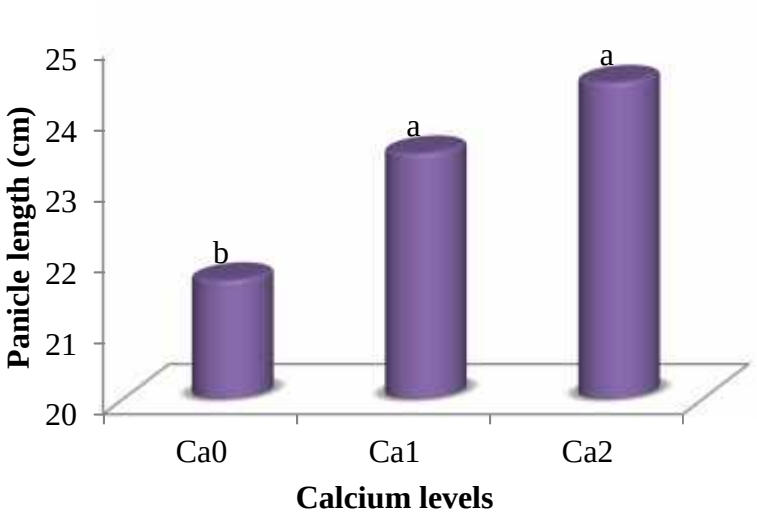


S_0 =0 dSm^{-1}, S_1 = 4 dSm^{-1}, S_2 =6 $dSm^{-1}$and S_3 =8 dSm^{-1}

Figure 13. Effect of different salt concentrations on the panicle length of rice (LSD $_{(0.05)}$ = 1.80)

Effects of calcium (Ca)

The application of calcium in different amount showed variation among the length of panicle. Highest length of panicle was recorded in Ca_2 (24.44 cm) which was statistically same with Ca_1 (23.45 cm) and lowest length of panicle was recorded in Ca_0 (21.67 cm) (Appendix IX).



Ca_0 = 0 ppm of Ca, Ca_1 = 80 ppm of Ca, Ca_2 = 160 ppm of Ca

Figure 14. Effect of different calcium levels on the panicle length of rice (LSD $_{(0.05)}$ =1.56)

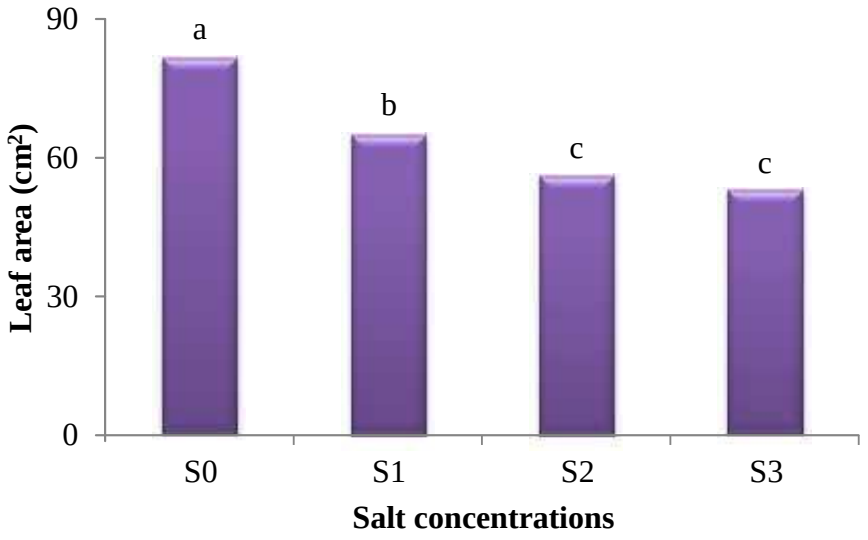
Combined effect of salinity and calcium

The effect of application of Ca in different amount under different salinity level of BRRI dhan67 showed the result significantly. Highest length of panicle was recorded in S_0Ca_2 (29.03 cm) and lowest length of panicle was recorded in S_3Ca_0 (17.95 cm) which was statistically similar with S_2Ca_0, S_3Ca_1, S_3Ca_2 (19.32, 19.67, 20.43 cm) (Table 4.3). Ebrahimi *et al.* (2012) reported that the different soil salinity levels significantly decrease panicle length with the increasing level of salinity. Aslam *et al.* (2001) found same effect by calcium under salt stress.

4.7 Leaf Area

Effects of salinity

Leaf area plant^{-1} was significantly affected by different salinity levels. Leaf area (cm^2) decreased with increasing concentration of salinity in BRRI dhan67 (Figure 15 and Appendix VIII). The maximum leaf area (81.48 cm^2) was recorded from control, S_0 (without salt) where the minimum leaf area (52.78 cm^2) was found from S_3 (8 dSm^{-1}). Similar result was also reported by Munns and Tester (2008). According to Hernandez *et al.* (2003) salt stress inhibited the cell division and cell expansion, consequently leaf expansion and as a result leaf area is reduced.

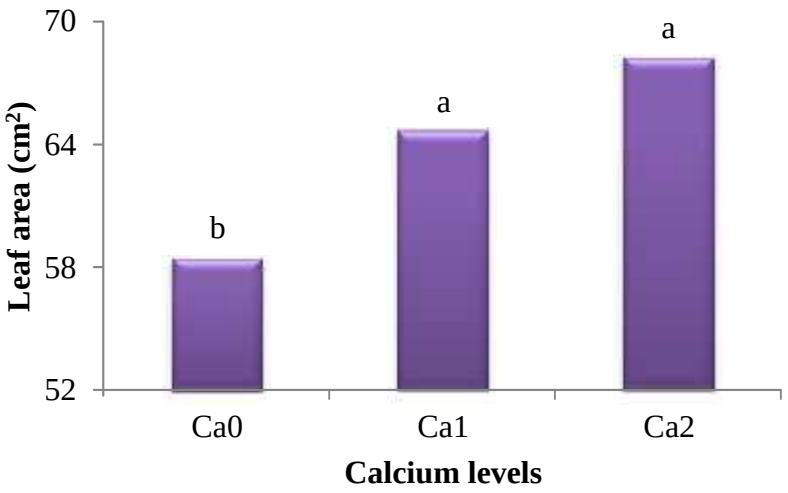


S_0 =0 dSm^{-1}, S_1 = 4 dSm^{-1}, S_2 =6 dSm^{-1}and S_3 =8 dSm^{-1}

Figure 15. Effect of different salt concentrations on the leaf area of rice (LSD $_{(0.05)}$ = 5.16)

Effects of calcium (Ca)

Different levels of calcium affected significantly on leaf area plant^{-1} (Figure 16 and Appendix VIII). The highest leaf area plant^{-1}(68.31 cm^2) was found from Ca_2 where the lower leaf area (58.49 cm^2) plant^{-1} was recorded from Ca_0.



Ca_0 = 0 ppm Ca , Ca_1 = 80 ppm of Ca, Ca_2 = 160 ppm of Ca

Figure 16. Effect of different calcium levels on the leaf area of rice ($LSD_{(0.05)}$ =4.47)

Combined effect of salinity and calcium

The combined effect of salt and calcium was a significant effect on the leaf area plant^{-1} (Table 4.4 and Appendix VIII). The maximum leaf area (84.66 cm^2) was recorded from S_0Ca_2 which was statistically similar (77.82 and 81.95 cm^2) to S_0Ca_0 and S_0Ca_1. The leaf area was found higher with 160 ppm concentration of calcium in case of 0, 4, 6 and 8 dSm^{-1} (84.66, 69.41, 61.23 and 57.95 cm^2 respectively) treated plants. From the above results it can be concluded that the calcium has important role in mitigating salt stress.

Table 4.4. Interaction effect of different salt concentrations and calcium levels on the root length, leaf area, leaf membrane stability and relative water content (%) of rice

Treatment combinations	Leaf area (cm^2)	Leaf membrane stability	Relative water content (%)
S_0Ca_0	77.82 ab	81.02 bc	87.56 ab
S_0Ca_1	81.95 a	87.26 ab	91.40 a
S_0Ca_2	84.66 a	88.72 a	93.61 a
S_1Ca_0	61.07 c-e	64.87 e	82.89 a-c
S_1Ca_1	64.80 cd	73.99 d	87.06 ab
S_1Ca_2	69.41 bc	79.93 cd	90.14 ab
S_2Ca_0	49.11 fg	52.22 f	72.41 cd
S_2Ca_1	57.29 d-f	61.54 e	78.41 b-d
S_2Ca_2	61.23 c-e	63.49 e	81.69 a-c
S_3Ca_0	45.97 g	30.49 h	60.13 e
S_3Ca_1	54.41 e-g	32.89 h	67.90 de
S_3Ca_2	57.95 d-f	44.82 g	71.35 c-e
LSD $_{(0.05)}$	**8.94**	**6.58**	**11.98**
Significance Level	*	*	NS
CV (%)	**8.28**	**6.13**	**8.8**

S_0 =0 dSm^{-1}, S_1 = 4 dSm^{-1}, S_2 =6 $dSm^{-1}$and S_3 =8 dSm^{-1}

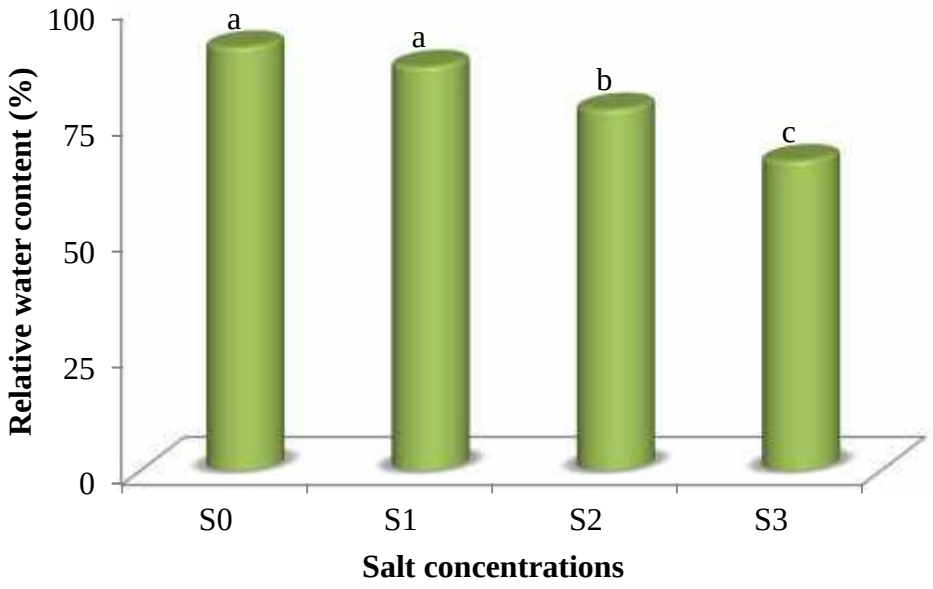
Ca_0 = 0 ppm of Ca, Ca_1 = 80 ppm of Ca, Ca_2 = 160 ppm of Ca, *-Significant at 5% level, NS-Non Significant

4.8 Relative water content (RWC)

Effects of salinity

The analysis of various salinity levels on relative leaf water content showed that, increase in salinity concentration caused significant reduction in RWC (Figure: 17 Apendix VIII). The maximum leaf relative water content (90.86) was recorded from control, S_0 (without salt) where the minimum leaf relative water content (66.46) was

found from S_3 (8 dSm^{-1}). A decrease in RWC indicates a loss of turgor that results in limited water availability for cell extension processes. Steudle (2000) reported that in transpiring plants, water is thought to come from the soil to the root xylem through apoplastic pathway due to hydrostatic pressure gradient. However, under salt stressed condition, this situation changes because of the restricted transpiration. Under these situations, more of water follows cell-to-cell path, flowing across membranes of living cells (Vysotskaya *et al*. 2010).

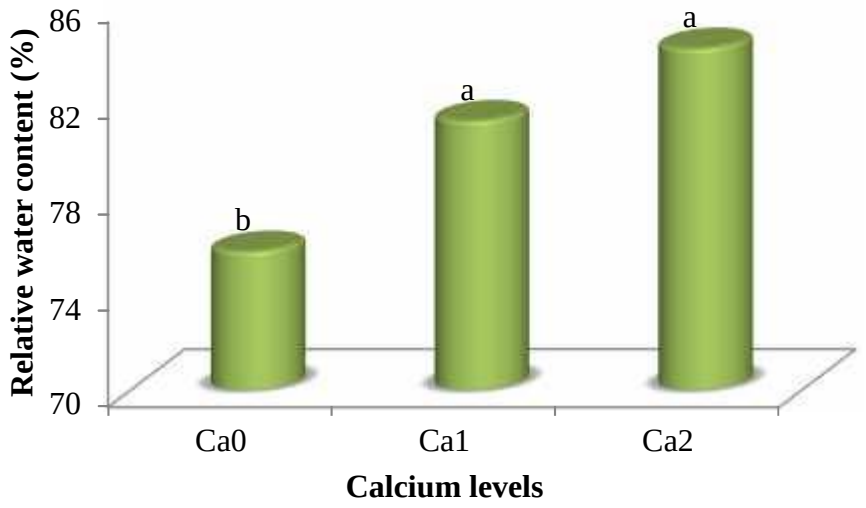


S_0 =0 dSm^{-1}, S_1 = 4 dSm^{-1}, S_2 =6 $dSm^{-1}$and S_3 =8 dSm^{-1}

Figure 17. Effect of different salt concentrations on the relative water content of rice (LSD $_{(0.05)}$ = 6.92)

Effects of calcium (Ca)

Different levels of calcium affected significantly on leaf relative water content (Figure 18 and Appendix VIII). The highest leaf relative water content (84.20) was found from Ca_2 where the lower leaf relative water content (75.75) was recorded from Ca_0.



Ca_0 = 0 ppm of Ca , Ca_1 = 80 ppm of Ca, Ca_2 = 160 ppm of Ca

Figure 18. Effect of different calcium levels on the relative water content of rice

(LSD $_{(0.05)}$ = 5.99)

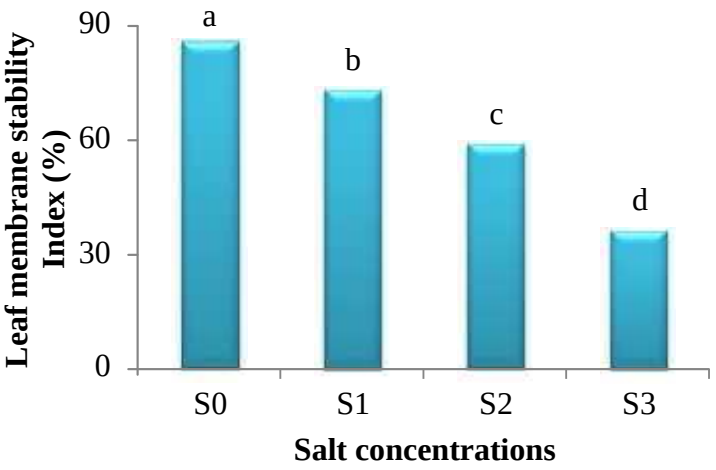
Combined effect of salinity and calcium

By increasing the concentration of calcium, the relative water content was increased (Table 4.4 and Appendix VIII). The maximum leaf relative water content (93.61) was recorded from S_0Ca_2. The leaf area was found higher with 160 ppm concentration of calcium in case of 0, 4, 8 and 8 dSm^{-1} treated plants. In this experiment it was observed that S_1Ca_1 (87.06), S_1Ca_2 (90.14) gave better result than S_1Ca_0 (82.89), S_2Ca_1 (78.41), S_2Ca_2 (81.69) gave better result than S_2Ca_0 (72.41) and S_3Ca_1 (67.90), S_3Ca_2 (71.35) gave better result than S_3Ca_0 (60.13). From the above results it can be concluded that the calcium has important role in mitigating salt stress. This result goes with harmony with the results found by Tabatabaeian (2014).

4.9 Leaf Membrane Stability Index

Effects of salinity

It was observed that, as the salinity level increased, the membrane stability of leaves decreased significantly (Figure 19). Highest membrane stability was recorded in S_0 (85.67) and lowest was recorded in S_3 (36.07) (Appendix VIII).

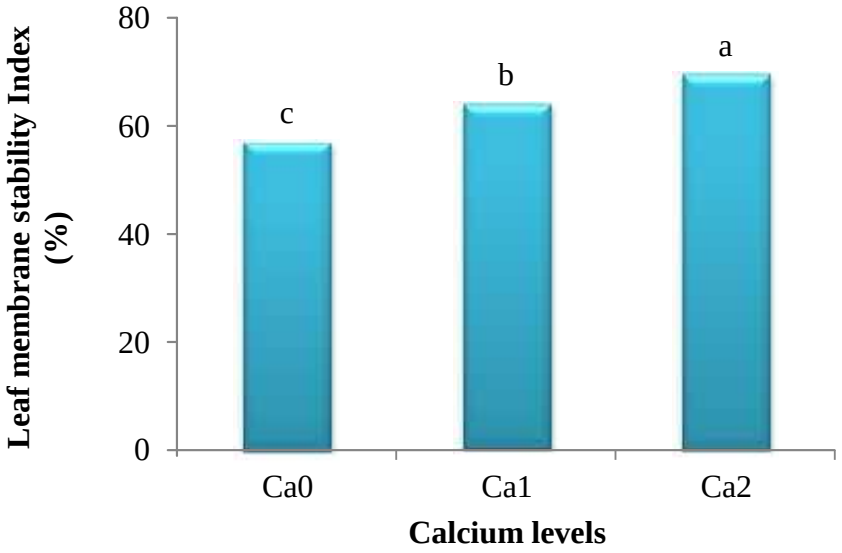


S_0 =0 dSm^{-1}, S_1 = 4 dSm^{-1}, S_2 =6 $dSm^{-1}$and S_3 =8 dSm^{-1}

Figure 19. Effect of different calcium levels on the leaf membrane stability of rice (LSD $_{(0.05)}$ = 3.29)

Effects of calcium (Ca)

The application of different amount of calcium showed variation among the membrane stability of leaves. Highest membrane stability was recorded in Ca_2 (69.24) and lowest was recorded in Ca_0 (57.15) (Appendix VIII).



Ca_0 = 0 ppm of Ca , Ca_1 = 80 ppm of Ca, Ca_2 = 160 ppm of Ca

Figure 20. Effect of different calcium levels on the leaf membrane stability of rice (LSD $_{(0.05)}$ = 3.29)

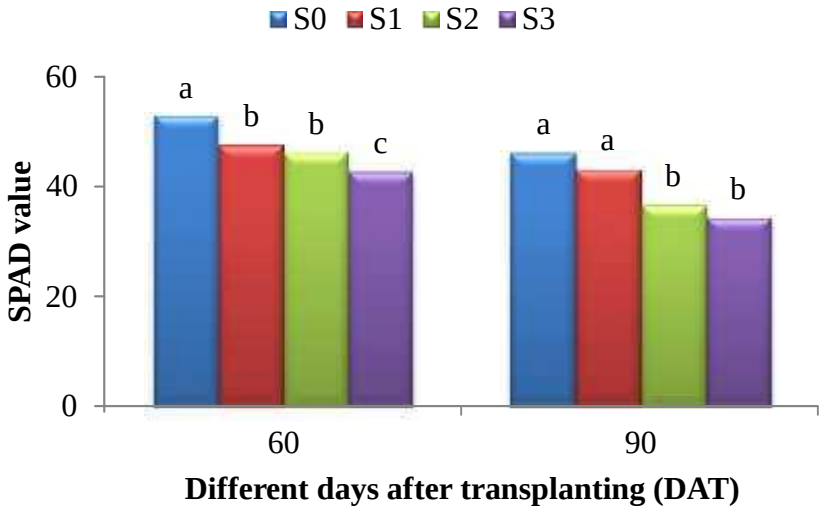
Combined effect of salinity and calcium

The combined effect of salt and calcium was a significant effect on the leaf membrane stability (Table 4.4 and Appendix VIII). The maximum leaf membrane stability (88.72) was recorded from S_0Ca_2 which was statistically similar (87.26) to S_0C_1. The lowest leaf membrane stability was found from S_3Ca_0 (30.49) which is statistically similar with S_3Ca_1 (32.89). In this experiment it was observed that S_1Ca_1 (73.99), S_1Ca_2 (79.93) gave better result than S_1Ca_0 (64.87), S_2Ca_1 (61.54) S_2Ca_2 (63.49) gave better result than S_2Ca_0 (52.22) and S_3Ca_1 (32.89), S_3Ca_2 (44.82) gave better result than S_3Ca_0 (30.49). .Increasing the salt concentration in the cells of plant caused the cellular leakages to be increased and membrane stability of tissue to be reduced. Therefore, we can see that the leaf membrane stability index in the treatments containing the calcium were significantly higher than other solutions. In this regard, Calcium acts as a secondary messenger and by effecting the membrane stability and activity of enzymes causes to protect the cells in stress terms. Thus, calcium plays an important role in the membrane stability under the salinity stress (Girija *et al*., 2002).

4.10 Chlorophyll content (SPAD Value)

Effects of salinity

Salinity treatment reduced total chlorophyll content (Figure 21 Appendix VI). The chlorophyll content (SPAD reading) in leaves decreased with increasing salinity levels. At 60 DAT, the highest chlorophyll content (52.79 SPAD units) was recorded from S_0 and the lowest value (42.60 SPAD units) was found from S_3. At 90 DAT, the highest chlorophyll content (46.07 SPAD units) was found from S_0 which was statistically similar (43.11 SPAD units) to S_1 and the lowest value (34.31 SPAD units) was observed in case of S_3 which was statistically similar (36.70 SPAD units) to S_2. From these results, it was found that the high levels of salinity (8 dSm^{-1}) induced a significant decrease in the total chlorophyll content as compared to control plants. Salt stress often causes alteration in photosynthetic pigment biosynthesis (Maxwel and Jhonson, 2000). Similar decrease in chl content was observed by Amirjani (2011) in rice.

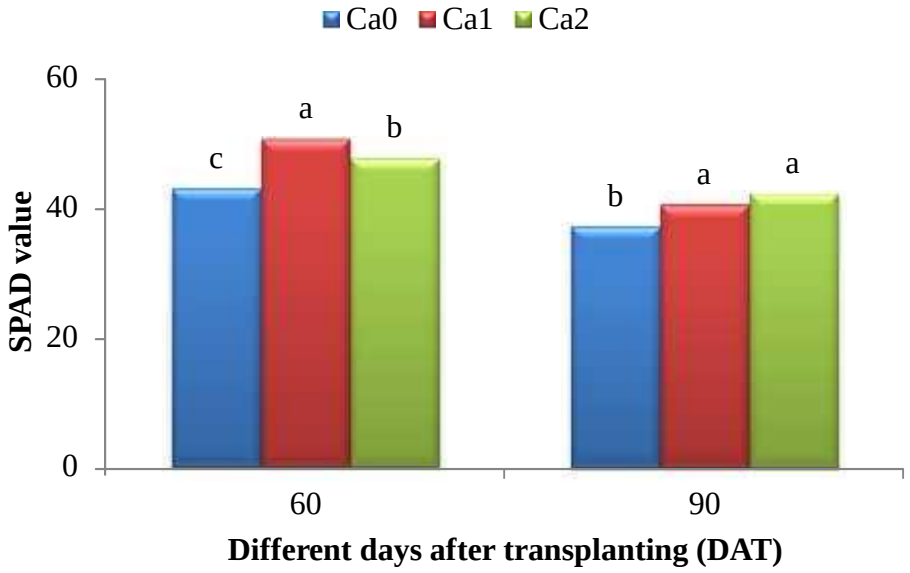


S_0 =0 dSm^{-1}, S_1 = 4 dSm^{-1}, S_2 =6 $dSm^{-1}$and S_3 =8 dSm^{-1}

Figure 21. Effect of different salt concentrations on the SPAD value of rice at different das after transplanting (LSD $_{(0.05)}$ = 3.30 and 3.20at 60 and 90 DAT, respectively)

Effects of calcium (Ca)

Significant effect of calcium on leaf chlorophyll content of BRRI dhan67 plant was found at 60 and 90 DAT (Figure 22 and Appendix VI). The highest value (50.73 SPAD units) was found from Ca_1 and lowest value (43.16 SPAD units) was recorded from Ca_0 at 60 DAT. At 90 DAT, the highest chlorophyll content (42.34 SPAD units) was found from Ca_2 which was statistically similar (40.57 SPAD units) to Ca_1. The lowest value (37.25 SPAD units) was recorded from Ca_0. Thus, calcium reduced the toxic effect on leaf chlorophyll content which was supported by Howladar and Rady (2012). This study suggests that, exogenous Ca^{2+} supply improves the total chlorophyll content in plant which was strongly related to the yield.



Ca_0 = 0 ppm Ca , Ca_1 = 80 ppm of Ca, Ca_2 = 160 ppm of Ca

Figure 22. Effect of different calcium levels on the SPAD value of rice at different das after transplanting (LSD $_{(0.05)}$ = 2.86 and 2.77 at 60 and 90 DAT, respectively)

Combined effect of salinity and calcium

The interaction effect between salinity and calcium levels on leaf chlorophyll content of BRRI dhan67 plant was statistically Non-significant at 60 and significant at 90 DAT (Table 4.5 and Appendix VI). At 60 DAT, the highest leaf chlorophyll content (55.88 SPAD units) was found from S_0Ca_1 which was statistically similar (54.99 SPAD units) to S_0Ca_2. The lowest value (39.61 SPAD units) was found from S_3Ca_0 which was statistically identical (45.12, 40.43 and 42.86 SPAD units) to S_1Ca_0, S_2Ca_0 and S_3Ca_2. At 90 DAT, the highest value (48.27 SPAD units) was observed in S_0Ca_2 which was statistically similar (45.97 SPAD units) to S_0Ca_1. The lowest value (35.44 SPAD units) was found from S_3Ca_0 which was statistically identical (34.74 and 28.90 SPAD units) to S_2Ca_0 and S_3Ca_1. In each case where calcium were supplemented (S_1Ca_1, S_1Ca_2, S_2Ca_1, S_2Ca_2, S_3Ca_1 and S_3Ca_2) with salt, mitigating effect was observed.

Table 4.5. Interaction effect of different salt concentrations and calcium levels on the SPAD value of rice at different days after transplanting

Treatment combinations	SPAD value at different days after transplanting (DAT)	
	60	90
S_0Ca_0	47.49 b-d	43.98 ab
S_0Ca_1	55.88 a	45.97 a
S_0Ca_2	54.99 a	48.27 a
S_1Ca_0	45.12 c-f	39.70 b-d
S_1Ca_1	51.10 ab	43.68 a-c
S_1Ca_2	46.07 b-e	45.96 a
S_2Ca_0	40.43 ef	34.74 de
S_2Ca_1	50.62 a-c	37.18 d
S_2Ca_2	47.30 b-d	38.19 cd
S_3Ca_0	39.61 f	30.57 e
S_3Ca_1	45.33 c-e	35.44 de
S_3Ca_2	42.86 d-f	36.93 d
LSD $_{(0.05)}$	**5.72**	**5.54**
Significance Level	**NS**	*****
CV (%)	**7.15**	**8.17**

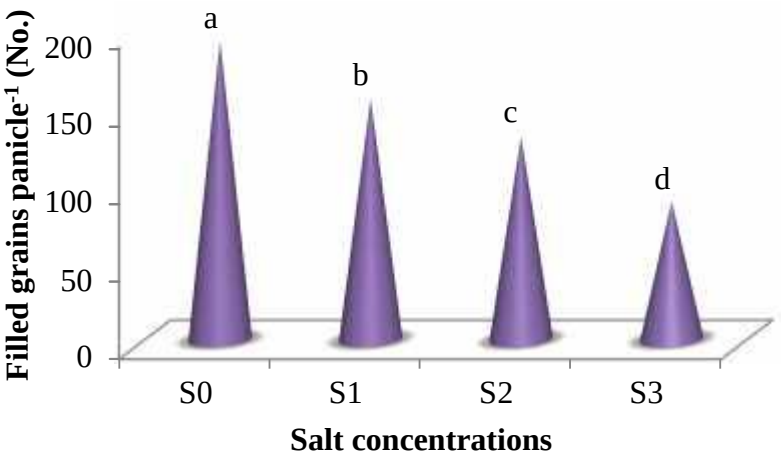
S_0 =0 dSm^{-1}, S_1 = 4 dSm^{-1}, S_2 =6 $dSm^{-1}$and S_3 =8 dSm^{-1}

Ca_0 = 0 ppm, of Ca, Ca_1 = 80 ppm of Ca, Ca_2 = 160 ppm of Ca, *-Significant at 5% level, NS-Non Significant

4.11 Number of filled grains panicle^{-1}

Effects of salinity

It was observed that, as the salinity level increased, the number of filled grain panicle^{-1} decreased significantly (Figure 23). Highest number of filled grain panicle^{-1} was recorded in S_0 (192.3) and lowest number of filled grain panicle^{-1} was found from S_3 (89.00) (Appendix X). The filled grain and grain weight also significantly decrease with the increased salinity level was found by Hasanuzzaman *et al.* (2009).

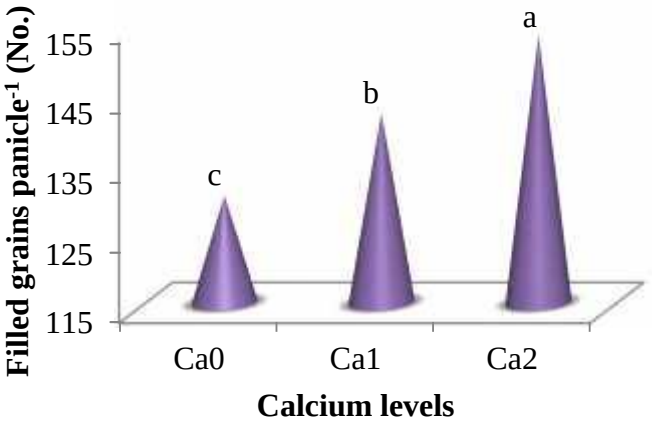


S_0 =0 dSm^{-1}, S_1 = 4 dSm^{-1}, S_2 =6 $dSm^{-1}$and S_3 =8 dSm^{-1}

Figure 23. Effect of different salt concentrations on the filled grains panicle^{-1} of rice (LSD $_{(0.05)}$ =10.90)

Effects of calcium (Ca)

It was recorded that, application of Ca in different amount under different salinity level of BRRI dhan67 showed the number of filled grain panicle^{-1} increased significantly with the increase of calcium level (Figure 24). Highest number of filled grain panicle^{-1} was recorded in Ca_2 (153.1) and lowest number of filled grain panicle^{-1} was found from Ca_0 (130.3) (Appendix X).



Ca_0 = 0 ppm of Ca, Ca_1 = 80 ppm of Ca, Ca_2 = 160 ppm of Ca

Figure 24. Effect of different calcium levels on the filled grains panicle^{-1} of rice (LSD $_{(0.05)}$ =9.44)

Combined effect of salinity and calcium

The effect of application of Ca in different amount under different salinity level of BRRI dhan67 showed the result significantly. Highest number of filled grain panicle^{-1} was recorded in S_0Ca_2 (202.0) and lowest number of filled grain panicle^{-1} was recorded in S_3Ca_0 (80.00). Result showed that the number of filled grain increase with the Calcium increase and decrease with the salinity increase. As it was observed that S_1Ca_1 (155.3), S_1Ca_2 (166.7) gave better result than S_1Ca_0 (143.3), S_2Ca_1 (132.7) S_2Ca_2 (144.3) gave better result than S_2Ca_0 (115.0) and S_3Ca_1 (87.67), S_3Ca_2 (99.33) gave better result than S_3Ca_0 (80.00). This is agreed with Puteh and Mondal (2013).

Table 4.6. Interaction effect of different salt concentrations and calcium levels on the filled grains panicle^{-1}, unfilled grains panicle^{-1}, 1000 grain weigh and grain yield plant^{-1} of rice

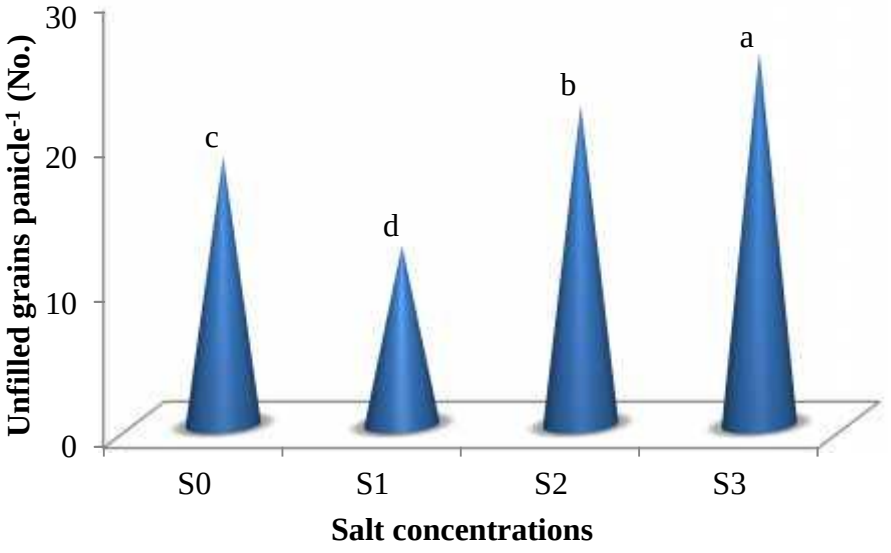
Treatment combinations	Filled grains panicle^{-1} (No.)	Unfilled grains panicle^{-1} (No.)	1000 grain weight (g)	Grain yield plant^{-1} (g)
S_0Ca_0	182.70 bc	20.33 cd	24.55 ab	30.23 bc
S_0Ca_1	192.30 ab	19.67 d	25.78 a	31.92 ab
S_0Ca_2	202.00 a	15.33 ef	26.59 a	34.10 a
S_1Ca_0	143.30 ef	14.67 fg	18.73 de	25.95 de
S_1Ca_1	155.30 de	11.00 h	19.66 cd	28.20 cd
S_1Ca_2	166.70 cd	11.33 gh	21.92 bc	31.63 a-c
S_2Ca_0	115.00 gh	26.00 b	14.31 fg	14.65 g
S_2Ca_1	132.70 fg	21.67 cd	16.99 d-f	19.99 f
S_2Ca_2	144.30 ef	18.33 de	18.62 de	23.90 e
S_3Ca_0	80.00 j	33.67 a	12.45 g	6.453 h
S_3Ca_1	87.67 ij	23.33 bc	15.74 ef	11.53 g
S_3Ca_2	99.33 hi	20.00 cd	16.95 d-f	13.18 g
LSD $_{(0.05)}$	**18.89**	**3.44**	**3.03**	**3.46**
Significance Level	*	*	*	*
CV (%)	**7.87**	**10.37**	**9.25**	**9.02**

S_0 =0 dSm^{-1}, S_1 = 4 dSm^{-1}, S_2 =6 $dSm^{-1}$and S_3 =8 dSm^{-1}, Ca_0 = 0 ppm of Ca, Ca_1 = 80 ppm of Ca, Ca_2 = 160 ppm of Ca, *-Significant at 5% level, NS-Non Significant

4.12 Number of unfilled grain panicle^{-1}

Effects of salinity

It was observed that, as the salinity level increased, the number of unfilled grain panicle^{-1} increased significantly except in S_1 where number of unfilled grain panicle^{-1} decreased (Figure 25). Highest number of unfilled grain panicle^{-1} was recorded in S_3 (25.67) and lowest number of unfilled grain panicle^{-1} was found from S_1 (12.33) (Appendix X).

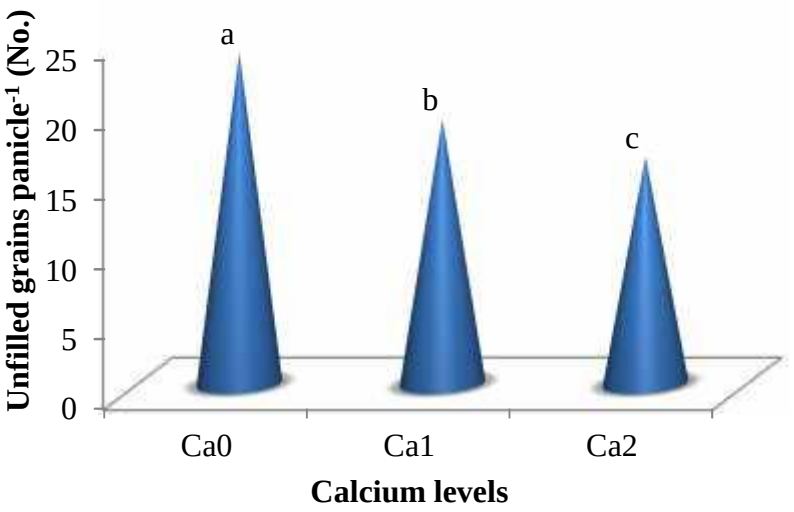


S_0 =0 dSm^{-1}, S_1 = 4 dSm^{-1}, S_2 =6 $dSm^{-1}$and S_3 =8 dSm^{-1}

Figure 25. Effect of different salt concentrations on the unfilled grains panicle^{-1} of rice (LSD $_{(0.05)}$ =1.99)

Effects of Calcium (Ca)

The application of calcium in different doses showed variation in number of unfilled grain panicle^{-1} significantly decreased (Figure 26 and Appendix X). Highest number of unfilled grain panicle^{-1} was recorded in Ca_0 (23.67) and lowest number of unfilled grain panicle^{-1} was found from Ca_2 (16.25).



Ca_0 = 0 ppm of Ca, Ca_1 = 80 ppm of Ca, Ca_2 = 160 ppm of Ca.

Figure 26. Effect of different calcium levels on the unfilled grains panicle^{-1} of rice (LSD $_{(0.05)}$ =1.72)

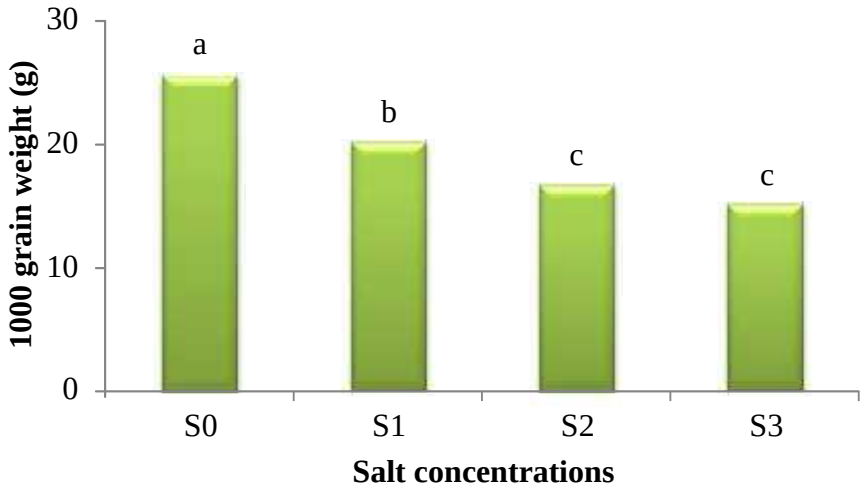
Combined effect of salinity and calcium

The effect of application of Ca in different amount under different salinity level of BRRI dhan67 showed the result significantly (Table 4.6). Highest number of unfilled grain panicle^{-1} was recorded in S_3Ca_0 (33.67) and lowest number of unfilled grain panicle^{-1} was recorded in S_1Ca_1 (11.00) statistically similar with S_1Ca_2 (11.33). The results indicate number of unfilled grains panicle^{-1} increased with the increase of salinity level and calcium application reduces that mostly at 160 ppm concentration, as for consideration we can see in S_3Ca_1 (23.33) and S_3Ca_2 (20.00) gave lower number of unfilled grains panicle^{-1} than S_3Ca_0 (33.67).

4.13 1000- grain weight (g)

Effects of salinity

Weight of 1000- grain of BRRI dhan67 decreased as the level of salinity increased .It is shown significantly. Highest weight of 1000- grain was recorded in S_0 (25.64 g) and lowest weight of 1000- grain was recorded in S_3 (15.05 g) statistically similar with S_2 (16.64 g). Uddin *et al.* (2007) and Hasanuzzaman *et al.* (2009)) stated that salinity reduced the number of 1000-grain weight and yield plant^{-1} of rice.

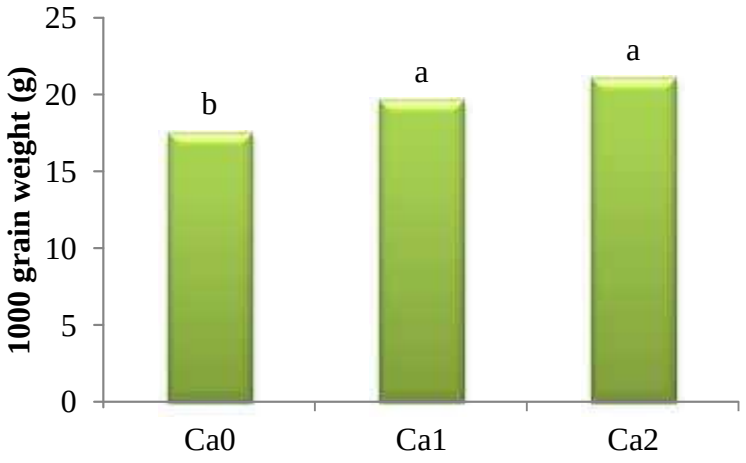


S_0 =0 dSm^{-1}, S_1 = 4 dSm^{-1}, S_2 =6 $dSm^{-1}$and S_3 =8 dSm^{-1}

Figure 27. Effect of different salt concentrations on the 1000 grain weight of rice (LSD $_{(0.05)}$ =1.75)

Effects of Calcium (Ca)

The application of calcium in different amount showed variation in weight of 1000-grain significantly. Highest weight of 1000- grain was recorded in Ca_2 (21.02 g) statistically similar with Ca_1 (19.54 g) and lowest weight of 1000- grains was found from Ca_0 (17.51g).



Ca_0 = 0 ppm of Ca, Ca_1 = 80 ppm of Ca, Ca_2 = 160 ppm of Ca

Figure 28. Effect of different calcium levels on the 1000 grain weight of rice (LSD $_{(0.05)}$ =1.52)

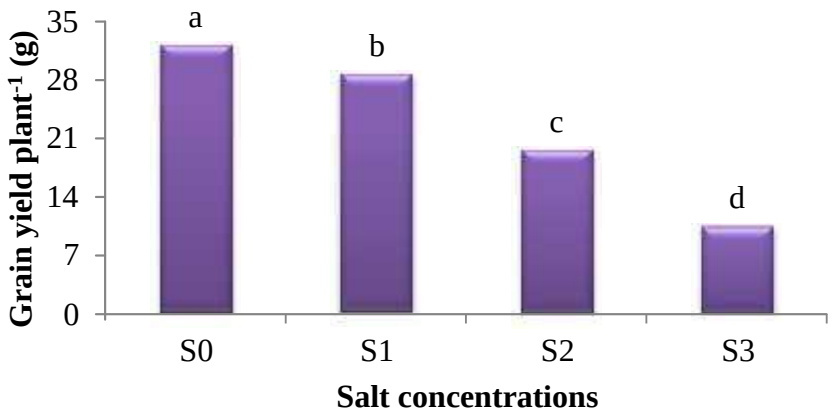
Combined effect of salinity and calcium

The effect of application of Ca in different amount under different salinity level of BRRI dhan67 showed the result significantly (Table 4.6 and Appendix X). . In this experiment, it was observed that S_1Ca_1 (19.66 g), S_1Ca_2 (21.92 g) gave better result and are statistically significant than S_1Ca_0 (18.73 g), S_2Ca_1 (16.99 g) S_2Ca_2 (18.62 g) gave better result than S_2Ca_0 (14.31 g) and S_3Ca_1 (15.74 g), S_3Ca_2 (16.95 g) gave better result than S_3Ca_0 (12.45 g).Highest weight of 1000- grain was recorded in S_0Ca_2 (26.59 g) statistically similar with S_0Ca_0 and S_0Ca_1 (24.55 and 25.78 g respectively) and lowest weight of 1000- grain was found from S_3Ca_0 (12.45 g) statistically similar with S_2Ca_0 (14.31g). This is agreed with Puteh and Mondal (2013).

4.14 Grain yield g plant^{-1}

Effects of salinity

Grain yield (g plant^{-1}**)** of BRRI dhan67 decreased significantly as the level of salinity increased. Highest grain yield ha^{-1} (25.64 g plant^{-1}) was recorded in S_0 which was followed by 0 dSm^{-1} and the lowest grain yield was found in treatment S_3 (15.05 g plant^{-1}) was observed in 8 dSm^{-1}at harvest. (Figure 29 and Appendix X). The loss of grain yield due to 150 mM salinity are 50%, 38%, 44% and 36% over control for the cultivars BR11, BRRI dhan41, BRRI dhan44 and BRRI dhan46, respectively (Hasanuzzaman *et al.*, 2009). He also studied that the all the yield components like grain yield also significantly decrease with the increased salinity level. Under salinity stress, the loss of grain yield results from a combination of reductions in plant stand, spikelet number per panicle, fertility, and harvest index. Among all these contributing components studied, the fertility of grain is found most severely affected and thus causes significant reduction in total yield of grain. In addition to fertility, panicle length and panicle numbers are two important affected characters that contribute to grain yield. The magnitude of salt induced yield losses could not be attributed to a single factor. Different physiological, and biochemical factors at different stages of rice plants might be involved. One factor may be the overall control mechanism (before flowering) of sodium uptake through root properties and its subsequent distribution in different vegetative and floral parts especially in leaves where it causes leaf mortality thereby reducing transportation of total assimilates to the growing region (Munns 2002).

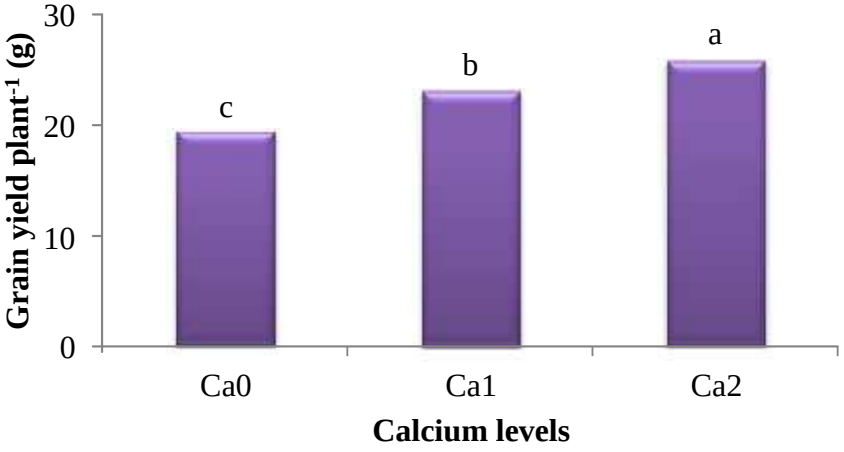


S_0 =0 dSm^{-1}, S_1 = 4 dSm^{-1}, S_2 =6 $dSm^{-1}$and S_3 =8 dSm^{-1}

Figure 29. Effect of different salt concentrations on the grain yield plant^{-1} of rice (LSD $_{(0.05)}$ =2.00)

Effects of Calcium (Ca)

Grain yield (g plant^{-1}**)** of BRRI dhan67 increased significantly on different amount of calcium application. Highest grain yield plant^{-1} (21.02 g plant^{-1}) was recorded in Ca_2 which was followed by 80 ppm Ca and the lowest grain yield was found in treatment Ca_0 (17.51 g plant^{-1}) was observed in 0 ppm Ca at harvest. (Figure 30 and Appendix X). The results indicated that application of fertilizer had a significant effect on grain yield which was agreed with Aslam *et al.* (2001).



Ca_0 = 0 ppm of Ca, Ca_1 = 80 ppm of Ca, Ca_2 = 160 ppm of Ca

Figure 30. Effect of different calcium levels on the grain yield plant^{-1} of rice (LSD $_{(0.05)}$ =1.73)

Combined effect of salinity and calcium

The effect of application of Ca in different amount under different salinity level of BRRI dhan67 showed the result significantly (Table 4.6). Highest weight of yield plant^{-1} was recorded in S_0Ca_2 (34.10 g) statistically similar with S_0Ca_1 and S_1Ca_2 (31.92 and 31.63 g respectively) and lowest weight of yield plant^{-1} was found from S_3Ca_0 (12.45 g) statistically similar with S_2Ca_0 (14.31 g). . In this experiment, it was observed that S_1Ca_1 (28.20 g), S_1Ca_2 (31.63 g) have given better yield and are statistically significant than S_1Ca_0 (25.95 g), S_2Ca_1 (19.99 g) S_2Ca_2 (23.90 g) have given better result than S_2Ca_0 (14.65 g) and S_3Ca_1 (11.53 g), S_3Ca_2 (13.18 g) have given better result than S_3Ca_0 (6.45 g). This is agreed with Puteh and Mondal (2013). The ameliorative effect of Ca was due to reduced shoot Na^+ and Cl^- concentration and better ratio of K^+ to Na^+ in shoot (Aslam *et al.* 2001). This may be due to reduced growth of rice as a result of reduced uptake of water and nutrients and reduction of enzymatic and photosynthetic efficiency and other physiological disorders. This increase might be due to the participation of calcium in mechanism of stomata movement, photosynthesis and osmoregulatory adaptation of plants to water stress in saline soils.

4.14 Leaf, Root, Shoot Dry Weight plant^{-1} (g)

The effect of salinity on dry mass production and distribution was significant (Table 4.7 and Appendix VII). Results showed that leaf weight, root weight and shoot weight decreased with increasing salinity levels. The highest leaf weight, root weight and shoot weight were recorded in control plant (27.98 , 14.15 and 30.01 g respectively) followed by 4 dSm^{-1}. In contrast, the lowest leaf weight, root weight and shoot weight were recorded in 8 dSm^{-1} (16.17, 9.034 and 13.72 g respectively). These results indicate that salinity seriously hampered dry matter accumulation in different plant parts of rice. The reduction in dry matter accumulation in different plant parts under saline condition might be due to lower amount of photosynthetic apparatus (chlorophyll), lower stomatal conductance and lower uptake of nutrients from soil (Mondal *et al.*, 2013). This result agrees with the findings of Moradi and Ismail (2007) in rice. A decrease in chlorophyll concentration of leaves in salinized plants could be attributed to increase activity of the chlorophyll degrading enzyme,

chlorophyllase and the production of the said enzyme enhance by deficiency of K^+ (Reddy and Vora, 1986).

Table 4.7. Effect of different salt concentrations and calcium levels on the leaf dry weight plant^{-1}, root dry weight plant^{-1}, shoots dry weight plant^{-1}and total dry weight plant^{-1} of rice

Treatments	Leaf dry weight plant^{-1} (g)	Root dry weight plant^{-1} (g)	Shoot dry weight plant^{-1} (g)	Total dry weight plant^{-1} (g)
Effect of different salt concentrations				
S_0	27.98 a	14.15 a	30.01 a	72.15 a
S_1	21.96 b	12.74 b	27.12 b	61.83 b
S_2	17.71 c	11.55 c	19.82 c	49.08 c
S_3	16.17 d	9.034 d	13.72 d	38.88 d
LSD $_{(0.05)}$	1.28	1.19	2.67	4.95
Significance Level	*	*	*	*
CV (%)	6.23	10.23	12.04	9.13
Effect of different calcium levels				
Ca_0	17.72 b	10.26 b	18.95 b	46.93 b
Ca_1	22.25 a	12.42 a	23.42 a	58.06 a
Ca_2	22.91 a	12.93 a	25.64 a	61.47 a
LSD $_{(0.05)}$	1.11	1.03	2.31	4.29
Significant Level	*	*	*	*
CV (%)	6.23	10.23	12.04	9.13

S_0 =0 dSm^{-1}, S_1 = 4 dSm^{-1}, S_2 =6 dSm^{-1}and S_3 =8 dSm^{-1}

Ca_0 = 0 ppm of Ca, Ca_1 = 80 ppm of Ca, Ca_2 = 160 ppm of Ca, *-Significant at 5% level, NS-Non Significant

Effects of Calcium (Ca)

Dry weight (g plant^{-1}**)** of BRRI dhan67 increased significantly on different amount of calcium application. The highest leaf weight, root weight and shoot weight were

recorded in Ca_2 (22.91, 12.93 and 25.64 g respectively) which was followed by 80 ppm Ca and the lowest grain yield was found in treatment Ca_0 (17.72, 10.26 and 18.95 g respectively) was observed in 0 ppm Ca at harvest. (Table 4.7).

Combined effect of salinity and calcium

Table 4.8. Interaction effect of different salt concentrations and calcium levels on the leaf dry weight plant^{-1}, root dry weight plant^{-1}, shoots dry weight plant^{-1}and total dry weight plant^{-1} of rice

Treatment combinations	Leaf dry weight plant^{-1} (g)	Root dry weight plant^{-1} (g)	Shoot dry weight plant^{-1} (g)	Total dry weight plant^{-1} (g)
S_0Ca_0	24.21 b	12.32 c-e	26.28 c-e	62.80 cd
S_0Ca_1	28.98 a	14.76 ab	31.05 ab	74.80 ab
S_0Ca_2	30.76 a	15.39 a	32.71 a	78.85 a
S_1Ca_0	17.71 c-e	11.51 d-f	23.29 d-f	52.51 e
S_1Ca_1	25.04 b	13.13 b-d	27.45 b-d	65.62 c
S_1Ca_2	23.14 b	13.59 a-c	30.62 a-c	67.35 bc
S_2Ca_0	15.66 e	9.50 f-h	16.40 gh	41.56 f
S_2Ca_1	17.65 c-e	12.97 b-d	20.73 fg	51.35 e
S_2Ca_2	19.83 c	12.18 c-e	22.33 ef	54.34 de
S_3Ca_0	13.29 f	7.72 h	9.85 i	30.86 g
S_3Ca_1	17.32 de	8.83 gh	14.43 hi	40.58 f
S_3Ca_2	17.90 cd	10.55 e-g	16.89 gh	45.34 ef
LSD $_{(0.05)}$	**2.21**	**2.06**	**4.62**	**9.09**
Significance level	*	**NS**	*	*
CV (%)	**6.23**	**10.23**	**12.04**	**9.67**

S_0 =0 dSm^{-1}, S_1 = 4 dSm^{-1}, S_2 =6 dSm^{-1}and S_3 =8 dSm^{-1}

Ca_0 = 0 ppm Ca , Ca_1 = 80 ppm of Ca, Ca_2 = 160 ppm of Ca, *-Significant at 5% level, NS-Non Significant

In interaction effect of salinity and calcium on leaves, roots and shoot dry weight exhibited a significant effect (Table 4.8 and Appendix VII). The highest leaves dry weight (30.76 g), root dry weight (15.39 g) and shoot dry weight (32.71 g) was found in S_0Ca_2 where the lowest leaves dry weight (13.29 g), root dry weight (7.723 g) and shoot dry weight (9.85 g) was found in S_3Ca_0. Manivannan *et al.* (2007) reported that, calcium had the ameliorative effect on salt stress and increased the total dry weight of plant through increasing the vegetative growth of plant. In this experiment, this trend was also found as it was observed that S_1Ca_1 (25.04 g), S_1Ca_2 (23.14 g) have given better result than S_1Ca_0 (17.71 g), S_2Ca_1 (17.65 g), S_2Ca_2 (19.83 g) have given better result than S_2Ca_0 (15.66 g) and S_3Ca_1 (17.32 g), S_3Ca_2 (17.90 g) have given better result than S_3Ca_0 (13.29 g) in case of Leaf dry weight plant^{-1}. Same result was found in case of Root dry weight plant^{-1} (g) and Shoot dry weight plant^{-1} (g). In most of the cases mitigating effect of calcium was found significant.

CHAPTER 5

SUMMARY AND CONCLUSION

The experiment was conducted in the net house of the Department of Agricultural Botany and in the Laboratory of Agricultural Botany of Sher-e-Bangla Agricultural University (SAU), Dhaka, during the period of December-June 2017-2018 to evaluate the response of BRRI dhan67 to calcium supplementation at different salinity levels. In this experiment, the treatments consisted of four different salinity levels viz. S_0= without salt (0 dSm^{-1}), S_1= 4 dSm^{-1}, S_2 = 6 dSm^{-1}, S3= 8 dSm^{-1}, and three different levels of calcium viz. Ca_0= 0 ppm, Ca_1= 80 ppm and Ca_2 = 160 ppm. The experiment was laid out in two factors Completely Randomized Design (CRD) with three replications. Data on different growth parameters, physiological parameters and yield with yield contributing characters of rice were recorded. The collected data were statistically analyzed for evaluation of the treatment effect. A significant variation among the treatments was found while different salinity levels and calcium levels were applied in different combinations.

There are significant differences among the influence of different levels of salinity in case of almost all the parameters. In this experiment, rice plants were subjected to salinity by applying saline water throughout the life cycle of rice plant after transplanting to keep the soil in saline condition. Plant grown on normal soil (control treatment) showed the maximum height more or less over the growth period whereas the lowest height was recorded from 8 dSm^{-1} treated plants. At 30, 60, 90 DAT and at harvest, the highest plant height were 76.49, 103.7, 124.7 and 122.7 cm respectively under a controlled condition whereas the lowest height were 57.92, 73.23, 93.41and 91.97 cm at 8 dSm^{-1}. The maximum number of tillers $plant^{-1}$ were 14.56, 19.78 and 18.44 at 30, 60 and 90 DAT from S_0 (control) whereas the lowest were 7.44, 9.56 and 9.11with 6, 8 and 8 dSm^{-1} respectively. Maximum number of effective tillers $plant^{-1}$ was 16.44 under controlled treatment, whereas the lowest was 6.56 with 8 dSm^{-1} of salinity. Maximum number of ineffective tillers $plant^{-1}$ was 2.76 under 8 dSm^{-1} level of salt stress, whereas the lowest was 1.88 with 6 dSm^{-1}.The maximum days required for flowering for the first time was recorded from 8 dSm^{-1} of salt stress (65.67 DAT) and the lowest was recorded under control (55.56 DAT). Highest Panicle length was

recorded 27.52 cm under controlled treatment whereas the lowest was recorded 19.35 cm from 8 dSm^{-1}. The maximum leaf area $plant^{-1}$ (81.48 cm^2) was observed from controlled treatment whereas the lowest (52.78 cm^2) was found from 8 dSm^{-1}. The highest leaf relative water content (90.86) was recorded from controlled plant whereas the lowest (66.46) was recorded from 8 dSm^{-1} of salt stress. The highest leaf membrane stability (85.67) was recorded from control treatment whereas the lowest (36.07) was recorded from 8 dSm^{-1} of salt stress. The leaf chlorophyll content was degraded with the increase of salinity whereas the maximum chlorophyll content was recorded from no or low levels of salt with minimum from 8 dSm^{-1}.The highest leaf, root and shoot dry weight (27.98, 14.15 and 30.01 gm) was recorded from control treatment whereas the lowest (16.17, 9.03 and 13.72 gm) was recorded with 8 dSm^{-1} of salinity. The maximum number of filled grain $panicle^{-1}$(192.3), 1000-grain weight (25.64 gm), yield $plant^{-1}$(32.08 gm) was recorded from control and the lowest value of all these parameters were found from 8 dSm^{-1} of salt stress. 8 dSm^{-1} salinity level was responsible for maximum number of unfilled filled grain $panicle^{-1}$ (25.67) whereas the lowest (12.33) was recorded from 4 dSm^{-1}.

Calcium significantly influenced maximum parameters selected for data collection. At 30, 60, 90 DAT and at harvest the highest plant height (71.63, 93.95, 113.6 and 112.6 cm) was obtained from Ca_2 (160 ppm Ca^{2+}), Ca_1 (80 ppm Ca^{2+}), Ca_2 and Ca_2 respectively over the control plants whereas the lowest height was 62.60, 84.11, 102.6 and 101.5 cm under Ca_0. The maximum number of tillers $plant^{-1}$ was 11.25, 15.50 and 14.58 at 30, 60 and 90 DAT from Ca_1, Ca_2 and Ca_2 respectively whereas the lowest was 8.17, 12.17 and 11.08 from Ca_0. Then maximum number of effective tillers $plant^{-1}$ was 12.92 under Ca_2 treatment, whereas the lowest was 9.50 (Ca_0). Maximum number of ineffective tillers $plant^{-1}$ was 2.65 under Ca_0 treatment, whereas the lowest was 1.74 from Ca_2 .The maximum days required for flowering for the first time was recorded from Ca_0 (63.42 DAT) and the lowest was recorded under Ca_2 (60.67 DAT). Highest Panicle length was recorded 24.44 cm under Ca_2 treatment whereas the lowest was recorded 21.67 cm from Ca_0. The maximum leaf area $plant^{-1}$ (68.31cm^2) was observed from Ca_2 treatment whereas the lowest (58.49 cm^2) was found from Ca_0. The leaf chlorophyll content was increased with the increase of calcium dose whereas the maximum chlorophyll content was recorded from 160 ppm Ca with minimum from control. The highest leaf, root and shoot dry weight (22.91, 12.93 and 25.64 gm) was recorded from Ca_2 whereas the lowest (17.72, 10.26 and 18.95 gm)

was showed by Ca_0. The maximum number of filled grain panicle^{-1}(153.1), 1000-grain weight (21.02 gm), yield plant^{-1}(25.70 gm) was recorded from Ca_2 and the lowest value of all these parameters were observed under Ca_0. Ca_0 calcium dose was responsible for maximum number of unfilled filled grain panicle^{-1} (23.67) whereas the lowest (16.25) was recorded from Ca_2.

The combinations of salinity and calcium significantly influenced almost all the parameters. At 30, 60, 90 DAT and at harvest the highest plant height (80.07, 106.4, 128.0 and 126.6 cm) was obtained from S_0Ca_2 over the control plants whereas the lowest height was 53.68, 63.37, 82.11 and 82.00 cm under S_3Ca_0. Mitigating effect of calcium were found most significantly at 90 DAT where it was observed that S_3Ca_1 (97.77 cm) and S_3Ca_2 (100.3 cm) showed better results than that of S_3Ca_0 (82.00 cm). The maximum number of tillers plant^{-1} was 17.67, 21.67 and 20.00 at 30, 60 and 90 DAT with S_0Ca_1, S_0Ca_2 and S_0Ca_2 respectively whereas the lowest was 6.00, 7.67 and 6.67with S_2Ca_2, S_3Ca_0 and S_3Ca_0 respectively. At 90 DAT, it was observed that S_3Ca_1 (9.67) and S_3Ca_2 (11.00) performed better than that of S_3Ca_0 (6.67). Similar trend was observed in each case where calcium was supplemented with salt. Then maximum number of effective tillers plant^{-1} was 19.00 under S_0Ca_2 treatment, whereas the lowest was (4.67) S_3Ca_0. It was observed that S_1Ca_1 (11.00) and S_1Ca_2 (13.00) produced higher number of effective tillers plant^{-1} than S_1Ca_0 (10.67). The treatments S_2Ca_1 (10.67) and S_2Ca_2 (11.67) helped to increase the number of effective tillers plant^{-1} compared to S_2Ca_0 (8.67). The treatments S_3Ca_1 (7.00) and S_3Ca_2 (8.00) produced higher number of effective tillers plant^{-1} than that of S_3Ca_0 (4.67). Maximum number of ineffective tillers plant^{-1} was 3.27 under S_3Ca_0 treatment, whereas the lowest was 1.33 recorded from S_0Ca_2. The treatments S_1Ca_1 (2.00) and S_1Ca_2 (1.67) produced lower number of ineffective tillers plant^{-1} compared to S_1Ca_0 (2.33). The treatments S_2Ca_1 (1.67) and S_2Ca_2 (1.63) showed lower number of ineffective tillers plant^{-1} than that S_2Ca_0 (2.33), S_3Ca_1 (2.67) and S_3Ca_2 (2.33) gave lower number of ineffective tillers plant^{-1} than that of S_3Ca_0 (3.27). The maximum days required for flowering for the first time was recorded from S_3Ca_1 (66.67 DAT) and the lowest was recorded under S_0Ca_2 (51.33 DAT). It was observed that days required for flowering was reduced by calcium supplementation mostly at higher level of salinity (8 dSm^{-1}) as we can see plants under the treatment S_3Ca_2 (64.33 DAT) required shorter duration of time to flower than that of S_3Ca_0 (66.00 DAT). The

highest panicle length was recorded 29.03 cm under S_0Ca_2 treatment whereas the lowest was recorded 17.95 cm from S_3Ca_0 which was statistically similar to S_2Ca_0, S_3Ca_1 and S_3Ca_2 (19.32, 19.67 and 20.43 cm respectively). The leaf area was found higher with 160 ppm concentration of calcium in case of 0, 4, 6 and 8 dSm^{-1} of salt (84.66, 69.41, 61.23 and 57.95 cm^2 respectively) treated plants. In case of leaf relative water content, it was observed that S_1Ca_1 (87.06) and S_1Ca_2 (90.14) performed better than S_1Ca_0 (82.89). The treatments S_2Ca_1 (78.41) and S_2Ca_2 (81.69) facilitated better results than that of S_2Ca_0 (72.41). The treatments S_3Ca_1 (67.90) and S_3Ca_2 (71.35) showed better results compared to S_3Ca_0 (60.13). Similar results were found in case of leaf membrane stability. The maximum leaf chlorophyll content was recorded from S_0Ca_1 at 60 DAT and S_0Ca_2 at 90 DAT (55.88 and 48.27 SPAD units respectively) whereas the lowest were recorded from S_3Ca_0 at both 60 and 90 DAT (39.61 and 30.57 SPAD units respectively). In each case where calcium was supplemented (S_1Ca_1, S_1Ca_2, S_2Ca_1, S_2Ca_2, S_3Ca_1 and S_3Ca_2) with salt, mitigating effect was observed. The highest leaf, root and shoot dry weight (30.76, 15.39 and 32.71gm) was recorded from S_0Ca_2 whereas the lowest (13.29, 7.72 and 9.85 gm) was recorded from S_3Ca_0. Calcium facilitated mitigation of salt stress in each case where calcium was supplemented (S_1Ca_1, S_1Ca_2, S_2Ca_1, S_2Ca_2, S_3Ca_1 and S_3Ca_2) with salt. In case of filled grain $panicle^{-1}$ it was observed that S_1Ca_1 (155.3) and S_1Ca_2 (166.7) performed better than that of S_1Ca_0 (143.3). The treatments S_2Ca_1 (132.7) and S_2Ca_2 (144.3) showed better results compared to S_2Ca_0 (115.0). The treatments S_3Ca_1 (87.67) and S_3Ca_2 (99.33) provided better results than that of S_3Ca_0 (80.00). More or less similar trend was observed in case of 1000-grain weight. Plant yield was found significantly affected by salinity and the stress condition was observed to be mitigated by supplemental calcium, as S_1Ca_1 (28.20 g) and S_1Ca_2 (31.63 g) were statistically significant and found to perform than that of S_1Ca_0 (25.95 g). Similarly S_2Ca_1 (19.99 g) and S_2Ca_2 (23.90 g) provided better yield compared to S_2Ca_0 (14.65 g). The treatments S_3Ca_1 (11.53 g) and S_3Ca_2 (13.18 g) produced better results than that of S_3Ca_0 (6.45 g). The maximum number of filled grain $panicle^{-1}$(202.0), 1000-grain weight (26.59 gm), yield $plant^{-1}$(34.10 gm) was recorded from S_0Ca_2 and the lowest value of all these parameters were found from S_3Ca_0 (80.00, 12.45 gm and 6.45 gm respectively). S_3Ca_0 calcium dose was responsible for maximum number of unfilled filled grain $panicle^{-1}$ (33.67) whereas the lowest (11.00) was recorded from S_3Ca_1. In most of the parameter 160 ppm of calcium application was found to have better

mitigating potential than 80 ppm of calcium even at higher level of salt stress (8 dSm^{-1}).

Conclusion

Considering the above mentioned results, it may be concluded that, the yield of BRRI dhan67 was gradually decreased by the increase of salinity levels and this reduction rate was decreased by exogenous supply of calcium. Among the calcium levels, 160 ppm showed the highest results in case of growth, physiology and yield parameters as compared to 80 ppm. The best results were mostly found at sole 160 ppm Ca treatment which indicates that calcium has important roles in different physiological and metabolic processes of plants. When plants are subjected to salt stress then calcium played a crucial role to ameliorate stress condition.

Scope of Future Research

Further studies are needed to find out:

- More accurate dosage of Calcium or effective application method
- Actual cellular or metabolical mechanism of Calcium in ameliorating salt stress
- Other methods or chemicals to mitigate salt stress
- The grain protein content and other quality attributes of rice under salt stress and also when supplemented with calcium. Such studies should be carried out to different saline prone areas of the country.

References

Abdel Latef, A. A. (2011). Ameliorative effect of calcium chloride on growth, antioxidant enzymes, protein patterns and some metabolic activities of canola (*Brassica napus* L.) under seawater stress. *J. Plant Nutr.* **34**: 1303-1320.

Ahanger, M. A., Agarwal, R. M. (2017). Salinity stress induced alterations in antioxidant metabolism and nitrogen assimilation in wheat (*Triticum aestivum* L) as influenced by potassium supplementation. *Plant Physiol Biochem.* **115**:449–460

Ahmad, P., Hashem, A., AbdAllah, E.F., Alqarawi, A.A., John, R, Egamberdieva, D., Gucel, S. (2015). Role of Trichoderma harzianum in mitigating NaCl stress in Indian mustard (*Brassica juncea* L) through antioxidative defense system. *Front Plant* Sci. **6**:868.

Ahmad, P., Sarwat, M., Bhat, N. A., Wani, M. R., Kazi, A. G. and Tran L. P. (2015). Alleviation of cadmium toxicity in *Brassica juncea* L. (Czern. & Coss.) by calcium application involves various physiological and biochemical strategies. *PLoS ONE* **10**:e0114571 10.1371/journal.pone.0114571

Ahmed, I. M., Nadira, U. A., Bibi, N., Zhang, G. and Wu, F. (2015). Tolerance to combined stress of drought and salinity in barley. In combined stresses in plants physiological, molecular, and biochemical aspects R. Mahalingam, (ed.). Springer, Basel, Switzerland. pp. 93–121.

Ahmed, I. M., Cao, F., Zhang, M., Chen, X., Zhang, G. and Wu, F. (2013). Difference in yield and physiological features in response to drought and salinity combined stress during anthesis in Tibetan wild and cultivated barleys. *PloS ONE*. **8**(10): e77869.

Akram, M. S. and Ashraf, M. (2011). Exogenous application of potassium dihydrogen phosphate can alleviate the adverse effects of salt stress on sunflower (*Helianthus annuus* L.). *J. Plant Nutr*. **34**: 1041–1057.

Alim, A. (1982). Bangladesh Rice. Alim Production, 18, Garden Road, Dhaka.

Amirjani, M. R. (2011). Effect of salinity stress on growth, sugar content, pigments and enzyme activity of rice. *Int. J. Bot.* **7**(1):73–81.

Anbumalarmathi, A. and Mehta, P. (2013). Effect of Salt Stress on Germination of *indica* Rice Varieties. *Eur. J. of Biol. Sci*.**6**(1):1-6.

Apel, K. and Hirt, H. (2004). Reactive oxygen species: metabolism, oxidative stress, and signal transduction. *Ann. Rev. Plant Biol.* **55**: 373–99.

Aslam, M., Mahmood, I. H., Qureshi, R. H., Nawaz, S., Akhter, J. and Ahmad, Z. (2001). Nutritional Role of Calcium in Improving Rice Growth and Yield under Adverse Conditions. *Int. J. Agric. Biol*. :1560–8530/2001/03–3–292–297

Aslam, M., Muhammad N., Qureshi R.H., Akhtar J. and Ahmed Z. (2000). Role of Ca^{2+} in salinity tolerance of rice. Symp. On Integ. Plant Manage. No. 8-10 (1998), Islamabad.

Bhattacharjee S. (2008). Triadimefon pretreatment protects newly assembled membrane system and causes up-regulation of stress proteins in salinity stressed *Amaranthus lividus* L. during early germination. *J. Environ. Biol*. **29**: 805-810.

Bhusan, D., Das, D. K., Hossain, M., Murata, Y. and Hoque, M. A. (2016). Improvement of salt tolerance in rice (*Oryza sativa* L.) by increasing antioxidant defense systems using exogenous application of proline. *Aust. J. Crop Sci*. **10**(1): 50–56.

Brar D. S. and Khush G. S. (2003). Utilization of wild species of genus *Oryza* in rice improvement In: JS Nanda, and SD Sharma (Eds) Monographon Genus *Oryza* pp: 283-309.

Cha-um, S., Singh, H. P., Samphumphuangm, T. and Kirdmanee, C. (2012). Calcium-alleviated salt tolerance in indica rice (*Oryza sativa* L. spp. *indica*): Physiological and morphological changes. *AJCS* **6**(1):176-182.

Chakraborty, K., Sairam, R.K. and Bhaduri, D. (2015). Effects of different levels of soil salinity on yield attributes, accumulation of nitrogen, and micronutrients in *Brassica* Spp. *J. Plant Nutr*. doi:10.1080/01904167.2015.1109105

Choudhury, S., Panda, P., Sahoo, L. and Panda, S. K. (2013). Reactive oxygen species signaling in plants under abiotic stress. *Plant Signal. Behav*. **8**(4): 236-281.

Chunthaburee, S., Dongsansuk, A., Sanitchon, J., Pattanagul, W. and Theerakulpisut, P. (2016). Physiological and biochemical parameters for evaluation and clustering of rice cultivars differing in salt tolerance at seedling stage. *Saudi J. Biol. Sci.* **23**(4): 467–477.

Colmer, T. D., Teresa W. M. F., Richard M. H. and Andre L. (1994). Interactions of Ca^{2+} and NaCl stress on the ion relations and intercellular pH of sorghum bicolor root tips: An in vivo 31P. NMR study. *J. Expt. Bot*., **45**: 1037–44.

Curtiss, J., Rodriguez-Uribe, L., Stewart, J. M., Zhang, J. (2011). Identification of differentially expressed genes associated with semigamy in pima cotton (*Gossypium barbadense* L.) through comparative microarray analysis. *BMC Plant Biol*. 11: 49.

DAE (2015). http://www.bbs.gov.bd/annual report.

Demidchik V. and Tester M. (2002). Sodium fluxes through nonselective cation channels in the plasma membrane of protoplasts from Arabidopsis roots. *Plant Physiol.* **128**:379–387.

Dionisio-Sese, M. L. and Tobita, S. (1998). Antioxidant responses of rice seedlings to salinity stress. *Plant Sci*.:135,1-9.

Ebrahimi, R. F., Rahdari, P., Vahed, H. S., Shahinrokhsar, P. and S. Babazadeh. (2012). Rice Response to Different Methods of Potassium Application under Salinity Stress Condition. *Am-Euras. J. Agric. & Environ. Sci*.**12**(11): 1441-1445.

El-Shabrawi H., Kumar, B, Kaul, T., Reddy, M.K., Singla-Pareek, S.L. and Sopory, S.K. (2010). Redox homeostasis, antioxidant defense, and methyl glyoxal detoxification as markers for salt tolerance in Pokkali rice. *Protoplasma.* **245**:85–96.

FAO (2007). Water Profile of Bangladesh. (www.fao.org/nr/water/aquastat/countries_regions/bgd/index.stm)

FAO (2014). http://www.fao.org/faostat/en/#data/QC

FAO (2015). www.fao.org/docrep/003/x6905e/x6905e04.htm.

Farooq, M., Wahid, A., Kobayashi, N., Fujita, D. and Basra, S. M. A. (2009). Plant drought stress: effects, mechanisms and management: a review. *Agron. Sustain. Dev*. **29**: 185-212.

Fomicheva, A. S., Tuzhikov, A. I., Beloshistov, R. E., Trusova, S. V., Galiullina, R. A. and Mochalova, L.V. (2012). Chichkova, N.V.; Vartapetian, A.B. Programmed cell death in plants. *Biochem*. **77**: 1452–1464.

Flowers T. J., Colmer T. D.(2008). Salinity tolerance in halophytes. *New Phytol*. **179**:945–963.

Ghosh, S., Mitra, S. and Paul, A. (2015). Physiochemical studies of sodium chloride on mungbean (*Vigna radiata* L. Wilczek) and its possible recovery with spermine and gibberellic acid. *Sci. World J*. 2015: 8. doi:10.1155/2015/858016

Gilroy S., Suzuki N. and Miller G.(2014). A tidal wave of signals: calcium and ROS at the forefront of rapid systemic signaling. Trends Plant Sci. **19**: 623–630.

Girija, C., Smit B. N. and Swamy, P. (2002). Interactive effects of sodium chloride and calcium chloride on the accumulation of proline and glycine- betaine in peanut (*Arachis hypogaea* L.). *Environ. Exp. Bot*. **47**: 1-10.

Gopalan C., Rama Sastri B. V. and Balasubramanian S. (2007). Nutritive Value of Indian Foods, published by National Institute of Nutrition (NIN), ICMR (Indian Council of Medical Research).

Govindaraju, P. and Balakrishanan, K. (2002). Effect of salinity in certain exzyme activity, physiological traits and yield of rice cultivars. *Madras Agric. J.* **89**(1-3): 67-69.

Greenberg, J.T.(1996). Programmed cell death: A way of life for plants. *Proc. Natl. Acad. Sci. USA*: **93**: 12094–12097.

Gulzar, S., Khan, M. A. and Ungar, I. A. (2003). Salt tolerance of a coastal salt marsh grass. Commu. *Soil Sci. Plant Anal*. **34**: 2595–605.

Gul, B. and Khan, A. (2008). Role of calcium in alleviating salinity effects in coastal halophytes, In: Khan MA, Weber DJ (Eds.). Ecophysiology of high salinity tolerant plants. *Springer*, Dordrecht. p. 107-114.

Haggag, W.M., Abouziena, H.F., Abd-El-Kreem, F. and El Habbasha, S. (2015). Agriculture biotechnology for management of multiple biotic and abiotic environmental stress in crops. J. *Chem. Pharm. Res*. **7**(10): 882–889.

Hakim, M. A., Juraimi, A. S. and Hanafi, M. M. (2014). The effect of salinity on growth, ion accumulation anr yield of rice varieties. *J. Anim. Plant Sci*. **24**: 874–885.

Harris, B. N., Sadras, V. O. and Tester, M. (2010). A water-centred framework to assess the effects of salinity on the growth and yield of wheat and barley. *Plant Soil*. **336**: 377–89.

Hasanuzzaman, M., Alam, M. M., Rahman, A., Hasanuzzaman, M., Nahar, K. and Fujita, M. (2014). Exogenous proline and glycine betaine mediated upregulation of antioxidant defense and glyoxalase systems provides better protection against salt-induced oxidative stress in two rice (*Oryza sativa* L.) varieties. *Biom. Res. Int.* 2014: 757219.

Hasanuzzaman, M., Hossain, M. A., da Silva, J. A. T. and Fujita, M. (2012a). Plant responses and tolerance to abiotic oxidative stress: antioxidant defenses is a key factors. In: Crop stress and its management: perspectives and strategies.V. Bandi, A.K. Shanker, C. Shanker, M. Mandapaka,(ed.). *Springer*, Berlin. pp. 261–316.

Hasanuzzaman M., Nahar K., and Fujita M. (2013b). Plant response to salt stress and role of exogenous protectants to mitigate salt-induced damages, in Ecophysiology and Responses of Plants Under Salt Stress, eds P. Ahmed, M. M. Azooz, and M. N. V. Prasad (New York, NY: *Springer*), 25–87. doi: 10.1007/978-1-4614-4747-4_2

Hasanuzzaman, M., Nahar, K., Fujita, M., Ahmad, P., Chandna, R., Prasad, M.N.V. and Öztürk, M. (2013a). Enhancing plant productivity under salt stress—

relevance of poly-omics. In: Salt stress in plants: Omics, signaling and responses. P. Ahmad, M. M. Azooz, and M.N.V Prasad, (ed.). *Springer*, Berlin, Germany. pp. 113–156.

Hasanuzzaman, M., Fujita, M., Islamm, M.N., Ahamed, K.U and Kamrun Nahar. (2009). Performance of four irrigated rice varieties under different levels of salinity stress. *Int. J. Integr. Biol.* **6**(2): 85-89.

Heenan, D. P., Lewin, L. G.and McCaffery, D. W. (1988). Salinity tolerance in rice varieties at different growth stages. *Aust. J. Exp. Agric*. **28**: 343–349.

Hengartner, M. O. (2000). The biochemistry of apoptosis. *Nature*. **407**: 770–776.

Hernandez, J., Aguilar, A. B., Portillo, B., Lopez-Gomez, E., Beneyto, J. and Legaz, M. (2003). The effect of calcium on the antioxidant enzymes from salt-treated loquat and anger plants. *Funct. Plant Biol*. **30**: 1127–1137

Hoang, T., Tran, T., Nguyen, T., Williams, B., Wurm, P., Bellairs, S., Mundree, S. (2016). Improvement of salinity stress tolerance in rice: challenges and opportunities. *Agronomy*. **6**: 54.

Howladar and Rady (2012). Effects of calcium paste as a seed coat on growth, yield and enzymatic activities in NaCl stressed-pea plants. *African J. Biotech*. **11**(77): 14140-14145.

Hussain, S.G. (2012). Role of Rice in Food Security of Bangladesh (Chapter 5). In: Food Security: Quality Management, Issues and Economic Implications.M.A. Jones and F.E. Hernandez, (ed.). Nova Science Publishers. pp. 127–148.

Husain, S., Von Caemmerer, S. and Munns, R. (2004). Control of salt transport from roots to shoots of wheat in saline soil. *Func. Plant Biol*. **31**: 1115-1126.

Hussain, B. M. N., Akram, S., Raffi, S. A., Burritt, D. J. and Hossain, M. A. (2016). Exogenous glutathione improves salinity stress tolerance in rice (*Oryza sativa* L.), *Plant Gene and Trait*. **7**(11): 1–17.

Ishak, N. K., Sulaiman, Z. and Tennakoon, K. U. (2015). Comparative study on growth performance of transgenic (over-expressed OsNHX1) and wild-type Nipponbare under different salinity Regimes. *Rice Sci.* **22**(6): 275–282.

Jafari, M. H. S., Kafi, M. and Astarie, A. (2009). Interactive effects of NaCl induced salinity, Calcium and potassium on physiomorphological traits of sorghum (*Sorghum bicolor* l.). *Pakistan J. Bot*. **41**(6): 3053-3063.

Jaleel C. A. and Azooz M. M. (2009). Exogenous calcium alters pigment composition, γ-glutamyl kinase and proline oxidase activities in salt stressed Withania somnifera. *Plant Omics.* J. **2**:85–90.

Jaleel C. A., Manivannan P., Kishorekumar A., Sankar, B. and Panneerselvam R. (2007a). Calcium chloride effects on salinity induced oxidative stress, proline metabolism and indole alkaloid accumulation in Catharanthus roseu. *Compt. Rend. Biol.* **330**: 674–683.

Jaleel, C. A., Manivannan, P., Sankar, B., Kishorekumar, A., Gopi, R.,Somasundaram, R. and Panneerselvam, R. (2007b). Water deficit stress mitigation by calcium chloride in Catharanthus roseus : Effects on oxidative stress, proline metabolism and indole alkaloid accumulation, Colloids and Surfaces B: *Biointerfaces* **60**: 110–116

Kazemi, K. and Eskandari, H. (2011). Effects of salt stress on germination and early seedlinggrowth of rice (*Oryza sativa*) cultivars in Iran. *Afr. J. Biotechnol.* **10**(77): 17789-17792.

Kellogg, E.A., Camara, P.E.A.S., Rudall, P.J., Ladd, P., Malcomber, S.T., Whipple, C.J. and Doust, A.N. (2013). Early inflorescence development in the grasses (Poaceae). *Front. Plant Sci*. **4**: 250.

Keunen , E., Peshev, D., Vangronsveld, J., Van den Ende, W. and Cuypers, A. (2013). Plant sugars are crucial players in the oxidative challenge during abiotic stress. Extending the traditional concept. *Plant Cell Environ*. **36**(7):1242-1255

Khan. (2013). Amelioration of salt stress in wheat (*Triticum aestivum* L.) by foliar application of nitrogen and potassium. *Sci. Tech. and Dev.* **32**(2): 85-98.

Khan, M. A., Shirazi, M. U., Khan, M. A., Mujtaba, S. A. I., Islam, E., Mumtaz, S., Shereen, A., Ansari, R. and Ashraf, M. Y. (2009). Role of proline, K/Na ratio and chlorophyll content in salt tolerance of wheat (*Triticum aestivum* L.). *Pak. J. Bot*. **41**: 633–638.

Khush, G. S. (2005).What will it take to feed 5.0 billion rice consumers in 2030. *Plant MolBiol* **59**: 1-6.

Kibria, M. G., Hossain, M., Murata, Y., and Hoque, M. A. (2017). Antioxidant defense mechanisms of salinity tolerance in rice genotypes. *Rice Sci*. **24**(3): 155–162.

Koyama, M.L., Levesley, A., Koebner, R.M.D., Flowers, T.J. and Yeo, A.R. (2001). Quantitative trait loci for component physiological traits determining salt tolerance in rice. *Plant Physiol*. **125**:406–422.

Kumar, V.; Shriram, V.; Nikam, T.D.; Jawali, N.; Shitole, M.G. (2008). Sodium chloride-induced changes in mineral nutrients and proline accumulation in indica rice cultivars differing in salt tolerance. *J. Plant Nutr*., **31**:1999–2017.

Lauchli, A. (1990). Calcium, salinity and the plasma membrane. *In:* R.T. Leonard, P.K. Hepler (Eds.), *Calcium in plant growth and development*. American Society of Plant Physiologist, Rockville, M.D: pp. 26–35.

Lawlor, D. W. (2002). Limitation to photosynthesis in water stressed leaves: Stomata vs. metabolism and the role of ATP. *Ann. Bot*. **89**: 1–15

Li, J. Y., Jiang, A. L. and Zhang, W. (2007). Salt stress-induced programmed cell death in rice root tip cells. *J. Integr. Plant Biol*. **49**: 481–486.

Lisenbee, C. S., Lingard, M. J. and Trelease, R. N. (2005). Arabidopsis peroxisomes possess functionally redundant membrane and matrix isoforms of monodehydroascorbate reductase. *Plant J.* **43**: 900–914.

Liu, S. H., Fu, B. Y., Xu, H. X., Zhu, L. H., Zhai, H. Q. and Li, Z. K. (2007). Cell death in response to osmotic and salt stresses in two rice (Oryza sativa L.) ecotypes. *Plant Sci*. **172**: 897–902.

Maas, E.V. and Hoffman, G.J. (1977). Crop salt tolerance: Current assessment. J. Irrig. Drain. Div. Amer. Soc. Civ. Eng. **103**: 115-132.

Maathuis, F. J. (2006). The role of monovalent cation transporters in plant responses to salinity. *J. Exp. Bot*. **57**: 1137–47.

Manivannan, P., Jaleel, C. A., Sankar, B., Somasundaram, R., Murali, P. V., Sridharan, R. and Panneerselvam, R. (2007). Salt stress mitigation by calcium chloride in *Vigna radiata* L. *Wilczek. Acta. Biol. Cracov*. **49**: 105-109

Marschner, H. (1995). Mineral nutrition of higher plants, 2nd ed. Academic Press, London, 889 p.

Martinez, V. and Lauchli A. (1993). Effect of Ca^{2+} on the salt stress response of barley roots as observed by in vivo 31P-nuclear magnetic resonance and in vitro analysis. *Planta.*, **1909**: 519–24.

Matysik, J., Alia Bhalu, B. and Mohanty, P. (2002). Molecular mechanisms of quenching of reactive oxygen species by proline under stress in plants. *Curr. Sci. India.* 82: 525–32.

Maxwell, K. and Johnson, G. N. (2000). Chlorophyll fluorescence—a practical guide. *J. Expt. Bot*. **51**(345):659–668.

Mendoza, I., Rubio, F., Rodriguez-Navarro, A. and Pardo, J. M. (1994). The protein phosphatase 6calcineurin is essential for NaCl tolerance of *Saccharomyces cerevisiae*. *J. Biol. Chem.* **269**: 8792-8796.

Miller, G. A. D., Suzuki, N., Ciftci-Yilmaz, S. and Mittler, R. O. N. (2010). Reactive oxygen species homeostasis and signalling during drought and salinity stresses. *Plant Cell Environ.* **33**: 453–467.

Mondal, M. M. A., Puteh, A. B., Malek, M. A. and Rafii, M. Y. (2013). Salinity induced morpho-physiological characters and yield attributes in rice genotypes. *J. Food Agric. Environ.,* **11**: 610-614.

Monirifar, H. and Barghi, M. (2010). Identification and selection for salt tolerance in alfalfa (*Medicago sativa* L.) ecotypes via physiological traits. *Notulae Sci. Biol.* **1**: 63–66.

Mohamed, A.K. and Basalah M.O. (2015). The Active Role of Calcium Chloride on Growth and Photosynthetic Pigments of Cowpea "Vigna unguiculata L. (Walp)" Under Salinity Stress Conditions. *American-Eurasian J. Agric. & Environ. Sci.,* **15** (10): 2011-2020, 2015

Mohanty S., Wassmann R., Nelson A., Moya P. and Jagadish S.V.K. (2013). Rice and climate change: Significance for food security and vulnerability. IRRI Discussion Paper Series No. 49. Los Baños.

Molazem, D., Qubanov, E. M. and Dunyamaliyev, S. A. (2010). Role of proline, Na and chlorophyll content in salt tolerance of corn (*Zea mays* L.). *American-Eurasian J. Agric. Environ. Sci.* **9** (3): 319–324.

Moradi, F., Ismail, A.M., Gregorio, G. and Egdane, J. (2003). Salinity tolerance of rice during reproductive development and association with tolerance at seedling stage. *Indian J. Plant Physiol*. **8**: 105–116.

Mostofa, M. G., Hossain, M. A. and Fujita, M. (2015). Trehalose pretreatment induces salt tolerance in rice seedlings: oxidative damage and co-induction of antioxidant defense and glyoxalase systems. *Protoplsasma*. **252**: 461–475.

Munns R. and Termaat A.(1986). Whole-plant responses to salinity. *Aust. J. Plant. Physiol*. **13**: 143–160.

Munns R, Tester M. (2008). Mechanisms of salinity tolerance. *Annu Rev Plant Biol*.**59**:651–681.

Munns, R. (2005). Genes and salt tolerance: bringing them together. *New Phytol.* **167**:645–663.

Munns, R. (2002). Comparative physiology of salt and water stress. *Plant Cell Environ.*, **25**: 239-250.

Muranaka, S., Shimizu, K., Kato, M. (2002). A salt-tolerant cultivar of wheat maintains photosynthetic activity by suppressing sodium uptake. *Photosynthetica*. **40**: 509–515.

Nahar, K., Hasanuzzaman, M., Rahman, A., Alam, M.M., Mahmud, J.A., Suzuki, T. and Fujita, M. (2016). Polyamines confer salt tolerance in mung bean (Vigna radiata L.) by reducing sodium uptake, improving nutrient homeostasis, antioxidant defense, and methylglyoxal detoxification systems. *Front. Plant Sci.* **7**: 1104.

Nahar, K., Hasanuzzaman, M., Alam, M. M. and Fujita, M. (2015a). Roles of exogenous glutathione in antioxidant defense system and methylglyoxal detoxification during salt stress in mung bean. *Biol. Plant*. **59**: 745–756.

Negrão, S., Schmöckel, S.M. and Tester, M. (2017). Evaluating physiological responses of plants to salinity stress. *Ann. Bot.* **119**: 1–11.

Nio, S., Cawthray, G., Wade, L. and Colmer, T. (2011). Pattern of solutes accumulated during leaf osmotic adjustment as related to duration of water deficit for wheat at the reproductive stage. *Plant Physiol. Biochem*. **49**: 1126–37.

Nishida, K., Khan, N. M. and Shiozawa, S. (2009). Effects of salt accumulation on the leaf water potential and transpiration rate of pot-grown wheat with a controlled saline groundwater table. *Soil Sci. Plant Nutri*. **55**: 375–384.

Nounjan, N., Nghia, P. T. and Theerakulpisut, P. (2012). Exogenous proline and trehalose promote recovery of rice seedlings from salt-stress and differentially modulate antioxidant enzymes and expression of related genes. *J. Plant Physiol*. **169**(6): 596–604.

Nxele, X., Klein, A. and Ndimba, B. K. (2017). Drought and salinity stress alters ROS accumulation, water retention, and osmolyte content in sorghum plants. *S. Afr. J. Bot.* **108**: 261–266.

Pande, A. and Arora, S. (2017). Molecular strategies for development of abiotic stress tolerance in plants. *Cell Cellular Lif. Sci*. J. **2**(2): 000112.

Pandey, S., Fartyal, D., Agarwal, A., Shukla, T., James, D., Kaul, T., Negi, Y.K., Arora, S. and Reddy, M.K. (2017). Abiotic stress tolerance in plants: Myriad roles of ascorbate peroxidase. *Front. Plant Sci*. **8**: 581.

Parvin, K., Ahamed, K. A., Islam M. M. and Md. Haque M. N. (2015). Response of Tomato Plant Under Salt Stress: Role of Exogenous Calcium. *J. Plant Sci*. **10** (6): 222-233.

Pérez-López, U., Robredo, S.A., Lacuesta, M., Sgherri, C., Munoz, A., Navari-Izzo, F. and Mena-Petite, A. (2010). The oxidative stress caused by salinity in two barley cultivars is mitigated by elevated CO_2. *Physiol. Plant*. **135**:29-42.

Perveen, S., Shahbaz, M. and Ashraf, M. (2010). Regulation in gas exchange and quantum yield of photosystem II (PSII) in salt stressed and non-stressed wheat plants raised from seed treated with triacontanol. *Pak. J. Bot.* **42**: 3073–3081.

Pitman, M. G. and Läuchli, A. (2002). Global impact of salinity and agricultural ecosystems. In: Salinity: environment – plants – molecules. A. Läuchli, and U. Lüttge, (ed.). Kluwer Dordrecht. pp. 3–20.

Prasad, I., Gururaja Rao, G., Chinchmalatpure, A. R., Kumar, S., Ramesh, N. V., Singh, S.and Sharma, D. K. (2016). Morphophysiological traits imparting salinity tolerance in maize (*Zea mays* L.) hybrids under saline water irrigation in vertisols. Commu. *Soil Sci. Plant Anal*. **47**: 1930–1939.

Purseglove, J.W. (1985). Tropical Crops: Monocotyledons. Vol 1 and 2 comb. English Language Book Society/Longman, England. p.168.

Puteh, A. B. and Mondal, M. M. A. (2013). Salinity Effect on Dry Mass Partitioning in Different Plant Parts and Ion Uptake in Leaves of Rice Mutants. *J. Environ. Sci. & Natural Resources*,**6**(1): 239 – 245.

Qadir M, Quillerou E, Nangia V. (2014). Economics of salt-induced land degradation and restoration. Natural Resources Forum **38**: 282–295.

Rahman, A., Nahar, K., Hasanuzzaman, M. and Fujita, M. (2016a). Calcium supplementation improves Na+/K+ ratio, antioxidant defense and glyoxalase systems in salt-stressed rice seedlings. *Front. Plant Sci*. 7. doi.10.3389/fpls.2016.00609.

Rahman, A., Hossain, M.S., Mahmud, J., Nahar, K., Hasanuzzaman, M. and Fujita, M. (2016b). Manganese-induced salt stress tolerance in rice seedlings: Regulation of ion homeostasis, antioxidant defense and glyoxalase systems. *Physiol. Mol. Biol. Plants*. doi:10.1007/s12298-016-0371-1

Rao, P. S., Mishra, B., Gupta, S. R., Rathore, A. (2008) Reproductive stage tolerance to salinity and alkalinity stresses in rice genotypes. *Plant Breed* **127**:256-261.

Raza, S. H., Athar, H. R. and Ashraf, M. (2006). Influence of exogenously applied glycinebetaine on the photosynthetic capacity of two differently adapted wheat cultivars under salt stress. *Pak. J. Bot*. **38**: 341–351.

Reddy, M. P. and Vora, A. B. (1986). Changes in pigment composition, hill reaction activity and saccharides metabolism in bajra leaves under NaCl salinity. *Photosynthica*, **20**: 50-55.

Rengasamy, P. (2006). World salinization with emphasis on Australia. *J. Exp. Bot*. **57**: 1017–1023.

Rengel, Z. (1992). The role of calcium in salt toxicity. *Plant Cell Environ*. **15**(6): 625-632.

Sabir, P., Ashraf, M., Hussain, M. and Jamil, A. (2009). Relationship of photosynthetic pigments and water relations with salt tolerance of proso millet (*Panicum Miliaceum* L.) accessions. *Pak. J. Bot*. **41**: 2957–2964.

Saruhan, N., Terzi, R., Saglam, A. and Kadioglu, A. (2009). The relationship between leaf rolling and ascorbate-glutathione cycle enzymes in apoplastic and symplastic areas of *Ctenanthe setosa* subjected to drought stress. *Biol. Res*. **42**: 315–326.

Scheres, B. and Van der Putten, W.H. (2017). The plant perception connects environment to development. *Nature.* **543**: 337–345.

Shabala, S., Demidchick, V., Shabala, L., Cuin, T. A., Smith, S. J. and Miller A. J.(2006). Extracellular Ca^{2+} ameliorates NaCl-induced K^+ loss from Arabidopsis root and leaf cells by controlling plasma membrane K^+ permeable channels. *Plant Physiol*. **141**: 1653–1665.

Shabala, S. and Pottosin, I. (2014). Regulation of potassium transport in plants under hostile conditions:implications for abiotic and biotic stress tolerance. *Physiol. Plant.* **151**: 257–279.

Shanker, A. K. and Venkateswarlu, B. (2011). Abiotic stress in plants mechanisms and adaptations. InTech.pp.2-4.

Sharma, P., Jha, A. B., Dubey, R. S. and Pessarakli, M. (2012). Reactive oxygen species, oxidative damage and antioxidative defense mechanism in plants under stressful conditions. *J. Bot*. 217037: 1–26.

Shelley, I., Takahashi-Nosaka, M., Kano-Nakata, M., Haque, M. and Inukai, Y. (2016). Rice cultivation in Bangladesh: present scenario, problems, and prospects: a review. J. *Intl. Cooper. Agric. Dev.* **14**: 20–29.

Shi, H. Z., Ishitani, M., Kim, C. and Zhu, J. K. (2000). The Arabidopsis thaliana salt tolerance gene SOS1 encodes a putative Na^+/H^+ antiporter. Proc. Nat. Acad. Sci. USA **97**: 6896-6901.

Sholi, N. J. Y. (2012). Effect of salt stress on seed germination, plant growth, photosynthesis and ion accumulation of four tomato cultivars. *Am. J. Plant Physiol.* **7**(6): 269–275.

Siddiqui, Z. S., Cho, J. I., Park, S. H., Kwon, T. R., Ahn, B. O., Lee, G. S., Jeong, M. J., Kim, K. W., Lee, S. K. and Park, S. C. (2014). Phenotyping of rice in salt stress environment using high-throughput infra-red imaging. *Acta Bot. Croat*. **73**: 149–158.

Singh, R.K, Gregorio, G.B. and Ismail, A.M. (2008). Breeding rice varieties with tolerance to salt stress. *J. Ind. Soc. Coastal Agric. Res*. **26**:16–21.

Smirnoff, N., Cumbes, Q. J. (1989). Hydroxyl radical scavenging activity of compatible solutes. *Phytochemistry*. **28**: 1057–60.

Soualem, S. Adda, A., Belkhodja, M., and Merah, O. (2014). Calcium supply reduced effect of salinity on growth in the Mediterranean shrub (*Atriplex halimus* L.). *Life SciJ* ;**11**(2):278-284.

Srivastava, R. K., Pandey, P., Rajpoot, R., Rani, A., Gautam, A. and Dubey R. S. (2014). Exogenous application of calcium and silica alleviates cadmium toxicity by suppressing oxidative damage in rice. *Protoplasma* 252 959–975. 10.1007/s00709-014-0731-z

Stepien, P. and Johnson G. N. (2009). Contrasting responses of photosynthesis to salt stress in the glycophyte Arabidopsis and the halophyte Thellungiella: role of the plastid terminal oxidase as an alternative electron sink. *Plant Physiol*. **149**: 1154–1165.

Steudle, E. (2000). Water uptake by roots: effects of water de fi cit. *J Exp Bot* **51**:1531–1542

Sultana, N., Alimon, A. R., Haque, K. S., Sazili, A. Q., Yaakub, H. and Hossain, S. M. J.(2014). The effect of cutting interval on yield and nutrient composition of rice.

Szabados, L. and Savouré, A. (2010). Proline: a multifunctional aminoacid. Trends *Plant Sci*. **15**: 89–97.

Tabatabaeian, J. (2014). Effect of Calcium Nutrition on Reducing the Effects of Salinity on Tomato Plant. *American Journal of Plant Nutrition and Fertilization Technology,* **4**: 11-17.

Tanji K. K. (2002). Salinity in the soil environment. In Lauchli A, Luttge U (eds.) Salinity Environment-Plant –Molecules. Kluwer Academic Publishers, Dordrecht, The Netherlands: 21-51.

Tanou, G., Job, C., Rajjou, L., Arc, E., Belghzi, M., Diamantidis, G., Molassiotis, A. and Job, D. (2009). Proteomics reveal the overlapping roles of hydrogen peroxide and nitric oxide in the acclimation of citrus plants to salinity. *Plant J.* **60**:795-804.

Termaat, A. and Munns, R. (1986). Use of concentrated macronutrient solutions to separate osmotic from NaCl specific effects on plant growth. *Func. Plant Biol.* **13**: 509–22.

Tavakkoli, E., Fatehi, F., Coventry, S., Rengasamy, P. and McDonald, G.K. (2011). Additive effects of Na^+ and Cl^- ions on barley growth under salinity stress. *J. Exp. Bot.* **62**: 2189–2203.

Tunçtürk, M., Tunçtürk, R., Yildirim, B. and Çiftçi, V. (2011). Changes of micronutrients, dry weight and plant development in canola (*Brassica napus* L.) cultivars under salt stress. *Afr. J. Biotechnol*. **10**: 3726–3730.

Tzortzakis, N. G. (2010). Potassium and calcium enrichment alleviate salinity-induced stress in hydroponically grown endives. *Hort. Sci. (Prague).* **37**: 155–162.

Uddin, I.M.D., Rashid, H.M.D., Khan, N., Perveen, M.S.T.F., Tai, T.H and Tanaka, K. (2007). Selection of promising salt tolerant rice mutants derived from cultivar 'DREW' and their antioxidant enzymatic activity under stress. SABRAO. *J. Breed. Genet*. **39**(2): 89-98.

Ueda, A., Yahagi, H., Fujikawa, Y., Nagaoka ,T., Esaka, M., Calcaño, M., González, M. M., Martich, J. D. H., Saneoka, H. (2013). Comparative physiological analysis of salinity tolerance in rice, *Soil Sci. Plant Nutri*. **59**(6): 896–903.

Umali, D.L. (1993). Irrigation-Induced Salinity a Growing Problem for Development and the Environment; The Word Bank, Washington, DC, USA.

Umar, S., Diva, I., Anjum, N. A., Iqbal, M., Ahmad, I. and Pereira, E. (2011). Potassium-induced alleviation of salinity stress in *Brassica campestris* L. *Cent. Eur. J. Biol.* **6**(6): 1054–1063.

United States Department of Agriculture. (2016). Bibliography on Salt Tolerance. Fibres, Grains and Special Crops; Brown, G.E., Jr., Ed.; Salinity Laboratory United States Department of Agriculture Research Service: Riverside, CA, USA :http://www.Ars.Usda.Gov/services/docs.Htm?Docid=8908

USDA (2018). Foreign Agricultural Service/USDA 26 October 2018.Office of Global Analysis. https://apps.fas.usda.gov/psdonline/circulars/grain-rice.pdf

Vysotskaya, L., Hedley, P. E., Sharipova, G., Veselov, D., Kudoyarova, G., Morris, J. and Jones, H. G. (2010). Effect of salinity on water relations of wild barley plants differing in salt tolerance. *AoB Plant*. doi: 10.1093/aobpla/plq006

Wang, H., Zhang, M., Guo, R., Shi, D., Liu, B., Lin, X. and Yang, C.(2012). Effects of salt stress on ion balance and nitrogen metabolism of old and young leaves in rice (*Oryza sativa* L.). *BMC Plant Biol*. **12**: 194.

Wang, X., Wang, W., Huang, J., Peng S. and Xiong, D. (2017). Diffusional conductance to CO_2 is the key limitation to photosynthesis in salt-stressed leaves of rice (*Oryza sativa*). *Physiol. Plant.* doi:10.1111/ppl.12653

Wani, A. S., Ahmad, A., Hayat, S., and Fariduddin, Q. (2013). Salt-induced modulation in growth, photosynthesis and antioxidant system in two varieties of *Brassica juncea*. *Saudi J.Biol. Sci*. **20**(2): 183–193.

Wei, X., Wang, R., Cao, L., Yuan, N., Huang, J., Qiao, W., Zhang, W., Zeng, H. and Yang, Q. (2012). Origin of Oryza sativa in China inferred by nucleotide polymorphisms of organell DNA. *PLoS ONE.* **7**(11): e49546.

Wei, J., Li, C., Li, Y., Jiang, G. and Cheng, G. (2013). Effects of external potassium (K) supply on drought tolerances of two contrasting winter wheat cultivars. *PLoSONE.* 8: e69737. doi:10.1371/journal.pone.0069737

White P. J. and Broadley M. R. (2003). Calcium in plants. *Ann. Bot*. **92**: 487–511.

Wilmoth, J. (2015). Global population projections by the United Nations. Joint Statistical Meetings Session 151: ―Better demographic forecasts, better policy decisions‖ 10 August 2015

Wopereis, M.C.S., Ceuppens, J., Boivin, B., Ndiaye, A. M. and Kane, A. (1998). Preserving soil quality under irrigation in the Senegal river valley. *Nethreland J. Agril. Sci.*. **46**: 97-107.

Wu, G. Q., Wang, S. M. (2012). Calcium regulates K^+/Na^+ homeostasis in rice (*Oryza sativa* L.) under saline conditions. *Plant Soil Environ*. **58**: 121–127.

Yang, J. Y., Zheng, W., Tian, Y., Wu, Y. and Zhou, D. W. (2011). Effects of various mixed salt-alkaline stresses on growth, photosynthesis, and photosynthetic pigment concentrations of *Medicago ruthenica* seedlings. *Photosynthetica*. **49**: 275–284.

Yokoi, S., Bressan, R.A., and Hassegawa, P.M. (2002). Salt stress tolerance of plants. JIRCAS working Report. pp.25-33.

Yousuf, P.Y., Ahmad, A., Hemant, Ganie, A. H., Aref, I. M. and Iqbal M. (2015). Potassium and Calcium Application Ameliorates Growth and Oxidative Homeostasis in Salt-Stressed Indian Mustard (*Brassica Juncea*) Plants. *Pak. J. Bot*. **47**(5): 1629-1639.

Zain, N. A. M., Ismail, M. R., Puteh, A., Mahmood, M. and Islam, M. R. (2014). Drought tolerance and Ion accumulation of rice following application of additional potassium fertilizer. *Commun. Soil Sci. Plant Anal*. doi:10.1080/00103624.2014.932374

Zeid, I. M. (2009). Trehalose as osmoprotectant for maize under salinity-induced stress research. *J. Agr. Biol*. Sci. **5**: 613–622.

Zeng, L., Shannon, M. C. and Lesch, S. M. (2001). Timing of salinity stress affects rice growth and yield components. *Agric. Water Manag*. **48**: 191–206.

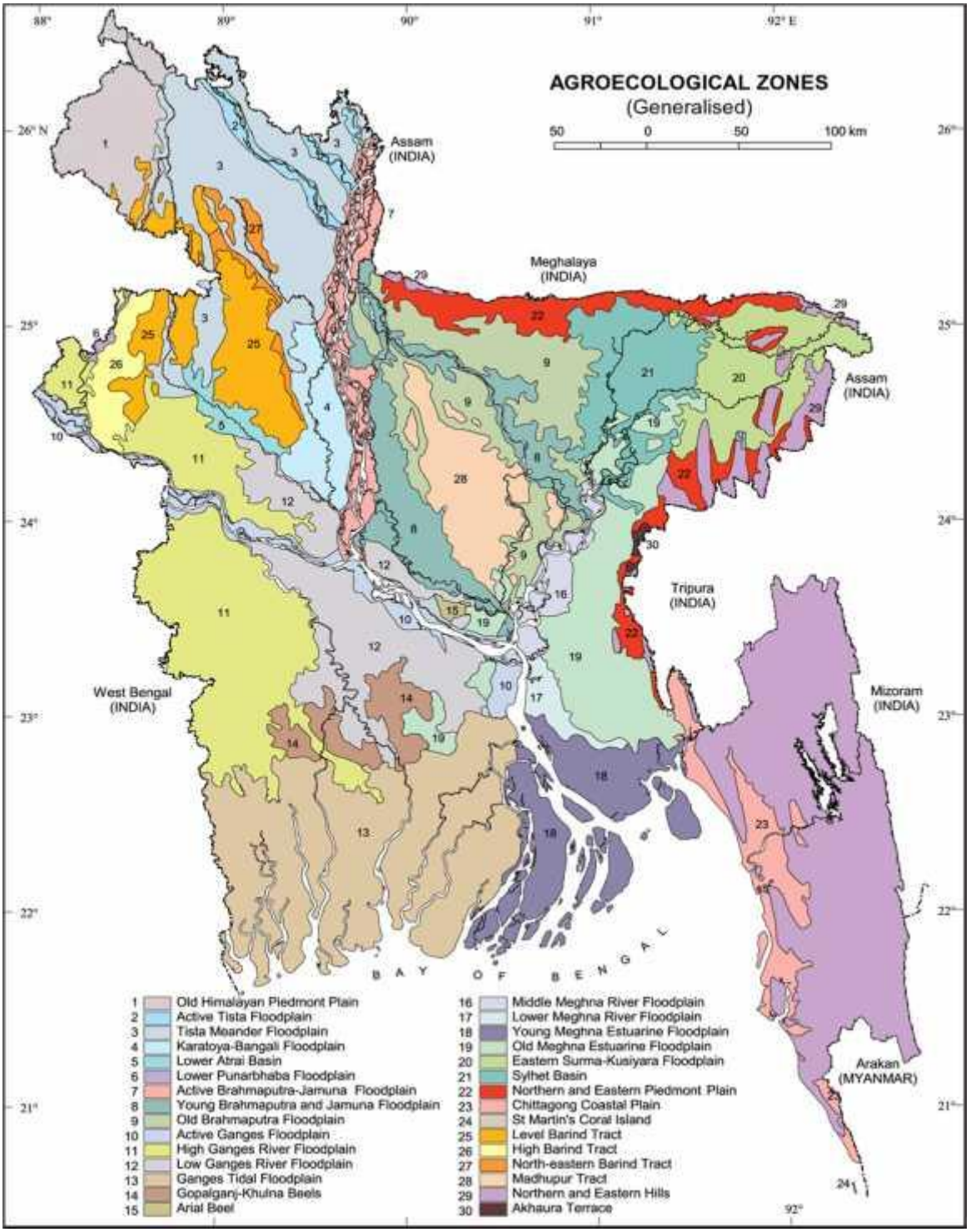
Zeng, L. and Shannon, M. C. (2000). Salinity effects on seedling growth and yield components of rice. *Crop Sci.* **40**(4): 996-1003.

Zhang, L., Ma, H., Chen, T., Pen, J., Yu, S. and Zhao, X. (2014). Morphological and physiological responses of cotton (*Gossypium hirsutum* L.) plants to salinity. *PLoS ONE*. 9(11): e112807. doi. 10.1371/journal.pone.0112807

Zhang, P., Senge, M. and Dai, Y. (2017). Effects of salinity stress at different growth stages on tomato growth, yield and water use efficiency. *Commun. Soil Sci. Plant Anal.* doi:10.1080/00103624.2016.1269803

APPENDICES

Appendix I. Experimental location on the map of Agro-ecological Zones of Bangladesh



Appendix II. Morphological characteristics of the experimental field

Morphology	Characteristics
Location	SAU Farm, Dhaka.
Agro-ecological zone	Madhupur Tract (AEZ- 28)
General Soil Type	Deep Red Brown Terrace Soil
Parent material	Madhupur Terrace.
Topography	Fairly level
Drainage	Well drained
Flood level	Above flood level

(SAU Farm, Dhaka)

Appendix III. Initial physical and chemical characteristics of the soil

Characteristics	Value
Mechanical fractions: % Sand (2.0-0.02 mm) % Silt (0.02-0.002 mm) % Clay (<0.002 mm)	 22.26 56.72 20.75
Textural class	Silt Loam
pH (1: 2.5 soil- water)	5.9
Organic Matter (%)	1.09
Total N (%)	0.028
Available K (ppm)	15.625
Available P (ppm)	7.988
Available S (ppm)	2.066

(SAU Farm, Dhaka)

Appendix IV. Analysis of variance of the data on plant height as influenced by combined effect of salt concentrations and calcium levels of rice

Source of variation	df	Mean square of plant height at different days after transplanting (DAT)			
		30	60	90	At harvest
Replication	2	21.92	3.104	278.13	225.20
Salt concentrations (A)	3	575.33**	1502.63**	1582.66**	1463.57**
Calcium levels (B)	2	249.74**	336.69*	414.08**	383.70*
Salt concentrations (A) X Calcium levels (B)	6	2.15**	27.31**	24.80**	18.13**
Error	22	34.70	68.57	67.51	86.45

**Significant at 1% level of significance
*Significant at 5% level of significance
^{NS} Non significant

Appendix V. Analysis of variance of the data on tillers plant^{-1} as influenced by combined effect of salt concentrations and calcium levels of rice

Source of variation	df	Mean square of tillers plant^{-1} at different days after transplanting (DAT)		
		30	60	90
Replication	2	2.90	1.44	0.58
Salt concentrations (A)	3	99.04**	169.88**	139.89**
Calcium levels (B)	2	28.58**	33.36**	40.75**
Salt concentrations (A) X Calcium levels (B)	6	6.51**	0.55**	0.86**
Error	22	0.61	1.26	1.71

**Significant at 1% level of significance
*Significant at 5% level of significance
^{NS} Non significant

Appendix VI. Analysis of variance of the data on SPAD value as influenced by combined effect of salt concentrations and calcium levels of rice

Source of variation	df	Mean square of SPAD value at different days after transplanting (DAT)	
		60	90
Replication	2	3.74	12.38
Salt concentrations (A)	3	160.74**	269.39**
Calcium levels (B)	2	174.93**	80.21**
Salt concentrations (A) X Calcium levels (B)	6	8.53**	2.15**
Error	22	11.42	10.71

**Significant at 1% level of significance
*Significant at 5% level of significance
^{NS} Non significant

Appendix VII. Analysis of variance of the data on leaf dry weight plant^{-1}, root dry weight plant^{-1}, shoot dry weight plant^{-1} and total dry weight plant^{-1} as influenced by combined effect of salt concentrations and calcium levels of rice

Source of variation	df	Mean square of			
		Leaf dry weight plant^{-1}	Root dry weight plant^{-1}	Shoot dry weight plant^{-1}	Total dry weight plant^{-1}
Replication	2	2.89	1.46	0.83	4.61
Salt concentrations (A)	3	251.57**	42.37**	485.60**	1899.49**
Calcium levels (B)	2	95.88**	23.98**	139.03**	694.28**
Salt concentrations (A) X Calcium levels (B)	6	4.67*	1.15NS	0.51**	3.19**
Error	22	1.71	1.47	7.46	28.81

**Significant at 1% level of significance
*Significant at 5% level of significance
NS Non significant

Appendix VIII. Analysis of variance of the data on root length, leaf area, leaf membrane stability and relative water content as influenced by combined effect of salt concentrations and calcium levels of rice

Source of variation	df	Mean square of		
		Leaf area	Leaf membrane stability	Relative water content
Replication	2	38.56	30.75	85.80
Salt concentrations (A)	3	1495.11**	4056.91**	1054.85**
Calcium levels (B)	2	295.19**	440.30**	220.20*
Salt concentrations (A) X Calcium levels (B)	6	7.03**	22.57*	4.40**
Error	22	27.89	15.11	50.05

**Significant at 1% level of significance
*Significant at 5% level of significance
NS Non significant

Appendix IX. Analysis of variance of the data on days to flowering, panicles plant^{-1}, effective tillers plant^{-1}, ineffective tillers plant^{-1}and panicle length as influenced by combined effect of salt concentrations and calcium levels of rice

Source of variation	df	Mean square of			
		Days to flowering	Effective tillers plant^{-1}	Ineffective tillers plant^{-1}	Panicle length
Replication	2	3.08	3.36	0.01	4.29
Salt concentrations (A)	3	178.74**	149.85**	1.38**	121.22**
Calcium levels (B)	2	22.75**	35.03**	2.47**	23.56**
Salt concentrations (A) X Calcium levels (B)	6	21.94**	1.44*	0.15*	0.21**
Error	22	2.30	1.15	0.07	3.40

**Significant at 1% level of significance
*Significant at 5% level of significance
^{NS} Non significant

Appendix X. Analysis of variance of the data on filled grains panicle^{-1}, unfilled grains panicle^{-1}, 1000 grain weight and grain yield plant^{-1} as influenced by combined effect of salt concentrations and calcium levels of rice

Source of variation	df	Mean square of			
		Filled grains panicle^{-1}	Unfilled grains panicle^{-1}	1000 grain weight	Grain yield plant^{-1}
Replication	2	261.36	2.53	2.24	7.54
Salt concentrations (A)	3	16927.78**	290.11**	197.97**	853.53**
Calcium levels (B)	2	1564.53**	169.36**	37.23**	122.72**
Salt concentrations (A) X Calcium levels (B)	6	20.97**	20.58**	1.48**	4.86*
Error	22	124.42	4.13	3.21	4.17

**Significant at 1% level of significance
*Significant at 5% level of significance
^{NS} Non significant