POTASSIUM-INDUCED ANTIOXIDANT DEFENSE AND REGULATION OF PHYSIOLOGICAL PROCESSES TOWARDS DROUGHT STRESS TOLERANCE IN WHEAT

ABDUL AWAL CHOWDHURY MASUD

DEPARTMENT OF AGRONOMY

SHER-E-BANGLA AGRICULTURAL UNIVERISITY,

DHAKA-1207

JUNE, 2017

POTASSIUM-INDUCED ANTIOXIDANT DEFENSE AND REGULATION OF PHYSIOLOGICAL PROCESSES TOWARDS DROUGHT STRESS TOLERANCE IN WHEAT

BY

ABDUL AWAL CHOWDHURY MASUD

REGISTRATION NO. 10-03844

A Thesis

Submitted to the Faculty of Agriculture,

Sher-e-Bangla Agricultural University, Dhaka,

In partial fulfillment of the requirements

for the degree of

MASTER OF SCIENCE

IN

AGRONOMY

SEMESTER: JANUARY-JUNE, 2017

Approved by:

(Prof. Dr. Md. Fazlul Karim)

Department of Agronomy

Supervisor

(Prof. Dr. Parimal Kanti Biswas)

Department of Agronomy

Co-supervisor

(Prof. Dr. Md. Shahidul Islam)

Chairman

Examination Committee

Department of Agronomy
Sher-e Bangla Agricultural University
Sher-e-Bangla Nagar, Dhaka-1207

CERTIFICATE

This is to certify that thesis entitled, ***"Potassium-induced antioxidant defense and regulation of physiological processes towards drought stress tolerance in wheat"*** *submitted to the Faculty of Agriculture, Sher-e-Bangla Agricultural University, Dhaka, in partial fulfillment of the requirements for the degree of* ***MASTER OF SCIENCE in AGRONOMY****, embodies the result of a piece of bonafide research work carried out by* ***Abdul Awal Chowdhury Masud****, registration No.* ***10-03844*** *under my supervision and guidance. No part of the thesis has been submitted for any other degree or diploma.*

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

Dated:
Place: Dhaka, Bangladesh

(Dr. Md. Fazlul Karim)
Professor
Department of Agronomy
Sher-e-Bangla Agricultural University
Supervisor

DEDICATED
TO MY
BELOVING PARENTS

ACKNOWLEDGEMENTS

*The author strongly believes that, Mere words are inadequate to express the sense of gratitude and indebtedness to those whose assistance was indispensable for the completion of his thesis work. However, fore mostly, All praises to Almighty **ALLAH**, The Omnipotent Creator of this universe for his granted bounties and who has bestowed the author with wisdom and patience for successful accomplishment of this piece of research work. The pleasure is to express profound gratitude and enormous thankfulness to the author's respected parents, who endured much struggle and hardship to prosecute his studies, thereby receiving proper education and guidelines for being a good human.*

*At the very outset, the author wishes to express his sincere gratitude and earnest appreciation to respected Supervisor, **Prof. Dr. Md. Fazlul karim**, Department of Agronomy, Sher-e-Bangla Agricultural University, Dhaka, for continuous direction, constructive criticism, encouragement and valuable suggestions throughout the research period and to prepare the manuscript.*

*The author feels unfathomable euphoria and enormous indebtedness to pronounce his heartfelt adoration and gratitude to one of his ever seen most dynamic, versatile and sagacious pedagogue, **Prof. Dr. Mirza Hasanuzzaman**, Department of Agronomy, Sher-e-Bangla Agricultural University, Dhaka, for scholastic supervision, relentless guidance, unfailing support and encouragement, during stay in Japan for the purpose of research. His supervision has provided an ideal intellectual environment to learn science for which author will always be grateful to him. Furthermore, the author will always be thankful to him for paving the opportunity to conduct research work in Laboratory of Plant Stress Responses, Department of Applied Biological Sciences, Faculty of Agriculture, and Kagawa University, Japan.*

*The author also feels to share his deep sense of regards to his respected Co-Supervisor, **Prof. Dr. Parimal Kanti Biswas,** Department of Agronomy, Sher-e- Bangla Agricultural University, Dhaka, for his adept guidance, kind cooperation, and appropriate suggestions in conducting the research work. The author feels elevated to express his heartfelt thanks to the Departmental Chairman **Prof. Dr. Md. Shahidul Islam** along with all other teachers and staff members of the Department of Agronomy, Sher-e-Bangla Agricultural University, Dhaka, for their co-operation during the period of the study.*

*This is author's countless rejoice and proud privilege to express his heartiest gratitude and enormous thanks to **Prof. Dr. Masayuki Fujita**, Laboratory of Plant Stress Responses, Faculty of Agriculture, Kagawa University, Japan for inviting author as a foreign research student and providing laboratory facilities along with his kind appreciation, continuous supervision and constructive criticism and helpful suggestions in carrying out the research work. Author is duly grateful to him as without his intense and interactive co-operation this work would never been significantly possible.*

*The author avails this opportunity to express his sincere thanks and gratitude to the Government of Bangladesh through its **Ministry of Science and Technology (MoST)** for providing financial support (NST fellowship) to conduct the research work.*

The author must also acknowledge the Laboratory of Plant Stress Responses, Faculty of Agriculture, and Kagawa University, Japan through its Funded by ***Japan Student Services Organization (JASSO)*** *for bearing the expense in Japan and to conduct the research work.*

It would author injustice if not pay thanks to ***Dr. Kamrun Nahar,*** *Associate Professor, Department of Agricultural Botany and* ***Dr. Anisur Rahman****, Associate Professor, Department of Agronomy,* ***Jubayer Al Mahmud****, Associate Professor, Department of Agroforestry and Environmental Sciences, Sher-e-Bangla Agricultural University, Dhaka, for their incredible help and support, valuable suggestions and cooperation during the entire study period.*

The author wishes to convey his wholehearted appreciation to his lab mates, Sayed Mohammad Mohsin, Khurshida parvin Hira, Md. Shahadat Hossain, Mahmudol Hasan Sohag, Md. Shahdat Hossen, Moumita, Bushra Al-Jannat and last but not the least ***M.H.M Borhannuddin Bhuyan*** *for their earnest help as well as heartiest cooperation and encouragement.*

Finally, the author is highly delighted to express his heartfelt thanks to rest of other family members, and to remember his all friends, seniors and well-wishers from Sher-e-Bangla Agricultural University for their prayers, encouragement, constant inspiration and moral support for higher study. May Allah bless and protect them all.

The Author

POTASSIUM-INDUCED ANTIOXIDANT DEFENSE AND REGULATION OF PHYSIOLOGICAL PROCESSES TOWARDS DROUGHT STRESS TOLERANCE IN WHEAT

ABSTRACT

Drought imparts injuries in plant through elevated production of reactive oxygen species viz. ($O_2^{\bullet}$, $OH^{\bullet}$, H_2O_2 and $^1O_2$). Potassium (K) triggers numerous ameliorative functions against oxidative damages caused by drought. To investigate K attenuating oxidative damage and promotion of antioxidant defense in wheat (*Triticum aestivum* L. cv. BARI Wheat-21), an experiment was carried out at the Laboratory of Plant Stress Responses, Faculty of Agriculture, Kagawa University, Japan, under controlled environment of green house during June, 2017 to December, 2017. A total number of nine treatments were combined considering three K doses viz. 0 mM (-K), 6 mM (+K) & 12 mM (+2K) in Hoagland nutrient solution exposed to 3 distinct water regimes viz. 100% FC, 50% FC, and 20% FC for 9 days. The experiment was designed in CRD model. Imposed drought stress notably reduced Plant height, fresh weight (FW), dry weight (DW), leaf relative water content (RWC %) along with chlorophyll content in dose dependent manner. Increased drought stress elevated the amount of oxidative markers viz. malondialdelyde (MDA), hydrogen peroxide (H_2O_2), methyl glyoxal (MG) and proline (Pro). Wheat seedlings treated with drought stress resulted in notable decrease of ascorbate (AsA), reduced glutathione (GSH) and increase of glutathione disulphide (GSSG). Enzymatic activities of AsA-GSH cycle enzymes were declined except the ascorbate peroxidase (APX), which was increased under drought stress combination. On the contrary, potassium (K) supply resulted in improved Chlorophyll contents, and water status along with increased GSH content and reduced the GSSG content which ultimately improved GSH/GSSG ratio. However, K reduced MDA, H_2O_2, Proline contents and up regulated the antioxidant enzymes activities. Exogenous K reduced the ROS production through increasing AsA. Additionally, In K treated plant activities of glyoxalase I and glyoxalase II enzymes reduced the levels of MG and overall increased the activity of glyoxalase system. However, plant showed better physiological performances under 50% drought stress with 12 mM K was observed in the overall study. Therefore it can be concluded that K confers drought stress tolerance in wheat seedlings through up regulating antioxidant defense and glyoxalase system which helped in better seedling establishment.

LIST OF CONTENTS

LIST OF CONTENT Continue'd

LIST OF CONTENT Continue'd

LIST OF CONTENT Continue'd

LIST OF TABLES

LIST OF FIGURES

LIST OF FIGURES Continue'd

LIST OF PLATES

LIST OF APPENDICES

LIST OF ACRONYMS AND ABBREVIATION

ABBREVIATION	ELABORATION
µM	Micromolar
$1O_2$	Singlet Oxygen
ANOVA	Analysis of Variance
AO	Ascorbate oxidase
APX	Ascorbate peroxidase
AsA	Ascorbic acid/ Ascorbate
ATP	Adenosine triphosphate
BARI	Bangladesh Agricultural Research Institute
BAU	Bangladesh Agricultural University
BBS	Bangladesh Bureau of Statistics
CAT	Catalase
Chl	Chlorophyll
DAE	Department of Agricultural Extension
DHA	Dehydroascorbate
DHAR	Dehydroascorbate reductase
DW	Dry weight
et al.	and others
FAO	Food and Agriculture Organization
FAOSTAT	Food and Agriculture Organization Statistics
FW	Fresh weight
Gly I	Glyoxalase I
Gly II	Glyoxalase II
GPX	Glutathione peroxidase
GR	Glutathione reductase
GSH	Glutathione
GSSG	Glutathione disulfide

LIST OF ACRONYMS AND ABBREVIATION Continue'd

ABBREVIATION	ELABORATION
GST	Glutathione S-tranferase
IPCC	Intergovernmental Panel on Climate Change
LSD	Least Significant Difference
MDA	Malondiadehyde
MDHA	Monodehydroascorbate
MDHAR	Monodehydroascorbate reductase
MG	Methylglyoxalase
mM	Milimolar
NADPH	Nicotinamide adenine dinucleotide phosphate
Nm	Nanometer
$O_2^{\bullet}$	Superoxide radical
$OH^{\bullet}$	Hydroxyl radical
PAR	Photosynthetically active radiation
PEG	Poly Ethylene Glycol
POX/ POD	Peroxidase
Pro	Proline
PS I	Photosystem I
PS II	Photosystem II
ROS	Reactive Oxygen Species
RuBisCo	Ribulose-1,5-bisphosphate carboxylase/oxygenase
RWC	Relative water content
SLG	S-lactoyl-glutathione
SOD	Super Oxide Dismutase
SRDI	Soil Resource Development Institute
USDA	United States Department of Agriculture
XOD	Xanthine Oxidase

CHAPTER 1

INTRODUCTION

Wheat (*Triticum aestivum* L.) is considered as one of the predominant cereal crop under the *Poaceae* (*Gramineae*) family that occupies global rank one in aspect of worldwide production and consumption. It is the staple food of more than 36% of world population that supplies nearly 55% of the carbohydrates and 20% of the food calories consumed as human diet globally (Lobell and Gourdji, 2012; Hasanuzzaman *et al*., 2017). Around the world, about 30% area is covered by wheat cultivation (Lobell and Gourdji, 2012). In 2016, world wheat production was 749 million tons, which made it the second most produced cereal after maize (1.03 billion tons) (FAOStat, 2016). In Bangladesh it is the second most significant grain crop after the staple grain Rice. The national wheat production during the year 2016-2017 was 1.422 million metric tons from 0.429 million hectares of cultivated land with an average 3.32 metric t/ha while in 2015-2016 it was 1.348 million metric tons from 0.445 million hectares of cultivated land with an average 3.03 metric t/ha (DAE, 2018). However, the wheat yield of Bangladesh is much lower comparing the rest other wheat producing countries in the world due to the fact of growing wheat under rainfed condition (Bazzaz, 2013).

Estimated world population by 2050 will be around 9-10 billion which will require double the existing food production in order to feed this vast population (Waraich *et al*., 2011). The food productivity is seriously hampered due to the effect of various abiotic and biotic stresses. Survival, biomass production and crop yield are drastically reducing due to the negative impact of these stresses (Amtmann *et al*., 2004; Agarwal *et al*., 2006). Global climate changes due to unpredictable environmental conditions now become one of the most disastrous threat to the current and future agriculture. The deleterious effect of climate change impact on global agriculture in terms of both biotic and abiotic stresses,

which ultimately reduces plant growth followed by declined yield attributes (Hasanuzzaman *et al*., 2012a). Among various abiotic stresses, drought stress encompass a significant position due to its nature of destruction and losses to crops (Kusvuran *et al*., 2011). To encounter this challenge, increase of yield potential is a must through reducing the yield losses caused by different kinds of biotic and abiotic stresses including drought (Tuteja *et al*., 2012). Plants frequently have to experience a number of environmental adversities like drought, salinity, temperature extremes, toxic metals etc. more or less every year which accounts up to 50% yield reduction annually. These forms of abiotic factors imparts detrimental effect on plant survival, biomass production and yield (Mantri *et al*., 2012; Hasanuzzaman *et al*., 2017).

Drought or water deficit stress is an edaphic stress which is considered as the most pernicious from of abiotic stress (Cattivelli *et al*., 2008; Pennisi, 2008; Farooq *et al*., 2009;) and may limit more than 50% of world agricultural productivity (Reynolds *et al*., 2007; Comas *et al*., 2013) by considerable reduction of plant growth and shoot production (Ehdaie *et al*., 2008, 2012). Drought stress cumulatively effects on all plant tissues through impairing the agronomic, morphological, physiological, biochemical and metabolic traits and eventually decreases the yield attributes (Cochard *et al*., 2002).

Water stress drastically reduces the leaf respiration and stomatal conductance, photosynthetic efficiency, carboxylation and water-use efficiency (WUE), carbon dioxide (CO_2) diffusion and transpiration rate, enzymatic activities etc., (Demirevska *et al*., 2010; Hasanuzzaman *et al*., 2013). Considering the facts, increasing plant tolerance against the drought is a big challenge to future agricultural productivity (Chaves *et al*., 2003).

Production of reactive oxygen species (ROS) is a naturally occurring process in plants that associates both photosynthetic and respiratory metabolism in cells (Miyake, 2010). Cumulative interactions of these ROS radicals imparts oxidative

damage to fundamental biomolecules viz., nucleic acids, proteins, lipids along with cell membrane damages (Ahmed *et al*., 2010). While plant subject to drought stress, the balance between production and utilization of ROS is altered thus effect the antioxidant system (Gao *et al.*, 2008; Pandey and Sukla, 2015).
To check the oxidative damages, plant has developed a strong and complex chain of antioxidant defense mechanism comprising the up regulation of both enzymatic and non-enzymatic components (Gupta *et al*., 2009; Hasanuzzaman and Fujita, 2013). Each compartment of cell contains one or more antioxidants that acts on detoxifying a particular ROS (Mittler, 2002; Ahmed *et al*., 2010). Furthermore, methylglyoxal (MG) is another oxidative stress indicator that causes damage to plant under stress (Hasanuzzaman *et al*., 2011, 2012b). However, plant tolerant to drought is entirely dependent on the amount of excess ROS scavenged by plant antioxidant system (Parida and Das, 2005; Hasanuzzaman *et al*., 2011 and Alam *et al*., 2013).

Potassium is one of the most essential nutrient element for plant growth and development. In plant cell, potassium plays the irreplaceable role of enzyme activation and water content hence maintaining the turgidity for each cell. It plays an important role in survival of crop plants under water stress condition. Potassium application under water stress moderates the adverse effects of water shortage on plant growth (Sangakkara *et al*., 2001). Yield limiting effects of water deficit could be overcome by increasing potassium supply (Damon and Rengel, 2007). During stress conditions, reactive oxygen species (ROS) formation was induced and oxidative damage to cells was nullified by potassium application (Foyer *et al*., 2002). Potassium influences the water economy and crop growth through its effects on water uptake, root growth, maintenance of turgor, transpiration and stomatal regulation (Nelson, 1980). Potassium helps the plant to adjust to low soil water potential under drought stress (Bukhsh *et al*., 2012). For example, the contribution of K+ to changes of the osmotic potential in wheat caused by drought was in the range of 40–80% (Morgan, 1992). Drought is one of the most detrimental abiotic factors causing serious plant

injuries and alteration in growth and development. Potassium (K) as a macro nutrient plays a vital role in mitigating drought stress by increasing nutrient translocation and improving water balance in plants. Therefore, we investigated the ameliorative effect of K on water stress induced oxidative damage in wheat by excess production of reactive oxygen species and methylglyoxal, which resulted in high lipid peroxidation and disordered antioxidant defense system. Moreover, stunted growth, dehydration and chlorosis were evident at different extent under drought. Potassium supplementation resulted in increased antioxidant defense and upregulation of glyoxalase system with special notation in, osmolyte synthesis and maintenance of redox balance.

Considering these strategies the present study was undertaken keeping in mind the following objectives:

1. To study the effect of drought on growth, physiological and biochemical traits of wheat
2. To investigate the role of potassium and its reflection in alleviating drought stress in wheat
3. To study the underlying mechanism of potassium induced drought stress in improving plant growth, physiological and biochemical parameters.

CHAPTER 2

REVIEW OF LITERATURE

2.1 Wheat

The genus *Triticum* is composed of a number of wheat species among which about 95% of the world wheat produced from the species *Triticum aestivum* popularly known as bread wheat. Among other cereals, wheat is the most widely grown cereal with the highest monetary yield. Wheat belongs to the grass family poaceae is probably the first domesticated cereal crop which is now recognized worldwide as the most important cereal based on grain acreage and is ranked third position considering the global volume of production.

2.2 Global wheat scenario

Around 17 percent of worldwide arable land occupied with wheat cultivation and nearly 35 percent of world population consumes wheat beside rice (FAOStat, 2013). Currently, about 65 percent of the wheat crop is used for food, 17 percent for animal feed, and 12 percent in industrial applications, including biofuels (FAOStat, 2018). Wheat largely grown in cold region of the world as it was originated from temperate region. However, cultivars adaptable to diverse climatic conditions including subtropical regions are successfully developed by the scientists through modern and conventional breeding techniques with high potential yield. The global wheat production came to about 755 million metric tons in crop year 2016-2017 (Statista, 2017) while the total wheat production in 2016 was around 724 million metric tons, down 1.4% or 10 million tons, from the 2015 record (FAO, 2016). However, Global wheat production in 2017 has been forecast to 752.8 million tons (FAO, 2017). In 2015/2016, the global production volume of wheat amounted to approximately 735.3 million metric

tons. In that year, European Union was the leading global wheat producer. In 2016/2017, among the 10 leading global wheat producing countries European Union remain the same in position by producing around 145.7 million metric tons of wheat. The rest of the leading wheat producer is China, India, Russia, USA, Australia, Canada, Ukraine, Pakistan and Turkey (Statista, 2018). According to USDA (2018), the world wheat production was 750.44 million tons in 2016. Among which EU alone accounted for 19.36% production, whereas Asia shares 33.08%, former soviet union, south America, Middle east, Africa, produced 20.73%, 7.50%, 5.76%, 2.23% accordingly (USDA, 2018).

2.3 Wheat production and consumption dynamics: Bangladesh

Although Bangladesh is the world's fourth largest rice-producing and the largest rice- consuming country, Wheat consumption is increasing here over the decades. Wheat consumption has doubled from 1961 to 2013 in Bangladesh and now stands at 17.5 kg per capita, which is nearly a ninth of the rice consumption and 12% of total cereal consumption. Whereas, global wheat consumption has increased by 19 per cent from 54.9 kg yearly per capita in 1961 to 65.43 kg in 2013. Bangladesh has achieved self sufficiency in rice production, but still to go a long way to boost up domestic wheat production (Mottaleb *et al*., 2017). While Bangladesh has been highly successful in achieving self-sufficiency in rice production (Mottaleb and Krupnik, 2015), the increasing wheat consumption is met mainly by import. Bangladesh meets 75% of its wheat consumption needs through imports, sourcing lower quality wheat from Russia and Ukraine, and higher quality wheat from Canada, Australia and the United States. The inflow of wheat food aid to Bangladesh has been decreasing since the 1990s. In 2015–2016, it was estimated that the domestic production of wheat in the country would be 1.40 MT, and from July 2015 to February 2016, the country imported an additional 2.58 MT from abroad to meet consumption demands (GOB, 2016). According to trade sources, during the current MY 2016/17 about 40% of wheat imported is sourced from Russia and 30% from Ukraine (USDA, 2017).

Currently, domestic production can meet only less than 35% of the total wheat consumption in Bangladesh.

Wheat cultivation in Bangladesh was sporadic before liberation.0.126 million hectares area was under wheat cultivation with 0.103 million metric tons production in 1971 (Hasan, 2006). In 1975, the government imported high-yielding wheat seed from India and Mexico to expand wheat production in the country (Ahmed and Meisner, 1996). Introduction to modern agricultural practices, stress tolerant and high yielding varieties, lower production cost compared to others crops facilitates increasing wheat growing area and production over time. In 2016, 1.422 milion metric tons wheat produced from 0.429 milion hectares of cultivated land with 3.32 metric t/ha of average yield (DAE, 2018). While in 2015-2016 it was 1.348 million metric tons from 0.445 million hectares of cultivated land with an average 3.03 metric t/ha (DAE, 2018).

2.4 Abiotic stress

Abiotic and biotic factors immensely hamper crop growth and yield. Among abiotic stress, drought, salinity, heat extreme, nutrient imbalance, metal toxicity is so common. In Bangladesh, winter crop frequently experience scarcity of water, as a result drought is a common phenomenon at that time. Although wheat is moderately drought tolerant, but severe drought results in great yield loss (Ali *et al*., 2013). Water scarcity at spike forming stage results in reduced grain number per spike, per plant yield and finally yield loss occurred (Ali *et al.*, 2013). Due to salinity, the Southern region of Bangladesh is facing huge problem in wheat production and is almost no wheat production in this region due to lack of salt tolerant variety. At times, late sowing of wheat experience severe heat stress. Reduced above-ground biomass is attributed due to heat stress. While demand is increasing day by day, Wheat production is limiting due to these environmental stress factors. There is scope and urgent needs to develop resistant varieties to counter these environmental stresses. Diseases resistant and heat sensitive

wheat varieties like BARI Gom-27, BARI Gom 28, BARI Gom-29 and BARI Gom-30 are released by Bangladesh Agricultural Research Institute (BARI) but yet no drought tolerant, salt tolerant and other stress tolerant wheat varieties are released. However, Fellow lands need to bring under cultivation to gear up the national wheat production according to its ever increasing diverse demand.

The current world population of 7.3 billion is projected to reach 8.5 billion by 2030, 9.7 billion in 2050 (Hasanuzzaman *et al*., 2017). Although population is increasing, the crop productivity is not parallel to the amount of food demand it creating. World agriculture has to produce 70% more food for an additional 2.4 billion people while fighting with poverty and hunger, consuming scarce natural resources more efficiently and adapting to climate change (Hasanuzzaman *et al*., 2014a). In nature plants are frequently exposed to different stress factor(s) due to the incalculable nature of the environment and global climate change (Mittler and Blumwald, 2010). Abiotic stress is the collective term of all the negative impact exerted by environment on living organisms (Hasanuzzaman *et al.*, 2012a). The major abiotic stresses include salinity, drought, extreme temperature, flooding, toxic metal/metalloids, ozone, UV radiation, high light, etc. Some environmental factors, such as air temperature, can become stressful in just a few minutes; others, such as soil water content, may take days to weeks, and factors such as mineral deficiencies can take months to become stressful (Taiz and Zeiger, 2006).

The reason behind the lower productivity of crop plants in most of the cases, are the abiotic stresses (Shanker and Venkateswarlu, 2011). High concentrations of toxic or antagonistic substances are sources of plant abiotic stress where scanty supply of water (drought) or too much flooding can both impose stress on plants. Tremendous plant metabolic changes results from stress, imparts multifarious harmful effects on crop growth, development and productivity. When stress becomes too high and/or prolong 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.*, 2012a, b).

According to Araus *et al.* (2002), abiotic stresses along with limiting crop productivity can also influence the distribution of plant species in different types of environment. Ahmad and Prasad (2012) reported that abiotic stress cause changes in soil-plant-atmosphere continuum which is responsible for reduced yield in several of the major crops in different parts of the world. Upto 50% worldwide yield loss can be possible through abiotic stresses (Bray *et al.*, 2000). Mantri *et al.* (2012) also reported that the yield of food crops worldwide become reduced severely because of drought, cold, high-salinity and heat which are major abiotic stresses. Abiotic stresses greatly affect plant growth and metabolism and ultimately disturbs plant life cycle (Ahmad and Prasad, 2012). These stresses are associated with production of certain deleterious chemical entities called reactive oxygen species (ROS), which include singlet oxygen ($^1O_2$), hydrogen peroxide (H_2O_2), superoxide radical ($O_2^{\bullet}$), hydroxyl radical ($OH^{\bullet}$), etc. (Choudhury *et al.*, 2013). ROS are highly reactive and may cause cellular damage through oxidation of lipids, proteins, and nucleic acids. Besides damaging, reactive oxygen species (ROS) produce signalling effects at organellar and cellular levels (Apel and Hirt, 2004). Plants can respond and adapt to various stress condition by altering their cellular metabolism and regulating various defense mechanisms (Ghosh *et al.*, 2011). Survival of plants under this stressful condition also depend on their abilities to perceive the stimulus, generate and transmit a signal, and initiate various physiological and biochemical changes (El-Shabrawi *et al*., 2010).

In their review, Macedo (2012) concluded that plant abiotic stress has been a matter of concern for the maintenance of human life on earth and especially for the world economy. The outcome of an environmental challenge highly depends on the delicate balance between ROS production and scavenging by both metabolic and enzymatic antioxidants. Traditional plant breeding approaches to

improve abiotic stress tolerance of crops had limited success due to multigenic nature of stress tolerance. 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.

Collins *et al.* (2008) reported that the tolerance to abiotic stress is multigenic and quantitative in nature and thus a massive challenge exists to understand the key molecular mechanisms for advanced selective breeding purposes. Similarly, Patakas (2012) reported that the understanding abiotic stress responses in plants is difficult due to the complexity, interrelationship, and variability of mechanisms and molecules involved that consist their evaluation as an challenging topic in plant research.

2.5 Abiotic stress-induced oxidative stress

Oxidative stress occurs when the antioxidant capacity of a living system during normal metabolic courses is hampered by the excess generation of free radicals or reactive oxygen species (ROS) (Zima *et al*., 2001). These ROS highly disrupt the cellular homeostasis and are extremely harmful to organisms at high concentrations (Hasanuzzaman *et al*., 2012a, b). Reactive oxygen species produced in response to oxidative stress can cause permanent damage to the cellular apparatus. Excess free radicals and ROS attack biomolecules such as lipids, proteins, enzymes, DNA and RNA that lead to tissue or cellular injury (Khan *et al*., 2010). The enhanced production of ROS during environmental stresses can pose a threat to cells by causing peroxidation of lipids, oxidation of proteins, damage to nucleic acids, enzyme inhibition, activation of programmed cell death (PCD) pathway and ultimately leading to death of the cells (Mishra *et al*., 2011). Antioxidants are free-radical scavengers that provide protection to living organisms from ROS inducing cellular damage. Although almost all organisms possess antioxidant defense and repair systems, these systems are

insufficient to prevent the damage entirely.

According to Asada and Takahashi (1987), ROS are a group of free radicles, reactive molecules, and ions that are derived from O_2. It has been estimated that about 1% of O_2 consumed by plants is diverted to produce ROS in various sub cellular loci such as chloroplasts, mitochondria, depending on their concentration in plants.

Chloroplast is the primary site of ROS production in plant. Insufficient energy dissipation during photosynthesis lead to the formation of a Chl triplet state that transfer its excitation energy onto O_2 to make $^1O_2$ (Logan, 2005). Although mitochondrial ROS production is much lower compared to chloroplasts, mitochondrial ROS are important regulators of a number of cellular processes, including stress adaptation and PCD. In glyoxysomes, acyl-CoA oxidase is the primary enzyme responsible for the generation of H_2O_2. Plasma membrane-bound NADPH oxidases (NADPHox) as well as cell-wall associated peroxidases (POX) are the main sources of $O_2^{\bullet}$ (Mhamdi *et al*., 2010). Additional sources of ROS in plant cells include the detoxifying reactions catalyzed by cytochromes in both cytoplasm and the endoplasmic reticulum.

2.6 Drought stress

Drought is a meteorological term and is generally explained as a period without significant rainfall. It is one of the most detrimental environmental stresses that affects the growth and development of plants. Thus, reduction of crop production by drought stress has been recognized as the most complex and devastating one on a global scale (Pennisi, 2008; Mishra and Singh, 2010), and its frequency is increasing as a consequence of climate change and a growing water crisis (Harb *et al*., 2010; Ceccarelli *et al*., 2010). Due to continuous climate change the extremity of drought stress is increasing day by day and global crop production

will reduce up to 30% within 2025, compared with current yield if drought stress increases in similar trend as like as present.

Generally drought stress occurs when the available water in the soil is reduced and atmospheric conditions cause continuous loss of water by transpiration or evaporation. Due to drought and desertification each year 12 million hectares are lost, where 20 million tons of grain could have been grown (FAO, 2014). In Bangladesh a strong drought can cause greater than 40% damage to broadcast Aus, significant destruction to the T. Aman crop in Kharif season in 2.32 million ha and about 1.2 million ha of land during Robi season. Past droughts have naturally affected about 53% of the population and 47% of the country (Dey *et al*., 2011).

Drought stress severely limits crop productivity and the expansion of crop cultivation worldwide and thus a main barrier for crop production (Jaleel *et al*., 2009, Hasanuzzaman *et al*., 2012a). However, the adverse effects of drought stress on growth and development of crop plants are multidimentional in nature (Hasanuzzaman *et al*., 2012 a, b).

2.7 Oxidative stress in plants under drought

Drought stress primarily reduces plant growth which rely on cell division, cell enlargement, cell differentiation, and involves genetic, physiological, ecological, and morphological events, and their complex interactions. Drought stress seriously inhibits these events which adversely affects a variety of vital physiological and biochemical processes in plants, including stomatal conductance, membrane electron transport, carbon dioxide diffusion, carboxylation efficiency, water-use efficiency, respiration, transpiration, water loss, photosynthesis, and membrane functions. Disruption of these key functions limits growth and developmental processes, and leads to reductions in final crop yield. (Nahar *et al*., 2016)

A principal sign of drought stress at the molecular level is the accelerated production of ROS such as $1O_2$, $O_2^{\bullet}$, H_2O_2, $OH^{\bullet}$ to levels that are often beyond the plant's scavenging capacity. This causes oxidative stress that damages cells and cellular components, disrupts the physiological and biochemical life processes, and even leads to plant death (Faize *et al*., 2011).

The excess production of ROS is common in drought and results from impaired electron transport processes in the chloroplasts and mitochondria. Photorespiration is one of the major causes of ROS production under drought stress which accounts for more than 70% of the total H_2O_2 produced. ROS accumulation and oxidative stress increase under drought stress (Sorkheha *et al*., 2011) and drought induced oxidative stress significantly increases lipid peroxidation (Pandey *et al*., 2010). Plants have endogenous mechanisms for adapting to ROS production and are thought to respond to drought stress by strengthening these defense mechanisms. The mechanism of drought tolerance includes ion homeostasis, biosynthesis of osmolytes, scavenging of harmful radicals, water circulation and coordination of a long distance response system (Reddy *et al*., 2004). Numerous research findings describe the ROS generation and their successive damage effects under drought or water deficit stress.

With the increase of drought stress germination percentage, seedling fresh weight, seedling dry weight, shoot length and root length were significantly decreased whereas proline content increased as compared to control while an experiment was conducted in rapeseed (*Brassica napus* L.) with different levels of drought stresses (-4, -6, -8 and -12 bar) (Razaji *et al*., 2014).

Prasad and Staggenborg (2008) explained that morphological, physiological, and metabolic events of plants are seriously affected by drought stress that entirely reduces the grain yield and quality.

Fang *et al*. (2012) conducted a study to evaluate the performance of *Salix paragplesia* and *Hippophae rhamnoides* under three water regimes (80%, 40% and 20% field water capacity, FC). The result demonstrated that drought stress severely reduced the plant height, basal diameter, leaf number, biomass production. Drought increased the below ground plant parts that renders the increase of root/shoot ratio. However, the C, N and P of both species were diminished due to imposed drought.

Yield of barley has been reduced due to imposed drought at 50% field capacity. Compared to control (100% field capacity) yield reduction in Switir and Tlalit cultivar was 40% and 34% respectively (Thameur *et al*., 2012).

Assefa *et al*. (2010) reported that water stress at the vegetative stage and reproductive stage individually can reduce sorghum yield more than 36% and 55% respectively.

Study conducted by Alam *et al*. (2013) Showed that drought stress reduced tissue water content, leaf RWC and total chlorophyll content in *Brassica* Sp. However, growth reduction and reduced photosynthetic carbon assimilation is resulted from drought stress.

Photosynthetic pigment contents are reduced under water stress which is another vital reason for net photosynthesis reduction under drought stress. Reduced photosynthetic pigment content including chl a, chl b, total chl, and carotenoid under drought stress has been reported in several plant species, such as *Avena* sp., *Triticum* sp., and *Gossypium* sp. (Pandey *et al*., 2012). Similarly, extreme drought stress decreased net photosynthetic rate by 22% and 75%, respectively compared to the control treatment (Xu *et al*., 2009).

In another experiment Xu *et al*. (2010) also reported that the leaf net photosynthetic rate and stomatal conductance (gs) in *Phillyrea angustifolia*

plants were decreased about 90% under drought stress.

Similar results were investigated in *Albizia lebbeck* and *Cassia siamea* seedlings (Saraswathi and Paliwal, 2011), in canola (Din *et al*., 2011), in black gram (Pratap and Sharma, 2010), in bean (*Phaseolus vulgaris*) (Abass and Mohamed, 2011) and in water lettuce (Singh and Pandey, 2011),

Mahmood *et al*. (2012) conducted a study with 5 canola cultivars by imposing water stress at flowering and pod filling stages to observe the comparative drought tolerance of the cultivars. The data exposed significant differences among the various canola genotypes for leaf chlorophyll *a, b* and proline accumulation. The chlorophyll *a & b* content of all the Napus genotypes reduced due to drought stress at both the growth stages.

Pervez *et al*. (2009) conducted an experiment with tomato plants which was subjected to drought stress at different stages of growth. Among the stages viz., control, early drought stress (when first truss has set the fruits), middle stress (when fruits in first truss were fully matured and started changing their color), and late drought stress (when fruits on first truss were ripened fully) germination and emergence percentages were 99.36, 89.66, 91.67, 90 and 91.5, 77.5, 78.5, 81.0 respectively.

Uzilday *et al*. (2012) compared the differences between antioxidant responses to drought in C3 (*Cleome spinosa*) and C4 (*Cleome gynandra*). Both malondialdehyde (MDA), H_2O_2 content was remarkably increased in *Cleome spinosa* as compared to *Cleome gynandra* under drought stress because in *C. spinosa*, antioxidant defence system was inadequate to suppress the increasing ROS production under stress condition.

Al-Ghamdi (2009) reported that drought stress weakens antioxidant system (reduced activities of SOD, APX and CAT and ascorbate contents, AsA) in

Triticum aestivum L. which exhibited high H_2O_2 and oxidized ascorbate levels and leading to enhanced membrane damage during severe drought stress, indicated by the accumulation of malondialdehyde (MDA). Between two cultivars of wheat drought susceptible cultivar exhibited more oxidative stress, compared to drought resistant

ROS cause severe damages to reproductive organ has been reported by many studies. Reproductive organ development is extremely sensitive to drought stress across different crop species (Liu *et al*., 2006). During the anther development in rice, CAT, APX, DHAR enzymes were suppressed severely during the meiosis stage which enhanced ROS production (Nguyen *et al*., 2009).

Drought (–0.2 MPa and –0.4 MPa) affects antioxidant system and oxidative damage in melon seedlings. Significant rise of H_2O_2 level and MDA content was directly correlated to the changes of antioxidant components of melon seedlings. However, oxidative stress increased with the increase in severity of drought stress and the melon cultivar Galia is more tolerant than Kirkagac (Kavas *et al*., 2013).

2.8 Effects of drought stress on plant

Deficit water supply at any growth stage poses detrimental effects on crop growth and development in general but varies depending on the severity of stress and the crop growth stage. Effects of drought on morphological, physiological, and biochemical processes in plants are discussed below.

2.8.1 Effect of drought on plant growth and productivity

Establishment of an early and optimum crop stand is important for harvesting maximum productivity. However, if the crop experiences an early drought,

thereby affecting germination, then the suboptimal plant population is the major cause of low grain yield. Early season drought severely reduces germination and stand establishment principally due to reduced water uptake during the imbibition phase of germination, reduced energy supply, and impaired enzyme activities (Taiz and Zeiger, 2010). Growth is an irreversible increase in volume, size, or weight, which includes the phases of cell division, cell elongation, and differentiation. Both cell division and cell enlargement are affected under drought owing to impaired enzyme activities, loss of turgor, and decreased energy supply (Farooq *et al*., 2009). For example, drought decreases growth and productivity of sunflower (*Heliantus annuus* L.) owing to reductions in leaf water potential, rate of cell division, and enlargement primarily due to loss of turgor (Hussain *et al*., 2009).

Under drought, reduced dry matter accumulation occurs in all plant organs, although different organs manifest varying degrees of reduction. For instance, total growth duration of both bread wheat (*Triticum aestivum* L.) and barley (*Hordeum vulgare* L.) decreased under drought (McMaster and Wilhelm, 2003), which generally results in substantial yield reductions. The effect of drought is phase specific in most cases. For example, drought at pre-anthesis delayed flowering in quinoa (*Chenopodium quinoa* Wild.) and bread wheat plants (Geerts *et al*., 2008). Likewise, drought at anthesis commonly delays flowering in rice (*Oryza sativa* L.). In soybean (*Glycine max* L.), drought during grain filling hastened maturity but yield was down due to smaller grains (Desclaux and Roumet, 1996). Different crops respond to drought differently. For instance, upon exposure to drought flowering is delayed in quinoa, whereas in soybean, wheat, and barley drought hastened flowering and physiological maturity.

2.8.2 Effect of drought on vegetative and reproductive stage of plant

While drought occurs during the vegetative period of crop growth, it may substantially decrease economic yield. Drought stress during reproductive and

grain filling phases is more devastating (Table 1; Lafitte *et al*., 2007). Drought at flowering is critical as it can increase pollen sterility resulting in hampered grain set. In sunflower, for example, under drought at flowering, achene yield declined primarily due to less achenes (Hussain *et al*., 2008). In pearl millet (*Pennisetum glaucum* L. Leeke), drought at flowering increased the rate of ear abortion due to a decline in assimilate supply to developing ears (Yadav *et al*., 2004). In drought-stressed maize, kernel set was lost leading to low grain yield. Likewise, water deficit at anthesis increased pod abortion which reduced yield in soybean (Liu *et al*., 2003).

Table 1. Decrease in grain yield in different crops by drought stress

Sl. No.	**Growth stages**	**Crop**	**Stress type**	**Yield reduction (%)**	**Reference**
1.	Reproductive	Rice	Mild stress	54	Lafitte *et al*. (2007)
2.	Reproductive	Rice	Severe stress	94	Lafitte *et al*. (2007)
3.	Flowering	Rice	Short severe stress	54	Lanceras *et al*. (2004)
4.	Flowering and grain filling	Rice	Prolonged severe stress	84	Lanceras *et al*. (2004)
5.	Flowering and grain filling	Rice	Prolonged mild stress	52	Lanceras *et al*. (2004)
6.	Pre-anthesis	Wheat	Prolonged mild stress	18-53	Majid *et al*. (2007)
7.	Post-anthesis	Wheat	Prolonged mild stress	13-38	Majid *et al*. (2007)

Sl. No.	Growth stages	Crop	Stress type	Yield reduction (%)	Reference
8.	Flowering and grain filling	Wheat	Prolonged mild stress	58-92	Dhanda and Sethi (2002)
9.	Stem elongation	Wheat	Mild stress	18	Akram (2011)
10.	Anthesis	Wheat	Mild stress	8	Akram (2011)

2.8.3 Effect of drought on plant water relations

Relative water contents (RWC), leaf water potential, osmotic potential, pressure potential, and transpiration rate are the major attributes of plant water relations (Kirkham, 2005), which are significantly affected under water deficit owing to decrease in water supply. Drought lowered RWC in tomato *(Solanum lycopersicum* L.) and caper bush *(Capparis spinosa* L.) (Ozkur *et al*., 2009). Water potential significantly declined in soybean roots, leaves, and pods under drought in general; however, root water potential dropped much earlier than leaves and pods (Liu *et al*., 2004). Effects of drought also depend on the intensity and duration of drought. Tissue water contents decreased linearly with increased severity of drought (Reddy *et al*., 2004). Transpiration not only helps to maintain leaf temperature but also drives water and nutrient uptake and CO_2 influx. Rise in leaf temperature of bread wheat and rice plants is reported under drought owing to reduced transpiration rates (Siddique *et al*., 2001). de Campos *et al*. (2011) reported reduced turgor pressure and transpiration rate in citrumelo *(Citrus trifoliata* L.) rootstocks grown under drought. Dry matter produced per unit of water consumed is termed WUE. WUE of genotypes and crops varies under drought. Abbate *et al*. (2004) and Subramanian *et al*. (2006) reported higher WUE in wheat and tomato under drought than well- watered controls mainly due to reduced transpiration rates under drought. Crop stage is also

important in defining the effect of drought on WUE. For instance, drought stress decreased WUE in sunflower; however, the extent of the reduction was significantly higher when stress was imposed at flowering than at budding (Hussain *et al*., 2009).

2.8.4 Effect of drought on photosynthesis

A major effect of drought is reduction in photosynthesis, which arises by a decrease in leaf expansion, impaired photosynthetic machinery, premature leaf senescence and associated reduction in food production (Wahid and Rasul, 2005). When stomatal and non-stomatal limitations to photosynthesis are compared, the former can be quite small. This implies that other processes besides CO_2 uptake are being damaged. The role of drought-induced stomatal closure, which limits CO_2 uptake by leaves, is very important. During the last decade, stomatal closure was generally accepted to be the main determinant for decreased photosynthesis under mild to moderate drought (Yokota *et al*., 2002). When the amount of available soil water is moderately or severely limiting, the first option for plants is to close stomata (Cornic and Massacci, 1996). This decreases the inflow of CO_2 into the leaves and spares more electrons for the formation of active oxygen species. As the rate of transpiration decreases, the amount of heat that can be dissipated increases (Yokota *et al*., 2002). Comparing results from different studies is complex due to interspecific differences in the response of stomatal conductance and photosynthesis to leaf water potential and/or relative water content; the parameters most often used to assess the degree of. It is clear that stomata close progressively as drought progresses, followed by a parallel decline in net photosynthesis. However, stomatal conductance is not controlled by soil water availability alone, but by a complex interaction of intrinsic and extrinsic factors.

2.8.5 Effect of drought on mineral uptake and assimilation

Nutrients used for plant growth and biomass production generally come from the internal cycling of reserve materials, which require water for their solubilization and translocation. Limited nutrient uptake is a general phenomenon in crop plants grown under water deficit. Subramanian *et al.* (2006) reported reduced nitrogen (N) and phosphorous (P) contents in roots and shoots of tomato seedlings grown under drought. In marigold seedlings, P content under drought was severely reduced. Nutrient absorption is governed by interactions at the soil-root interface, including (1) root morphology and growth rate, (2) nutrient absorption kinetics of the roots; and (3) soil nutrient supply. Decreased soil water availability affects the rate of diffusion in many plant nutrients and finally the composition and concentration of soil solution (Singh and Pandey, 2011). With limited water supply, nutrient uptake by roots decreases because a decline in soil-water potential slows the diffusion rate of nutrients between the soil matrix and root surface (Farooq *et al*., 2009). Lower transpiration rate and impaired active transport, due to a lack of energy input and altered membrane permeability, decreases root nutrient adsorbing power of crop plants under drought. Impaired enzyme activity involved in nutrient assimilation under drought stress also disturbs nutrient acquisition. The activity of nitrate reductase in leaves and nodules of common bean *(Phaseolus vulgaris* L.) and dhainicha *(Sesbania aculeata* L.) is substantially decreased under drought (Ashraf and Bashir, 2003).

2.9 Drought resistance mechanism in plant

Plants adaptation and survival under drought stress happens through various morphological, biochemical and physiological changes. Drought tolerance is the ability to grow and display economic yield under suboptimal water supply. Drought stress affects the water relations of plants at cellular, tissue and organ levels, causing specific as well as unspecific reactions, damage and adaptation reactions (Beck *et al*., 2007). To cope with the drought, tolerant plants initiate

defense mechanisms against water deficit (Chaves and Oliveira, 2004). Plant drought tolerance involves changes at whole-plant, tissue, physiological and molecular levels. The morphological drought tolerance mechanism includes drought escape, drought avoidance and phenotypic flexibility where physiological mechanism involves Osmotic adjustment, osmoprotection, antioxidation and a scavenging defense system responsible for drought tolerance (Farooq *et al*., 2009). Available literature revealed that plants develop different mechanisms like reduced growth, metabolic alteration, accumulation of compatible solutes, activation of the antioxidant defense system, and suppression of energy consuming pathways to cope with a water limited environment (Nahar *et al*., 2016).

2.10 Effect of drought stress on growth and yield of wheat

Under drought stress or water deficit condition cell division reduces thus the growth of plant also reduces (Hasanuzzaman *et al*., 2013). Drought stress hampers critical physiological as well as biochemical mechanisms in plants (Hasanuzzaman *et al*., 2012a), which ultimately reduces crop yield (Shahbaz *et al*., 2011).

Generally the growing period of wheat is divided under: vegetative and reproductive stages which are affected by drought stress (Shi *et al*., 2010). Germination is the first step of plant establishment but drought reduces the germination percentage and results poor seedling establishment (Kaya *et al*., 2006). It was shown earlier that drought stress decreases root length (Nouri-Ganbalani *et al*., 2009), relative water content, total chlorophyll content and photosynthesis rate (Abdoli *et al*., 2013). In wheat, under drought stress length and area of flag leaf decreases while the width did not significantly changes (Lonbani and Arzani, 2011). Drought stress inhibits root and shoot growth, increases transpiration rate and reduces CO_2 uptake during photosynthesis. Water shortage at crown root initiation stage of wheat causes 27% yield loss. In

the past few decades drought stress drastically reduced production of wheat in many parts of Asia (IPCC, 2007). Crop yield is reduced by 70-80% due to a drought spell during the reproductive stage (Kulkarni and Deshpande, 2007). Spikelet of wheat became sterile due to water deficit at reproductive stage. Under drought stress number of grain per spike decreased (Chandler and Singh, 2008). Wheat yield reduced by 21.8 % and 40.7 % due to 25 and 50% reduction of water consumption, respectively (Ramezanpoor and Dastfal, 2004). Watering at crown root initiation, tillering, jointing and flowering stage gives good yield (Banker *et al*., 2008). Upto 25% and 46% reduction of wheat grain yield was found if water deficit respectively after anthesis period and stem elongation stage (Keyvan, 2010). Drought condition at tillering stage of wheat (when branching start) decreased yield loss up to 46%. Drought stress during booting stage of wheat reduces up to 21% yield loss (Schneekloth *et al*., 2012). Zhang *et al*. (2006) concluded that drought stress should be avoided at the booting and heading of wheat to reduce yield loss.

2.11 Drought induced oxidative stress in wheat

Under various abiotic stress conditions ROS production increased greatly which hinders almost all feature of plants biochemistry and physiology (Hasanuzzaman *et al*., 2012b). Like other abiotic stresses, drought stress increased the ROS production manifold. Under normal conditions plants maintain a balance in ROS productions.

Upon exposing to stress condition, there have an imbalance in ROS production. ROS production beyond the plant's quenching capability is often defined as a disruption of redox signaling and redox control (Jones, 2006), which can cause oxidative stress by damaging membrane lipids, proteins, photosynthetic pigments, and nucleic acids through oxidation process, and these are considerably increased under drought stress (Faize *et al*., 2011). Drought stress impair plants water uptake. So under drought stress condition plants reduce the

stomatal conductance. In that condition, abscisic acid carries stress signal from root to leaves. When leaves accept the signal, it activates closure of stomata (Cruz de Carvalho, 2008). Plants eventually achieved water saving approach by regulating stomatal opening, which decreases transpiration rate but the entrance of CO_2 also lessen and consequently internal CO_2 concentration decreased. Therefore, the rate of CO_2 reduction by the Calvin cycle becomes slow down which decrease regeneration of NADP+, thus provoking excess reduction of the photosynthetic electron transport chain (Reddy *et al*., 2004 and Hasanuzzaman *et al*., 2013). That means drought stress hinders carbon fixation by reducing the availability of CO_2. Reducing CO_2 availability and inhibiting carbon fixation results chloroplast to excessive excited energy from PS I and enhance the production of different toxic ROS (Gill and Tuteja, 2010 and Hasanuzzaman *et al*., 2013). Impaired electron transport procedures in the chloroplasts and mitochondria of the plant cell generate excess ROS production throughout the period of drought stress. On the other hand, decrease activity in PS II results in a disproportion between the generation and utilization of electrons, resulting in an alteration in yield of quantum. The resulted photochemial modification of the chloroplasts in drought-stressed plants leaves harvested excess light energy in the PS II and produce various active free radicals or ROS like $O_2^{\bullet}$, $1O_2$, H_2O_2 and $OH^{\bullet}$, which are very much hazardous for plants for generating oxidative damages (Hasanuzzaman *et al*., 2013; Apel and Hirt, 2004). ROS are off very active nature and harm to any organism when produced at a high concentration, causing lipid peroxidation, protein oxidation, DNA damage, inhibition of vital enzymes, activating programmed cell death (PCD) pathway, leading to cell (Mishra *et al*., 2011). Presence of ROS as low/ moderate concentration can act as a second messenger by stimulating antioxidant system and defending the plants from injury casued by overproduced ROS. Plants are well equipped with non-enzymatic (AsA, GSH; phenolic compounds; alkaloids; and α-tocopherol non-protein amino acids) and enzymatic (SOD, CAT, APX, GR, MDHAR, DHAR, GPX and GST) antioxidants which give protection against oxidative stress (Hasanuzzaman *et al*., 2012a).

Under various abiotic stress conditions, Methylglyoxal, a cytotoxic compound, can also create oxidative stress by over production of $O_2^{\bullet}$ (Yadav *et al*., 2005). Like other abiotic stresses, drought stress also increased the MG production which ultimately increases the ROS production. Glyoxalase system in living organism is well established always on the go to detoxify MG through two very important enzyme glyoxalase I (Gly I) and glyoxalase II (Gly II).

The antioxidant defense and glyoxalase systems cannot work properly to detoxify ROS and MG that produces under drought stress condition. As a result plants cell death. Different research findings demonstrated that drought stress considerably increases oxidative damage by overproducing of ROS and altering the antioxidant defense system based on drought intensity and plant growing stages.

Tan *et al*. (2008) reported that imposition of 15% Polyethylene glycol (PEG-6000) in wheat seedlings for 24 hours significantly increased oxidative stress by increasing MDA content, $O_2^{\bullet}$ production and decreasing antioxidant enzymes like SOD, CAT etc.

Not only short term water shortage but also long term water shortage can significantly increase oxidative stress. According to Ibrahim (2014), withholding irrigation from late tillering to the early flowering stage of *T. aestivum* L. (Giza 168) for 20 days increased oxidative stress markers MDA, H_2O_2 content 194% and 193% and decreased ROS scavenger enzymes activity(CAT, SOD). He also observed that membrane stability index and root viability decreased by 40 and 58%, respectively.

Farooq *et al. (*2013) reported that maintenance of 35% water-holding capacity in soil decreased the membrane stability index by 23% and increased MDA content

by 37% compared to control wheat plants. They also observed that soluble phenolics and leaf free Pro content increased by 30 and 57%, respectively.

Nahar *et al*. (2016) reported that 15% PEG (PEG-6000) induced drought stress rose the amount of MDA and H_2O_2, and $O_2^{\bullet}$ production rate compared to control mung bean plants. CAT which is well known as a ROS scavenger, also decreased under drought stress condition. Besides these, drought stress also increased cytotoxic MG production by decreasing gly II enzyme activity.

2.12 Antioxidant defense system

Oxidative stress commonly occurs along with drought stress. Antioxidant defense system is one of the drought response mechanisms. Aerobic metabolism which provides energy for plant growth and development is often accompanied by the generation of reactive oxygen species (ROS) as by-products such as $1O_2$, H_2O_2, $O_2^{\bullet}$, and $OH^{\bullet}$. The activity of ROS scavenging enzymes is highly correlated with antioxidant stress defense and abiotic stress tolerance. However, the activities vary with plant cultivar, stress duration and dose. The generation of ROS and increased activity of many antioxidant enzymes during abiotic stress have been reported in different plant studies with several reports indicating that the activity of antioxidant enzymes of tolerant genotypes increased in response to abiotic stress whereas the sensitive species failed to do so (Hasanuzzaman *et al*., 2012a). Under normal circumstances, the intracellular generation and removal of ROS is under dynamic equilibrium. When plants suffer from exposure to drought stress conditions, the dynamic equilibrium is broken and the excessive accumulation of ROS injures cells, and the oxidative deterioration may ultimately lead to cell death (Cruz de Carvalho, 2008). The membrane phospholipids and fatty acids which are sensitive to the over-accumulation of ROS are damaged, resulting in the peroxidation of membrane lipids. Under ROS stress, the spatial configurations of various membrane proteins or enzymes are disturbed, leading to increased membrane permeability and ion leakage,

chlorophyll destruction, metabolism perturbations, and even severe injury or death of plants (Gill and Tuteja, 2010). Plants produce ROS in chloroplasts, peroxisomes, mitochondria, endoplasmic reticulum, plasma membrane, and the cell wall due to the imbalance between the generation and utilization of electrons under drought stress conditions (Mittler, 2002). ROS attack the most sensitive biological macromolecules in plant cells to induce lipid peroxidation, protein carbonylation, and DNA damage, and impair their functions to result in a catastrophic cascade of events (Moller *et al*., 2007). To protect cells against the deleterious effects of excessive ROS, plants have evolved a series of sophisticated enzymatic and non-enzymatic antioxidant defense mechanisms to maintain the homeostasis of the intracellular redox state. The protective enzymes include superoxide dismutase (SOD), catalase (CAT), ascorbate peroxidase (APX), glutathione peroxidase (GPX), glutathione reductase (GR), glutathione S-transferase (GST), dehydroascorbate reductase (DHAR), monodehydroascorbate reductase (MDAR), thioredoxin peroxidase (TPX), alternative oxidase (AOX), peroxiredoxin (PrxR/POD), etc. (Mittler *et al*., 2011). SOD, the H_2O – H_2O cycle, the AsA-GSH cycle, GPX, and CAT cooperatively build up major ROS scavenging pathways (Miyake, 2010). The balances among the SOD, APX, and CAT activities are pivotal to maintaining the H_2O_2 homeostasis in plants. The non-enzymatic antioxidant system is comprised of several reducing substances, such as ascorbic acid (AsA), glutathione (GSH), carotenoids (CAR), a-tocopherol (vitamin E), cytochrome f (Cytf), flavanones, anthocyanins, and so on (Blokhina *et al*., 2003; Gill and Tuteja, 2010). To date, quite a few instructive cases have been reported to achieve enhanced drought tolerance by eliminating the excessive accumulation of ROS in cells or promoting the ROS-mediated signal transduction in plants (Hou *et al*., 2009; Reguera *et al*., 2012). Further characterization of genes controlling the antioxidant defense system may also provide useful candidate genes for improving drought tolerance by transgenic approach.

Hossain and Fujita (2009) also observed short-term enhanced drought tolerance with increased activities of APX, MDHAR, DHAR, GR, GPX, GST and CAT in mungbean seedlings, confirmed by lower levels of H_2O_2 and MDA.

Mohammadkhani and Heidari (2007) observed a positive and strong correlation between antioxidant enzymes and drought stress while investigating the responses of *Zea mays* L. var. 704 (drought-tolerant) and var. 301 (drought sensitive).

Shehab *et al*. (2010) reported an increase in the activity of various antioxidant defense enzymes (SOD, APX, GR and CAT) in rice, representing protective activity to counteract the oxidative injury caused by drought.

Selote and Khanna-Chopra (2010) demonstrated that drought acclimation induces oxidative stress tolerance of wheat seedlings, attributed to a well-coordinated induction of the ROS detoxification system.

2.13 Potassium

Potassium (K) is not only a constituent of the plant structure but it also has a regulatory function in several biochemical processes related to protein synthesis, carbohydrate metabolism, and enzyme activation (Hasanuzzaman *et al*., 2018). Potassium is a mineral element taken up in large amounts by plants and plays an important role in the regulation of water status (Mengel and Kirkby, 2001). Potassium is characterized by high mobility in plants at all levels (within individual cells and tissues as well as in long-distance transport via the xylem and phloem). Potassium is the most abundant cation in the cytoplasm. Its salts make a major contribution to the osmotic potential of cells and tissues in glycophyte species. It is accumulated passively by both the cytosol and vacuole except when extracellular K+ concentrations are very low, in which case it is taken up actively (Taiz and Zeiger, 2006).

The potassium ion is involved in many physiological processes, such as enzyme activation, protein synthesis, photosynthesis, osmoregulation, cell extension, stomatal movement and other processes (Mengel, 2007; Farooq *et al.,* 2009). It is well known that potassium activates various enzymes that may also be activated by other univalent cationic species of similar size and water mantle, such as NH4+, Rb+ and Cs+. These other cationic species, however, play no major role under natural conditions, as their concentration in the tis- sues is low and does not achieve the required activation concentration. A likely function of potassium is in polypeptide synthesis in the ribosome, since this process requires a high K+ concentration. It is not yet clear, however, what particular enzyme or ribosomal site is activated by K+ There is indirect evidence that protein synthesis requires K+ (Mengel, 2007). Water conditions in plants influence the K+ accumulation in leaves and interact with K+ nutritional status in some plant species (Restrepo-Diaz *et al.,* 2008). The stomatal opening mechanism is governed by the K+ concentration (Larcher 2006; Taiz and Zeiger, 2006; Mengel, 2007). The opening and closure of K+ channels are of particular importance to guard cells and this action mechanism is controlled by the reception of red light, which induces stomatal opening. Under mild water stress, plants tend to reduce the stomata aperture (Silva *et al*., 2003) and when water stress becomes severe, the stomata generally close. Mahouachi (2007) found reduced levels of K+ in banana plants under drought conditions. Similar results were found by Restrepo-Diaz *et al*. (2008) in the leaves of water-stressed olive plants, regardless of nutritional status.

It has been demonstrated that water is the main factor determining the availability of mineral nutrients such as K+ in the soil as well as absorption by plants and translocation from the roots to the shoot. However, some studies have shown that higher levels of K fertilization may allow plants such as maize

(Premachandra *et al*., 2008) and potato (Khosravifar *et al*., 2008) to tolerate water stress.

2.14 Role of Potassium in plant in response to abiotic stress

Potassium renders plant tolerance to different biotic and abiotic stress like salinity, drought, waterlogging, high temperature and ion toxicity (Reddy *et al*. 2004). Potassium directly or indirectly engaged in transportation, assimilation and development of storage tissue and thus it influences the plant growth and yield in a positive manner (Cakmak, 2005). A diverge ranges of plant metabolic functions like protein synthesis (Fernando *et al*., 1990), photosynthesis and activation of several enzymes are regulated by potassium (Marschner, 2002). Biotic and abiotic factors are responsible for plants to face different kinds of stresses during its lifecycle. Higer production of reactive oxygen species (ROS) frequently make plants suffer from severe oxidative damage. This ROS's are initially account for cellular impairment and growth under stress condition. Cellular membrane damage and degradation of chlorophyll is a common While K is deficit in plant. Net photosynthetic rate get lowered as a result of reduced stomatal conductance due to potassium deficiency. This physiological barrier under low K conditions is partially connected chloroplast.

inner membrane ATPase that controls the high stromal pH needed for efficient energy conversion from light to chemical energy by pumping protons out of the stroma into the cytosol while allowing K+ flux into the stroma (Berkowitz and Peters, 1993). Optimal application of K increases protein content of plants, carbohydrate content in tubers and grains. vitamin C, and the solid soluble contents in fruits also increased by applying K in an extra amount Ktells upon the quality of the produces due to decreased organic acids, that results in an increased pH (Mpelasoka *et al*., 2003).

The crucial importance of K in promoting the production of photosynthates and their transport to storage organs such as fruits, grains, and tubers and enhancing their conversion into starch, protein, vitamins, and oil is inevitable (Mengel and Kirkby, 2001). Shortage of K results in disregulation of many metabolic processes such as, reduced rate of photosynthesis, translocation and enzyme activities (Marschner, 2002).

2.14.1 Photosynthesis

Potassium plays an imperative role in the photosynthesis process and the subsequent carbohydrate translocation and metabolism, which eventually increase the crop yield and improve the grain quality (Lu *et al*., 2016). Both the leaf number and the leaf size are reduced while the plant is deficient in K. The leaf number and size reduction later hasten the diminished photosynthetic rate per unit leaf area and thus account for an overall reduction in the amount of photosynthetic assimilates available for growth (Pettigrew, 2008). Furthermore, K controls photosynthesis through sunlight interception.

2.14.2 Stomatal regulation

One of the major functions of the stomata is to control plant water loss via transpiration. During drought stress, quick stomatal closure and internal moisture preservation are essential for plant adaptation to drought conditions. Potassium plays a crucial role in turgor regulation within the guard cells during stomatal movement (Marschner, 2012). As stomatal closure is preceded by a rapid release of K+ from the guard cells into the leaf apoplast, it is reasonable to think that stomata would be difficult to remain open under K-deficient conditions. Some studies also stated that K deficiency may induce stomatal closure and inhibit photosynthetic rates in several crop plants (Jin *et al*., 2011). Benlloch 2008, explained that the low plant K status could inhibit water-stress-induced stomatal

closure via ethylene synthesis, and stomatal conductance could be significantly reduced in K+-starved plants after the adding of an ethylene synthesis inhibitor (cobalt). During drought stress, the stomata cannot function properly in K+-deficient plants, resulting in greater water loss. Drought stress did not decrease water use efficiency, whereas it did increase WUE by rapid stomata closing during water deficit. Adequate levels of K nutrition enhanced plant drought resistance, water relations, WUE and plant growth under drought conditions Egilla *et al*. (2005).

2.14.3 Water uptake

Potassium is engaged in nearly all the physiological processes of the plant which require water. These processes include stomatal regulation, the translocation of photoassimilates, enzyme activation, and heliotropic leaf movements. In addition, K assists in water transportation and mineral compound translocation for the entire plant through the xylem. In cases in which the K supply is not at its optimum level, the translocation of mineral compounds such as nitrates (NO_3^-), phosphates (PO_4^{3-}), calcium (Ca^{2+}), magnesium (Mg^{2+}) uptake is reduced (Tuma *et al.* 2004). Optimal K fertilization helped plants to mitigate the effect of the water deficit through better water use efficiency, which was related to the lower leaf ET. Martineau *et al*. (2017) reported the appearance of pronounced leaf rolling under a water deficit condition after K addition, which prevented water losses, and thus, they concluded that adding K to K-deficient soils can help maize to cope with drought and could be a new management option.

CHAPTER 3

MATERIALS AND METHODS

This chapter contains a precise description on experimental design and layout, time and location of the experiment, climatic condition of experimental site, seed or planting materials, plant growing procedure, nutrient and treatment doses, data collection and statistical analysis of the experiment.

3.1 Experimental location

The experiment was performed in the greenhouse of Kagawa University, Kagawa, Japan (134° 07' 60.00" E longitude and 34° 15' 60.00" N latitude) during the time span of April 2017 to October 2017. Complete experiment materials, both technical and biochemical analysis support was facilitated by the Laboratory of Plant stress responses under the Department of Applied Biological Sciences, Kagawa University.

3.2 Microclimate of greenhouse

Inside the greenhouse, a source of continued natural sunlight and ambient photoperiod condition was thoroughly maintained with artificially controlled airflow. The temperature set point was 24ºC at day and night but the actual day temperature varied between 20◦C and 35ºC at midday. On an average 70% relative humidity (RH) was maintained during the entire period of experiment by spraying water over the protective net shades around the experimental area.

3.3 Materials

3.3.1 Plant materials

Wheat (*Triticum aestivum)* cv. BARI Gom-21 (Satabdi) seed was used as plant material in conducting the entire experiment. The seed was collected from Bangladesh Agricultural Research Institute, Joydebpur, Gazipur, Bangladesh.

3.3.2 Plant growth behavior

This variety is high yielding, early in maturity and having good level of tolerance to terminal heat stress. It is semi-dwarf in nature with good tillering habit. The average plant height ranges in between 90-100 cm containing 4-6tiller /plant. The leaves are broad, recurved and light green in color whereas the flag leaves are also broad, half-erect and droopy in nature. The crop duration of the cultivar is around 105 days and yield is 3.6-5 t/ha on an average.

3.3.3 Wagner pot

Empty Wagner pot made of plastic white hay with a size specification of 10, 9, 11 inch; on diameter, bottom diameter, height accordingly; were used for carrying out this experiment. Pots were frequently washed and sundried for two days before pouring with sundried sand.

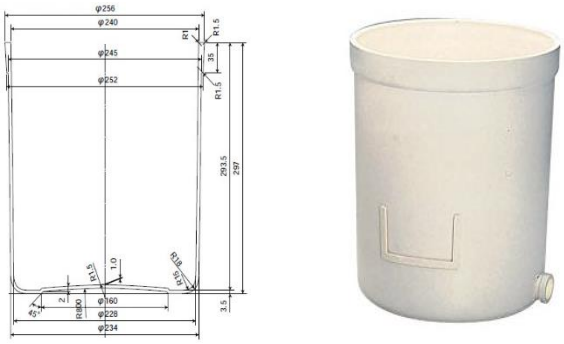

Figure 1. Wagner pot

3.4 Plant growth substrate

Sand was used as plant growth substrate for this experiment. Each Wagner pot was filled with 5kg of sundried sand and were ready for seed sowing.

3.5 Pot preparation

The collected sand was submerged under water in a big sized plastic pot for two days. Later, it was thoroughly washed in bulk for three times under running tape water to remove inert matters and mineral constituents if were present. After that it was sundried for two days in an open floor. Each Wagner pot was then filled up with 5 kg of sundried sand and placed at the greenhouse of Kagawa University, Japan. The pots were pre-labeled for each treatment and finally, water was added to bring soil water level to field capacity.

3.6 Seed sowing

The healthy and mechanical injury free seeds were sorted manually prior to seed soaking. 36 seeds were sown in 6 line maintaining equal line to line and seed to seed distance in each pot. After germination 15 seedlings were remained and rest are uprooted from each pot.

3.7 Plant protection measure

An iron framed structure covered by net were placed over the experimental unit to cut down excessive sunlight also to maintain balanced micro-climatic environment for the plants.

3.8 Design and layout

The experiment was plotted in completely randomized design (CRD) with 03 replications. Nine (9) pots for each replication amounting 27 pots in total along with control for each replication was allocated to complete the experiment.

3.9 Treatment

3.9.1 Protectant treatment

Potassium (K) was used as a protectant in this experiment. Three different concentration viz., 0 mM, 6 mM and 12 mM of K were mixed in Hoagland's nutrient solution (Table 2) considering nil, medium and high dose. 3 days after seed germination nutrient solution was provided for the first time. There after 3 days interval for 2 times water was provided and balanced up to the 100% field capacity level. According this sequence nutrient solution was provided 3 times at 3 DAS, 12 DAS, and 21 DAS.

Table 2. Hoagland's Nutrients Solution composition

Sl No.	Chemicals	Stock g/100 mL	Half Strength Working Solution Stock mL/L	Full Strength Working Solution Stock mL/L
	Macro nutrients			
1	$NH_4H_2PO_4$	11.5	0.5	1
2	KNO_3	10.11	3	6
3	$Ca(NO_3)_2.4H_2O$	16.405	2	4
4	$MgSO_4.7H_2O$	12.035	1	2
5	Micro nutrients			
	5.1 H_3BO_3	0.28	0.5	1
	5.2 $MnCl_2.4H_2O$	0.18		
	5.3 $ZnSO_4.7H_2O$	0.02		
	5.4 $CuSO_4.5H_2O$	0.008		
	5.5 $H_2MoO_4.H_2O$	0.0025		
6	Fe Stock Solution			
	6.1 EDTA.2Na	3.08	0.5	1
	6.2 FeSO4.7H2O	0.78		
	6.3 KOH	0.81		
	6.4 H_2O	100		

3.9.2 Stress treatments

Afterwards the 21 days old seedlings treated with or without protectant were subjected to artificially imposed drought stress by limiting water at two different water regime. Water amount was lowered to 50% and 20% of field capacity and maintained throughout the stress period and the effect was observed under above conditions for 9 days. Control plants were grown in 100% FC condition. Data were taken after 9 days stress period was finished when plants are of 30 day old seedlings. Biochemical assessment was done after this 9 days of stress treatment.

3.9.3 Treatment combinations

- The experiment consisted of eight treatments as follows:
- Well-watered (100% FC) + 0 mM K
- Well-watered + 6 mM K
- Well-watered + 12 mM K
- 50% FC+ 0 mM K
- 50% FC+ 6 mM K
- 50% FC+ 12 mM K
- 20% FC+ 0 mM K
- 20% FC+ 6 mM K
- 20% FC+ 12 mM K

3.10 General observation of the experimental pots

Day and night temperature and humidity were regularly observed and regulated as crop demand. The control plants looked normal green while K deprived plants showed slight yellowing and sporadic tip burn at the leaves.

3.11 Data collection

Growth parameters were collected two times at 10 DAS and 20 DAS where morphological and biochemical analysis of the parameters was recorded after the end of stress treatment period at 30 DAS.

Data were collected on following sequences:

3.11.1 Crop growth parameters

- Mortality rate
- Plant height
- Fresh weight plant^{-1}
- Dry matter weight plant^{-1}

3.11.2 Physiological parameters

- Relative water content (RWC)
- Photosynthetic pigments
- Determination of mineral contents

3.11.3 Oxidative stress indicators

- Lipid peroxidation
- H_2O_2 content
- Proline content
- Methylglyoxal content
- Ascorbic acid content
- Glutathione content
- Protein estimation
- Activities of antioxidant enzymes (APX, CAT, MDHAR, DHAR, GR, GPX, Gly I and Gly II)

3.12 Sampling procedure for growth study during the crop growth period

3.12.1 Plant height

Wheat plant height was recorded two times at equal intervals. First height was recorded at 15 days of seedlings age and second height was recorded at 30 days of seedlings age which was after the end of the stress duration. From the ground level to the highest tip of the leaf was measured by a measuring scale and counted as the plant height. 10 plants from each pot was randomly selected for measuring height. The average height of 10 plants was considered as the height of the plant for each pot.

3.12.2 Fresh weight plant^{-1}

Plant fresh weight was recorded after the end of estimated stress treatment period. Ten plants as sample from each treated pot was uprooted randomly and thoroughly washed in running tape water. After that the samples were weighed in a digital balance and averaged. The average data were counted as fresh weight plant^{-1}.

3.12.3 Dry weight plant^{-1}

After recording the fresh weight, the samples were dried in an electric dryer (ADVANTEK, SP-450, and Japan) at 80^{o}C for 48 h. Then they were weighed in an electric balance and finally averaged to derive the dry weight plant^{-1}. The data were collected after completion of treatment duration.

3.13 Sampling procedure for physiological parameter

3.13.1 Determination of leaf relative water content

According to Barrs and Weatherly (1962) leaf laminas of fully developed leaves were separated from randomly selected plants to measure the leaf relative water content. Leaf discs from randomly chosen plants were taken. At first, laminas were weighed as fresh weight and then immediately floated on distilled water in a petri dish for 8 h and kept in the dark condition of the incubator. Excess surface water was removed with paper towels and leaf turgid weights were obtained by weighing in a digital balance. Dry weights of leaves were measured after drying at 80°C for 48 h. finally, RWC was calculated using the following formula:

RWC (%) = [(FW-DW)/ (TW-DW)] ×100

3.13.2 Determination of photosynthetic pigments

To determine the photosynthetic pigments method of Arnon (1949) was followed. Firstly, 0.5g fresh leaf sample was taken randomly from each treatments. The samples were homogenized with 10 ml of acetone (80% v/v) using pre- cooled pestle and mortar. Later on, the homogenate was centrifuged at 2000×g for 10 min. After diluting the supernatant the absorbance of the supernatants was measured with a UV-visible spectrophotometer at 663, 645 and 470 nm for Chl *a*, chl *b* and carotenoid contents, respectively.

3.13.3 Determination of mineral contents

To determine the Na+ and K+ contents following the method of Rahman *et al*. (2016). Ten seedlings were uprooted from each treated pot after estimated stress period was finished. Samples were thoroughly washed with tap water and thereafter with distilled water to remove surface contaminants like soil, sand, dust and inert materials. The samples were then separated into root and

shoot. Later the samples were placed in an oven (ADVANTEK, SP-450, Japan) for drying at 80°C for 48 h. To determine mineral content 0.1 g dry sample were taken in a test tube and digested with 5 mL acid mixture [HNO_3:$HClO_4$ (5:1 v/v)] at 70^0C in a digestion chamber for 48h. During this operation Opening of test tube were closed by cotton to avoid volatilization. After that, 200 µL digested sample were taken in a test tube and volume up to 5 mL by 10% HNO_3. Finally the absorbance was observed in atomic absorption spectrophotometer by flame method and compare with the respective series of standards.

3.14 Sampling procedure of measuring oxidative stress indicators

3.14.1 Determination of lipid peroxidation

The level of lipid peroxidation was measured by estimating malondealdehyde (MDA) content according to Heath and Packer (1968) with slight modification by Hasanuzzaman *et al.* (2012b). The randomly sampled (0.5 g) leaves were homogenized by 3 mL 5% (w/v) trichloroacetic acid (TCA). The homogenate was then centrifuged at 11,500 × g for 15 min. thereafter, 1 mL supernatant was mixed with 4 mL of thiobarbituric acid (TBA) reagent (0.5% of TBA in 20% TCA). The reaction mixture was heated at 95 °C for 30 min in a water bath and then quickly cooled in an ice bath and centrifuged again at 11,500 × g for 10 min. The absorbance of the colored supernatant was measured at 532 nm and was corrected for non-specific absorbance at 600 nm. MDA content was calculated by using extinction coefficient 155 $mM^{-1}cm^{-1}$ and expressed as nmol g^{-1} FW.

3.14.2 Determination of H_2O_2 content

H_2O_2 was assayed according to the method described by Yu *et al.* (2003). 1/2 g of leaf samples was homogenized with 3 ml of 50 mM potassium-phosphate (K-P) buffer (pH 6.5) at 4°C. The homogenate was centrifuged at 11,500× g for 15

min. 3 ml of supernatant was mixed with 1 ml of 0.1% $TiCl_4$ in 20% H_2SO_4 (v/v) and kept in room temperature for 10 min. After that, the mixture was again centrifuged at 11,500× g for 12 min. The optical absorption of the supernatant was measured spectrophotometrically at 410 nm to determine the H_2O_2 content using extinction coefficient 0.28 $\mu M^{-1}cm^{-1}$ and expressed as $nmolg^{-1}$ fresh weight.

3.14.3 Determination of proline content

Free proline in leaf tissues was measured according to the protocol of Bates *et al*. (1973). Fresh leaf tissue (0.25 g) was homogenized well in 5 ml of 3% sulfo-salicylic acid with pre-cooled mortar and pestle on ice. After homogenization the homogenate was centrifuged at 11,500×g for 15 min. 1 ml of the supernatant was than mixed with 1 ml of acid ninhydrin (1.25 g ninhydrin in 30 ml glacial acetic acid and 20 ml 6 M phosphoric acid) and 1 ml of glacial acetic acid. The mixture was placed at 100°C in water bath for 60 minutes, and then the reaction was terminated by placing the tube in ice bath for 15 min, after the reaction mixture get cooled, 2 ml of toluene was added with the reaction mixture and thoroughly vortexed for 20-30sec. The upper aqueous layer (toluene layer) was immediately transferred to new test tube and kept in room temperature for next 10 minutes. Finally, the optical density of the chromophore containing toluene was read spectrophotometrically at 520 nm using toluene as a blank. The amount of proline was calculated from the standard curve using laboratory grad Proline.

3.14.4 Determination of methylglyoxal content

Method described by Wild *et al*. (2012) was followed to estimate Methylglyoxal. 0.5g Leaves were homogenized in 5% perchloric acid and then centrifuged at 4 °C for 10 min at 11,000×g. The supernatant was decolorized by adding charcoal. The decolorized supernatant was neutralized by adding a saturated solution of sodium carbonate at room temperature. The neutralized supernatant was used to

estimate MG by adding sodium dihydrogen phosphate and N-acetyl-L-cysteine to a final volume of 1 mL. Formation of the product N-α-acetyl-S-(1-hydroxy-2-oxoprop-1-yl) cysteine was recorded after 10 min at a wavelength of 288 nm, and the MG content was calculated using a standard curve of known concentration.

3.14.5 Extraction for ascorbate and glutathione content

Three mL ice-cold 5% meta-phosphoric acid containing 1 mM ethylene-diaminetetraacetic acid (EDTA) solution was used as extraction buffer to homogenize 0.5 g fresh wheat leaves using a pre-cooled mortar and pestle. The homogenate was centrifuged at 11,500 × g for 12 min at 4 °C. Later the supernatant was collected to analyze for AsA and GSH.

3.14.6 Determination of ascorbate content

Ascorbate content was determined following the method of Huang *et al.* (2005) with some modifications. The supernatant was neutralized with 0.5 M K-P buffer (pH 7.0), and the oxidized fraction was reduced by 0.1 M dithiothretitol. AsA was assayed spectrophotometrically at 265 nm in 100 mM K-P buffer (pH 7.0) with 0.5 units of ascorbate oxidase (AO). A specific standard curve of AsA was used for quantification.

3.14.7 Determination of glutathione content

The GSH pool was assayed according to a previously described method (Yu *et al*., 2003) with modifications as described by Paradiso *et al*. (2008). Aliquots (0.2 mL) of supernatant were neutralized with 0.3 mL of 0.5 M K-P buffer (pH 7.0). Based on enzymatic recycling, GSH is oxidized by 5,5-dithio-bis (2-nitrobenzoic acid) (DTNB) and reduced by nicotinamide adenine dinucleotide

phosphate (NADPH) in the presence of GR, and GSH content was evaluated by the rate of absorption changes at 412 nm of 2-nitro-5-thiobenzoic acid (NTB) generated from the reduction of DTNB. Oxidized glutathione (GSSG) was determined after removing GSH by 2-vinylpyridine derivatization. Standard curves with known concentrations of GSH and GSSG were used. The content of GSH was calculated by subtracting GSSG from total GSH.

3.14.8 Determination of protein

BSA (Bovin Serum Albumin) was used as standard and protein concentration of each sample was measured following the method of Bradford (1976).

3.15 Enzyme extraction and assays

Freshly sampled 0.5 g of wheat leaf tissue was homogenized in 1 ml of 50 mM ice-cold K-P buffer (pH 7.0) containing 100 mMKCl, 1 mM ascorbate, 5 mM β-mercaptoethanol and 10% (w/v) glycerol with the ice-cooled mortar and pestle. The homogenates were centrifuged at 11,500× g for 15 min and the supernatants were used for determination of enzyme activity. 0−4ºC temperature was maintained throughout the whole procedure.

3.15.1 Ascorbate peroxidase (APX, EC: 1.11.1.11)

APX (EC: 1.11.1.11) activity was determined following the method of Nakano and Asada (1981) containing 50 mM K-P buffer (pH 7.0), 0.5 mM AsA, 0.1 mM H_2O_2, 0.1 mM EDTA, and enzyme extract that finally volumed to 700 µl. The reaction was started by the addition of H_2O_2 and the activity was measured by observing the decrease in absorbance at 290 nm for 1 min using an extinction coefficient of 2.8 $mM^{-1}cm^{-1}$.

3.15.2 Catalase (CAT, EC: 1.11.1.6)

CAT (EC: 1.11.1.6) activity was assayed following the method described by Hasanuzzaman *et al*. (2012b). A reaction mixture contained 50 mM K-P buffer (pH 7.0), 15 mM H_2O_2, and enzyme solution in a final volume of 700 µL was used where the decrease in absorbance at 240 nm was observed for 1 min happened due to the defragmentation of H_2O_2. The reaction was initiated with the enzyme extract and activity was calculated using an extinction coefficient of 39.4 $M^{-1}cm^{-1}$.

3.15.3 Monodehydroascorbate reductase (MDHAR, EC: 1.6.5.4)

MDHAR (EC: 1.6.5.4) activity was determined by the method described by Hossain *et al*. (1984). The reaction mixture contained 50 mM Tris-HCl buffer (pH 7.5), 0.2 mM NADPH, 2.5 mM AsA, and 0.5 unit of AO and enzyme solution in a final volume of 700 µl. The reaction was started by the addition of AO. Change in absorbance was recorded at 340 nm for 1 min calculation was conducted with an extinction coefficient of 6.2 $mM^{-1}cm^{-1}$.

3.15.4 Dehydroascorbate reductase (DHAR, EC: 1.8.5.1)

DHAR (EC: 1.8.5.1) activity was determined by the procedure of Nakano and Asada (1981). The reaction buffer contained 50 mM K-P buffer (pH 7.0), 2.5 mM GSH, and 0.1 mM EDTA and 0.1 mM dehydroascorbate (DHA). The reaction was started by adding the sample solution to the reaction buffer solution. The activity was calculated from the change in absorbance at 265 nm for 1 min using extinction coefficient of 14 $mM^{-1}cm^{-1}$.

3.15.5 Glutathione reductase (GR, EC: 1.6.4.2)

GR (EC: 1.6.4.2) activity was measured by the method of Hasanuzzaman *et al*. (2011b). The reaction mixture contained 0.1 M K-P buffer (pH 7.0), 1 mM EDTA, and 1mM GSSG, 0.2 mM NADPH, and enzyme solution in a final volume of 1 ml. The reaction was initiated with GSSG and the decrease in absorbance at 340 nm was recorded for 1 min. The activity was calculated using extinction coefficient 6.2 mM–1cm–1.

3.15.6 Glutathione peroxidase (GPX, EC: 1.11.1.9)

GPX (EC: 1.11.1.9) activity was assayed using the method of Elia *et al.* (2003). The reaction mixture contained 100 mM K-P buffer (pH 7.0), 1 mM EDTA, 1 mM sodium azide (NaN_3), 0.12 mM NADPH, 2 mM GSH, 1 unit GR, 0.6 mM H_2O_2 (as a substrate), and 20 µL of sample solution. NADPH oxidation was recorded at 340 nm for 1 min. The activity was calculated using extinction coefficient 6.62 mM−1cm−1.

3.15.7 Glyoxalase I (Gly I, EC: 4.4.1.5)

Glyoxalase I (EC: 4.4.1.5) activity was recorded following the protocol of Hasanuzzaman *et al.* (2011a). The assay mixture contained 100 mM K-P buffer (pH 7.0), 15 mM magnesium sulphate, 1.7 mM GSH and 3.5 mM MG in a final volume of 700 µL. The reaction was started by the addition of MG. Increase in absorbance was recorded at 240 nm for 1 min. The activity was calculated using the extinction coefficient of 3.37 $mM^{-1}cm^{-1}$.

3.15.8 Glyoxalase II (Gly II, EC: 3.1.2.6)

Glyoxalase II (Gly II; EC: 3.1.2.6) activity was determined according to the method of Principato *et al.* (1987) by monitoring the formation of GSH at 412 nm for 1 min. The reaction mixture contained 100 mM Tris–HCl buffer (pH 7.2), 0.2 mM DTNB and 1 mM S-D-lactoylglutathione (SLG) in a final volume of 1 ml. The reaction was started by the addition of SLG and the activity was calculated using the extinction coefficient of 13.6 $mM^{-1}cm^{-1}$.

3.16 Statistical analysis

Data accumulated from different parameters were subjected to analysis of variance (ANOVA) using the software XLSTAT 2018 (AddinSoft, 2018). Mean separation was done by Fisher's LSD at 5% level of significance.

CHAPTER 4

RESULTS AND DISCUSSION

4.1 Growth parameters of wheat seedlings

4.1.1 Plant height

Significant reduction of plant height was observed in wheat seedlings due to imposed drought stress at two different water regimes compared with control (well watered). However, plant treated with K in stressed plant increased plant height under all three water regimes with an exception in 20% FC. 6 mM K application increased plant height by 15% & 18% under 50% and 20% FC accordingly, respect to their control. However, with 12 mM K application increased plant height 29% as well in 50% FC while in 20% FC it increased only 3% comparing to control (Table 3).

4.1.2 Shoot fresh weight and dry weight

Drought stress sharply decreased shoot FW at 50% and 20% water stress compared to control (well watered) seedlings. Similarly, DW of shoot were also reduced when exposed to drought stress. However, DW and FW of seedlings increased in drought-stressed plant while potassium applied as a protectant. A highest increase in FW was 16% at 50% FC with 12 mM K supply compared to control. Where in 20% FC 12 mM K showed less increase than 6 mM K apply in respect to individual's control. Similar trend of increase and decrease was observed in shoot DW due to K supplementation at 50% and 20% FC (Table 3).

Table 3. Effect of potassium on plant height, fresh weight and dry weight of wheat seedlings under different water regimes.

Treatments	Plant height (cm)	FW (mg seedling^{-1})	DW (mg seedling^{-1})
Well watered -K	32.21±1.66d	1.38±0.07c	0.18±0.010de
Well watered +K	39.88±1.40b	1.50±0.06b	0.22±0.008b
Well watered +2K	42.32±1.63a	1.66±0.04a	0.24±0.006a
50%FC -K	30.43±1.79e	1.23±0.04d	0.17±0.009ef
50%FC +K	35.07±2.14c	1.35±0.04c	0.19±0.011cd
50%FC +2K	39.13±1.61b	1.42±0.06bc	0.19±0.008c
20%FC -K	29.14±2.01f	1.11±0.06e	0.15±0.007g
20%FC +K	34.26±1.93c	1.23±0.06d	0.17±0.005ef
20%FC +2K	29.90±1.42ef	1.21±0.05de	0.16±0.004f
LSD (0.05)	1.08	0.09	0.01
CV (%)	1.80	4.28	3.25

Here, 3 water regimes defined as well watereded, 50% FC and 20% FC while 3 potassium doses were represented as –K, +K and ++K that stands for 0 mM K, 6 mM K and 12 mM K accordingly. Means (±SD) were calculated from three replications for each treatment. Values with different letters are significantly different at P≤0.05 applying the Fisher's LSD test

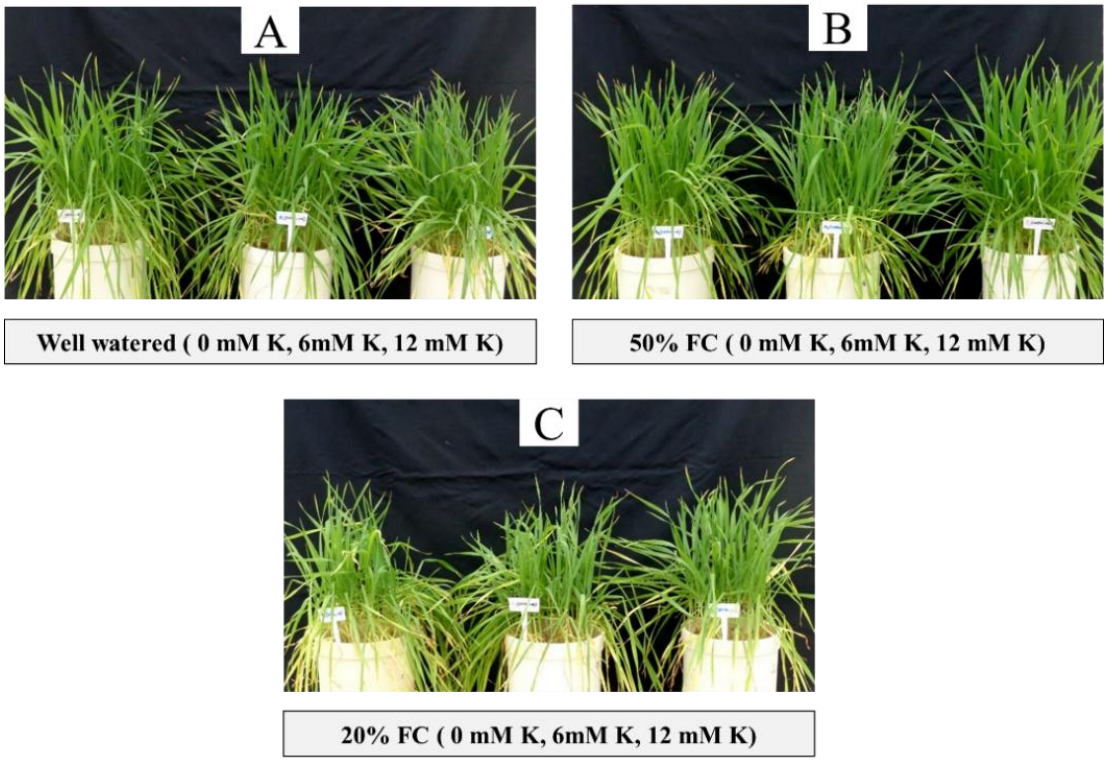

Plate 1. Effect of potassium (0 mM, 6 mM and 12 mM) on plant growth under (A) control, (B) 50% FC and (C) 20% FC condition in wheat seedlings

Cell is basic components of all living organism and water is the basic requirement for cell growth and development. Drought stress hinders cell expansion and reduces stomatal opening and carbohydrate supply ultimately affects growth and development of plants (Kang *et al.*, 2012). Plant growth is characterized by some morphological traits like plant height, Plant FW and DW, shoot length, root length etc. In the following experiment drought affected wheat seedlings exhibited growth retardation in terms of plant height, seedlings FW and DW. Many research publications have been reported the FW and DW of plants in response to drought are reduced significantly (Al Hassan *et al.*, 2017). In some cases due to water deficit condition at early vegetative stage the root biomass of plants increased relative to that of well-water controls which were reported by Sharp and Davies (1979) in maize and Malik *et al.* (1979) in cotton seedlings. However, the present study indicated the improvement of plant growth in terms of seedling length, FW, DW by seed potassium treatment under drought

stress. This results is consistent with the previous findings in which potassium treatment improved growth of plants under different abiotic stress condition (Patade *et al.*, 2009).

4.2 Physiological parameters of wheat seedlings

4.2.1 Relative water content

Drought stress significantly decreased relative water content of wheat seedlings compared to well watered condition. Whereas, supplemented K increased plant RWC in a significant manner. 6 mM K supply increased plant RWC by 21% and 35% under 50% and 20% FC compared to respective controls. However, in 20% FC, 12 mM K showed no significant increase in RWC as compared to control (0 mM K) while in 50% FC it showed 19% increase in RWC due to 12 mM K supply (Figure 2).

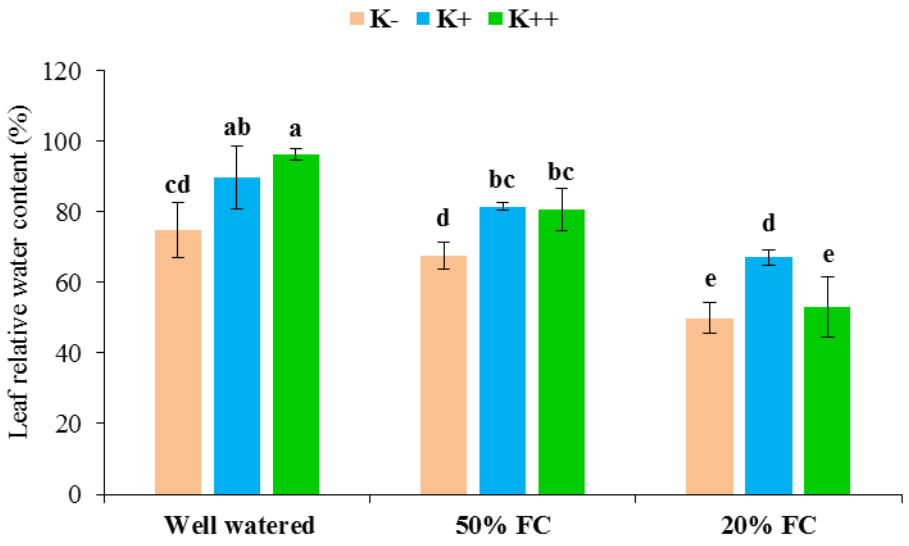

Figure 2. Effect of potassium doses on RWC of wheat seedlings under 3 water regimes. Here the treatments, well watered= 100% field capacity (control), 50% FC, 20% FC and -K, +K and +2K indicate 0 mM potassium, 6 mM potassium and 12 mM potassium respectively. ($LSD_{0.05}$ = 10.47)

Water is the most important component for the survival of all living organism. Under drought stress condition, water lost from the cell of plants that result in decreased RWC. Relative water content of plants is an indicator to measure the relative tolerance levels of plants under stress conditions. Drought reduces the soil water potential surrounding the root zone of plants, therefore water uptake from soil through roots was interrupted to maintain the turgor pressure (Pandey and Shukla, 2015). Leaf RWC is inversely correlated with drought stress (Hasanuzzaman *et al.*, 2013). For evaluating plants for tolerance to drought stress, leaf RWC is considered as an effective parameter. Potassium treatment helped in balancing and increasing RWC in the leaf tissue under drought stress condition. Similar results also observed by Atreya *et al.* (2009).

4.2.2 Chlorophyll content

Chlorophyll *a* and chl *b* contents of leaves were significantly decreased in wheat seedlings under drought stress compared to control, which in terms contributed to reduction of total chl (*a*+*b*) content also. However, seedlings exposed to drought stress with K treatment showed significantly higher amount of chl *a*, chl *b* and chl *(a+b)* content (Table 4). In both chl *a* and chl *b*, at 20% FC plant treated with 12 mM K showed 107% and 66% increase compared to their control (0 mM K). However in chl (a+b), 91% increase was observed with 12 mM K treated at 20% FC than control. Interestingly in chl b, no significant difference was observed at 50% FC either treated with 6 or 12 mM K.

Table 4. Effect of potassium on chl a, chl b and chl (a+b) content of wheat seedlings under control and 2 water regimes (50% FC and 20% FC) condition.

Treatments	Chl a	Chl b	Chl (a+b)
Well watered -K	0.72±0.020c	0.38±0.012c	1.09±0.02d
Well watered +K	0.85±0.076ab	0.47±0.01b	1.32±0.08b
Well watered +2K	0.91±0.020a	0.52±0.04a	1.43±0.06a
50%FC -K	0.49±0.035e	0.25±0.03e	0.74±0.06f
50%FC +K	0.56±0.046d	0.36±0.04c	0.93±0.06e
50%FC +2K	0.79±0.020bc	0.40±0.04c	1.20±0.06c
20%FC -K	0.30±0.025f	0.18±0.02f	0.47±0.04g
20%FC +K	0.43±0.062e	0.29±0.03de	0.72±0.03f
20%FC +2K	0.61±0.035d	0.29±0.03d	0.90±0.05e
LSD (0.05)	0.07	0.04	0.08
CV (%)	6.79	7.33	4.84

Here, 3 water regimes defined as well watereded, 50% FC and 20% FC while 3 potassium doses were represented as –K, +K and ++K that stands for 0 mM K, 6 mM K and 12 mM K accordingly. Means (±SD) were calculated from three replications for each treatment. Values with different letters are significantly different at $P \leq 0.05$ applying the Fisher's LSD test

Plants produce their own food and later converted this food into energy by the process of photosynthesis. Afterall for survival of plants photosynthesis is the most important factor. Water deficit conditions caused a marked suppression in plant photosynthetic efficiency, which has been found to be mainly due to closing of stomata which limits CO_2 diffusion into the leaf, or due to inhibition in rubisco, a non-stomatal factor (Rapacz *et al*., 2010). Water stress also causes a decline in photosynthetic pigments contents including chl, carotenoid, anthocynanin, etc. in various types of crop plants which is due to the oxidation

of pigments, impaired pigment biosynthesis and so on (Abbaspour *et al*., 2011; Pandey *et al*., 2012). Findings of the present study are also evident by reduced chl *a*, chl *b* and total chl *(a+b)* under drought stress. Addition of potassium with drought stress improved the photosynthetic pigment levels in studied species which might be due to the higher biosynthesis of these pigments. Present findings is corroborated with the results of previous studies on other stresses including drought stress (Hasanuzzaman *et al.*, 2014a; Hoque *et al*., 2008).

Effect of water stress (low, mild and severe) and K supply (0.2, 2.0 and 6.0 mM) on net photosynthesis rate in wheat leaves was studied by Gupta *et al*. (1989). Result showed under water stress, the photosynthetic efficiency of plants was reduced drastically (61%) as a consequence of chloroplast dehydration. Also, in drought stress conditions, spraying plants with K application in three levels 0.2, 2.0 and 6.0 mM, photosynthesis rate increased 17.3%, 75% and 92.8%, respectively. In wheat experiments, Pier and Berkowitz (1987) observed 66-113% higher photosynthetic rates in plants fertilized with above normal K+ than those under standard fertilization, indicating that leaves of plants grown in very high internal K+ levels have partially reversed the dehydration effects on photosynthesis.

4.2.3 Root-shoot mineral content

4.2.3.1 Shoot mineral content

Drought stress highly effect on the shoot mineral contents (K, Ca, Mg). In this study, increasing drought severity reduced all three mineral contents significantly. However, potassium supply increased the shoot mineral contents. The maximum shoot K content 26% was increased at 50% FC while plant treated with 12 mM K compared to control. Maximum shoot Ca content was observed at 20% FC while plant was exposed to 12 mM K. on the contrary, at 50% FC both the doses of K reduced Ca content in shoot and there was no significant difference between them. Meanwhile, highest Mg content 69% was accumulated

in 20% FC with 6 mM K supply compared to control (0 mM K) where the minimum 10% was observed at 50% FC with same dose of K application (Table 5).

Table 5. Effect of potassium supply on shoot mineral (K, Ca, Mg) content of wheat seedlings under three water stress condition.

Treatments	**Shoot mineral content**		
	Potassium (K)	**Calcium (Ca)**	**Magnesium (Mg)**
Well watered -K	547.20±32.50b	197.73±16.72b	143.50±6.84b
Wellwatered +K	635.20±12.69a	245.40±12.61a	155.22±10.33b
Well watered +2K	644.60±17.39a	281.25±2.00a	180.93±14.58a
50%FC -K	466.40±35.73c	180.78±9.13bc	143.58±9.37b
50%FC +K	562.80±30.42b	173.59±38.80bc	157.90±13.70b
50%FC +2K	586.95±51.82b	177.00±45.10bc	161.23±12.34ab
20%FC -K	363.38±24.06e	133.52±8.25d	38.35±10.52d
20%FC +K	439.65±18.87cd	146.21±16.89cd	64.96±7.90c
20%FC +2K	395.72±26.61de	162.03±5.66bcd	58.26±12.26cd
LSD (0.05)	4.35	7.96	3.25
CV (%)	5.35	11.70	9.38

Here, 3 water regimes defined as well watereded, 50% FC and 20% FC while 3 potassium doses were represented as –K, +K and ++K that stands for 0 mM K, 6 mM K and 12 mM K accordingly. Means (±SD) were calculated from three replications for each treatment. Values with different letters are significantly different at $P \leq 0.05$ applying the Fisher's LSD test

Wei *et al*. (2013) showed the response of two wheat cultivars in response to drought with potassium application. External K_2CO_3 significantly increased internal K+ contents of both cultivars under well-watered conditions. PEG6000

drastically reduced K+ contents in both shoot and root of two cultivars. Adequate K_2CO_3 application increased K+ content in both shoot and root, with the extent being larger in root than in shoot in both cultivars. The K+ content in shoot peaked at 7.5 mM K_2CO_3 in both cultivars, while the maximum values in roots were noted in 10 mM K $2CO_3$ treatment.

4.2.3.2 Root mineral content

Compared to control, increasing drought severity highly reduced the root mineral contents (K, Ca, and Mg). Due to drought, significant root mineral content reduction was visible in this study. Whereas, potassium supply increased the root mineral contents in different ways. The maximum root K content 46% was increased at 50% FC while plant treated with 12 mM K compared to control. Where in 20% FC with same K treatment it reduced 18% root K content compared to control. Besides, Maximum root Ca content was observed at 50% FC while plant was exposed to 12 mM K. on the contrary, at 20% FC with same K dose shoed the lowest increase of K content in the root. . Meanwhile, highest Mg content 79% was accumulated in 20% FC with 12 mM K supply compared to control (0 mM K) (Table 6).

Table 6. Effect of potassium supply on root mineral (K, Ca, Mg) content of wheat seedlings under three water stress condition.

Treatments	**Root mineral content**		
	Potassium (K)	**Calcium (Ca)**	**Magnesium (Mg)**
Well watered -K	165.99±6.35c	3396.37±166.48c	121.0±9.0bc
Well watered +K	240.70±27.00b	3959.00±189.43b	131.2±5.8ab
Well watered +2K	286.50±30.28a	4434.94±199.44a	143.2±6.4a
50%FC -K	159.33±11.68cd	479.29±69.41f	111.3±5.8c
50%FC +K	164.95±29.70cd	2393.07±199.02d	123.1±14.5bc
50%FC +2K	233.24±15.41b	2486.13±124.57d	131.7±12.2ab
20%FC -K	128.09±21.92de	545.96±80.98f	74.9±11.3d
20%FC +K	154.98±12.67cd	1280.93±72.66e	85.1±12.6d
20%FC +2K	105.18±21.82e	1093.86±44.88e	133.7±15.6ab
LSD (0.05)	3.25	1.21	3.64
CV (%)	11.90	6.66	8.64

Here, 3 water regimes defined as well watered, 50% FC and 20% FC while 3 potassium doses were represented as –K, +K and ++K that stands for 0 mM K, 6 mM K and 12 mM K accordingly. Means (±SD) were calculated from three replications for each treatment. Values with different letters are significantly different at $P \leq 0.05$ applying the Fisher's LSD test

4.3 Biochemical parameters of wheat seedlings

4.3.1 Oxidative stress markers

4.3.1.1 Levels of lipid peroxidation (MDA content)

Drought stress results in excessive production of ROS that interrupts cellular redox homeostasis and ultimately imparts serious oxidative damages to plants. A sharp increase in MDA content (the product of lipid peroxidation) was observed under drought stress which ultimately results in membrane damage. In this study, the highest lipid peroxidation was observed in 20% FC comparing to control. However, potassium treatment significantly reduced MDA content in all three water conditions. At 50% and 20% FC, 26% and 36% MDA content was reduced due to 6 mM K apply in respect to their control (0 mM K), accordingly. Meanwhile, 12 mM K addition reduced MDA content to 41% where in 20% it reduced only 24% from respective controls (Figure 3).

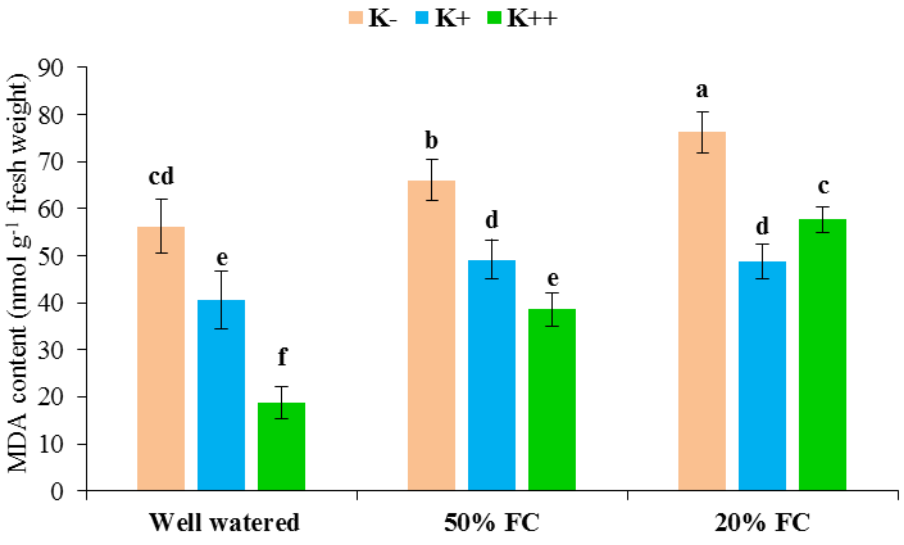

Figure 3. Effect of potassium on MDA content of wheat seedlings under control and 3 water regimes. Here the treatments, well watered= 100% field capacity (control), 50% FC, 20% FC and -K, +K and +2K indicate 0 mM potassium, 6 mM potassium and 12 mM potassium respectively. ($LSD_{0.05}$ =7.36)

MDA is a major cytotoxic compound which is produced through lipid peroxidation. MDA act as an indicator of oxidative damage due to increasing production of ROS (Mittler and Blumwald, 2010). ROS production increases under various abiotic stresses including drought stress. In our study, levels of MDA content increased markedly under drought stress condition where decreased levels of MDA content was observed in seedlings which were supplied with potassium at different concentrations and grown in drought stress condition. Similar protective effect of K was observed by (Fayez and Bazaid, 2013; Yan *et al.*, 2015). Improving membrane repairing and inducing responses of antioxidant enzymes (SOD, APX) helps to decrease the MDA content, which helps to provides protection against oxidative damage.

4.3.1.2 Levels of hydrogen peroxide

Hydrogen peroxide, an oxidative stress marker results in increased production due to various abiotic stresses including drought stress. In present study, under increasing drought stress the levels of H_2O_2 was increased in wheat seedlings compared to control (well watered) condition. However, potassium (K) treatment significantly decreased H_2O_2 content as compared to their respected control in all three water regimes. Notable decrease was found in plants treated with 6 mM K, where 21% and 23% H_2O_2 reduced under 50% FC and 20% FC accordingly, compared to their control (Figure 4). On the contrast, plants treated with 12 mM K reduced H_2O_2 content 27% under 50% FC, whereas only 7% reduced under 20% FC with same K treatment, compared to their respective controls.

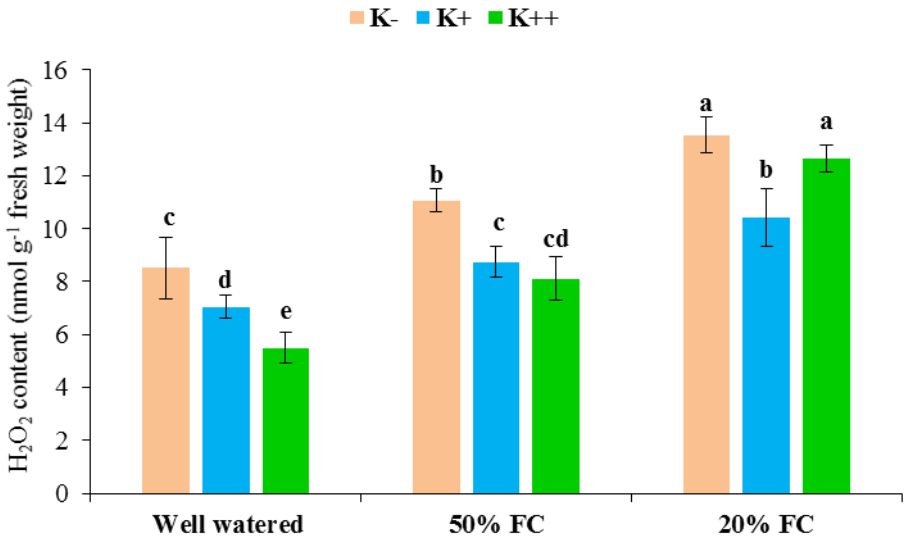

Figure 4. Effect of potassium on H_2O_2 content of wheat seedlings under control and 3 water regimes. Here the treatments, well watered= 100% field capacity (control), 50% FC, 20% FC and -K, +K and +2K indicate 0 mM potassium, 6 mM potassium and 12 mM potassium respectively. ($LSD_{0.05}$ = 1.30)

Hydrogen peroxide is a ROS, which is formed when oxygen acknowledges two electrons. Its metabolism determines formation of exceptionally toxic species; nonetheless, at the physiological concentration it is not dangerous. H_2O_2 converted to cytotoxic $HO^{\bullet}$ by the fenton or Haber–Weiss reactions. H_2O_2 content increased remarkably when plants falls under various abiotic stresses (Tian and Lei, 2007 and Hasanuzzaman *et al.*, 2014b). Higher H_2O_2 concentration may cause oxidative stress. However, during abiotic stresses the accumulation of H_2O_2 increased in plants cell and increased oxidative damage in cell membrane (Nahar *et al*., 2015; Rahman *et al*., 2016). Similarly, Nxele *et al.* (2017) were reported from their study that salinity and drought stress increased the H_2O_2 content significantly in sorghum plant. In the following experiment, drought stress caused a surprising increase of H_2O_2 which is clear sign for oxidative stress. Potassium treatment reduced the generation of H_2O_2 in stressed plants by upregualting the enzymes such as APX, CAT and GPX. This result is well agreed with Alam *et al.* (2013).

4.3.1.3 Proline (Pro) content

Compared to control (well watered) proline content was notably increased in wheat seedlings while exposed to increased drought severity. Plant treated with K as protectant under 50% FC and 20% FC showed significant reduction in proline content. For example, 50% and 20% stressed plant treated with 6 mM K results in declined Pro content by 25% and 35% accordingly, respect to their controls (0 mM K). On the contray, 12 mM K in 50% stress reduced pro content by 51% while only 21% reduction was observed in 20% FC with same K treatment comparing each controls (Figure 5).

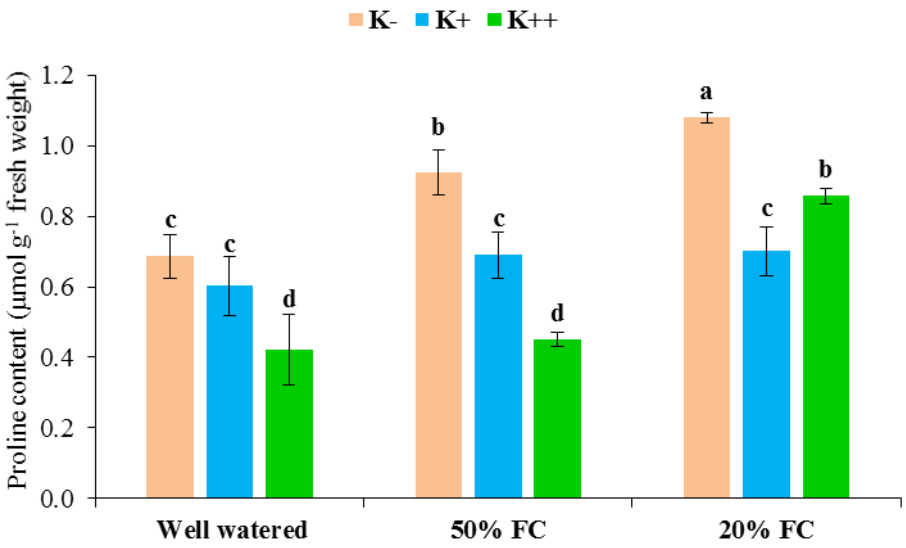

Figure 5. Effect of potassium on proline content of wheat seedlings under control and 3 water regimes. Here the treatments, well watered= 100% field capacity (control), 50% FC, 20% FC and -K, +K and +2K indicate 0 mM potassium, 6 mM potassium and 12 mM potassium respectively. ($LSD_{0.05}$ =0.11)

Proline plays very important role under abiotic stress conditions including drought stress. Proline has vital roles in osmotic adjustment, stress signal transduction and it also acts as an antioxidant. Increase of Pro level under physiological stresses including drought stress conditions were documented

previously (Nahar *et al*., 2013). Similarly profound increases of Pro levels under drought stresses were observed in wheat seedlings. In present study, Potassium treatment with drought stress reduced Pro levels in wheat seedlings. Proline biosynthesis due to potassium treatment under drought stress did not increase the Pro levels further. These results are corroborated to previous studies (Nounjan *et al*., 2012).

4.3.2 Antioxidant defense system

4.3.2.1 Ascorbate content

Ascorbate content was significantly reduced under increasing drought stress condition compared to control. On the contrast, K treatment ameliorated AsA content under drought stress condition. A highest 64% of AsA content increased was observed at 12 mM K treated plant in 20% FC compared to control (0 mM K). Whereas, at 50% FC condition the increasing content of AsA was 20% and 26% with 6 mM and 12 mM K supply respectively, compared to control (Figure 6).

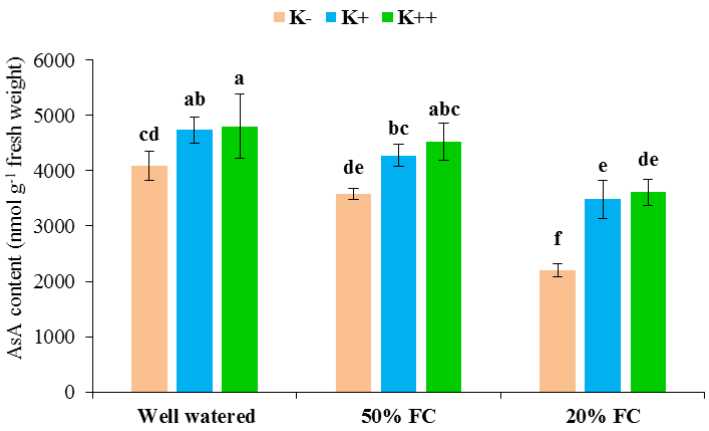

Figure 6. Effect of potassium treatment on ascorbate (AsA) content of wheat seedlings under 3 water regimes. Here the treatments, well watered= 100% field capacity (control), 50% FC, 20% FC and -K, +K and +2K indicate 0 mM potassium, 6 mM potassium and 12 mM potassium respectively. ($LSD_{0.05}$ = 2.12)

As a non-enzymatic antioxidant ascorbate (AsA) is one of the most important antioxidant for plant tissues to maintain the cellular redox state and protecting the plants from oxidative damage (Smirnoff, 2000), thus AsA content acts as a salinity and drought stress tolerance marker in plants (Hasanuzzaman *et al.*, 2011a). Moreover, among all antioxidative pathways the ascorbate-glutathione (AsA-GSH) cycle is one of the most important because in this cycle H_2O_2 the most long lived ROS converted into H_2O by ascorbate peroxidase (APX) along with the generation of monodehydroascorbate (MDHA), where AsA used as an electron donor (Smirnoff, 2000). After that MDHA is converted into AsA or disproportionate into AsA and dehydroascorbate (DHA). In previous study Rahman *et al.* (2016) suggested the AsA content and the AsA/DHA ratio are crucial for the maintaining of wheat seedlings redox homeostasis. Therefore, in this study we investigated the AsA, DHA and AsA/DHA ratio in wheat cultivars seedlings drought.

4.3.2.2 Reduced glutathione (GSH) content

In contrast to the well watereded condition, plant GSH content was drastically reduced under water deficit condition. However, Potassium supplementation increased GSH content under drought stress. Although in 50% FC, 6 mM and 12 mM K increased 24% and 28% GSH content accordingly. But there was no significant difference observed due to K supply at this water level. On the contrary at 20% FC, 11% GSH content increased due to 6 mM K, whereas 12 mM K increased only 1% content that is non-significant compared to control. (Figure 7).

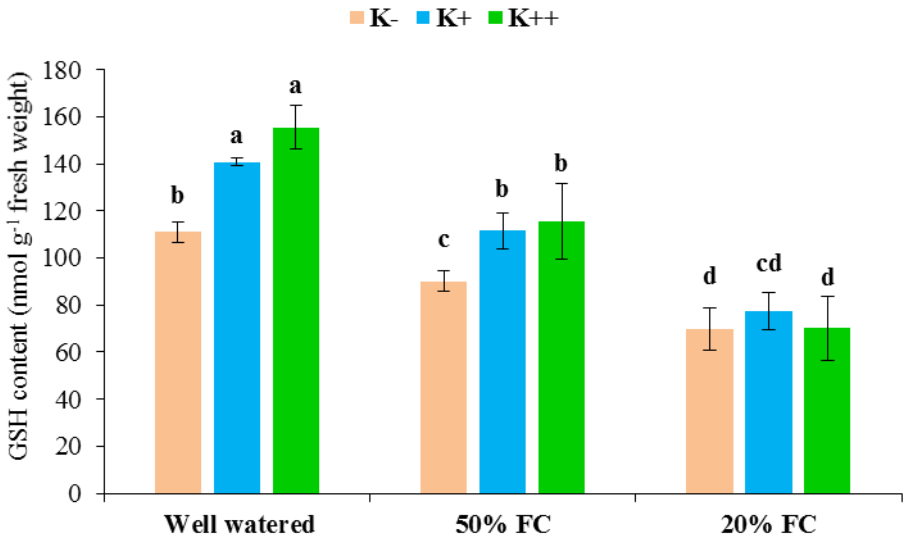

Figure 7. Effect of potassium treatment on glutathione (GSH) content of wheat seedlings under 3 water regimes. Here the treatments, well watered= 100% field capacity (control), 50% FC, 20% FC and -K, +K and +2K indicate 0 mM potassium, 6 mM potassium and 12 mM potassium respectively. ($LSD_{0.05}$ = 7.96)

4.3.2.3 Oxidized glutathione (GSSG) content

GSSG content was immensely increased under deficit condition compared to control (well watered). Oppositely, K treatment decreased GSSG level in drought-stressed seedlings. The highest GSSG content reduction, 36% and 27% was observed at 20% FC with 6 mM K and 12 mM K supply repectively, comparing the control (0 mM K). However, in 50% FC this decrease rate was 12% and 25& due to same K treatment accordingly (Figure 8).

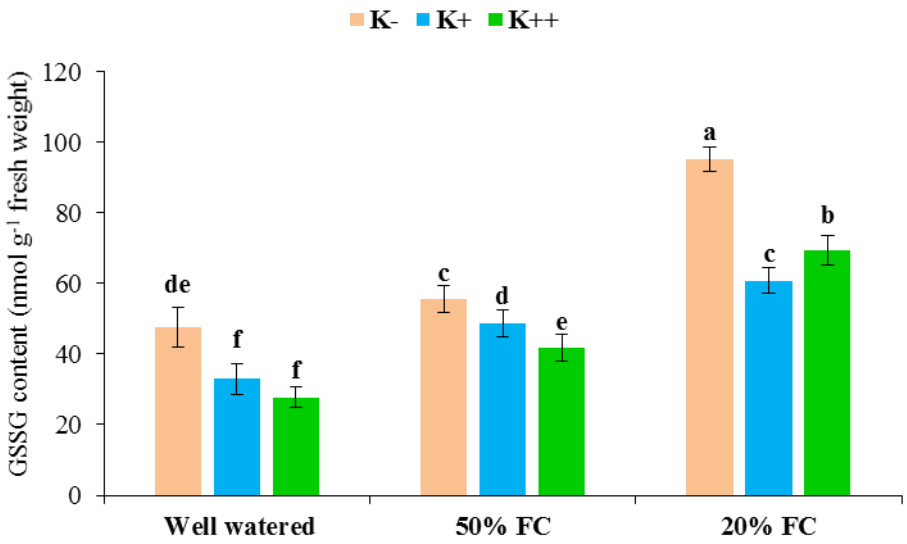

Figure 8. Effect of potassium treatment on oxidized glutathione (GSSG) content of wheat seedlings under 3 water regimes. Here the treatments, well watered= 100% field capacity (control), 50% FC, 20% FC and -K, +K and +2K indicate 0 mM potassium, 6 mM potassium and 12 mM potassium respectively. ($LSD_{0.05}$ =5.76)

4.3.2.4 GSH/ GSSG ratio

The GSH/GSSG ratio was reduced significantly compared to control due to stress severity. However, potassium treatment recovered GSH/GSSG ratio in drought-stressed seedlings. A sharply increased ratio was observed at 50% FC with 6 mM K and 12 mM K supply compared to control. Although, there was no significant difference between them. On the contray, at 20 % FC, the ratio increased by 74% with 6 mM K supply but only 39% with 12 mM K supply (Figure 9).

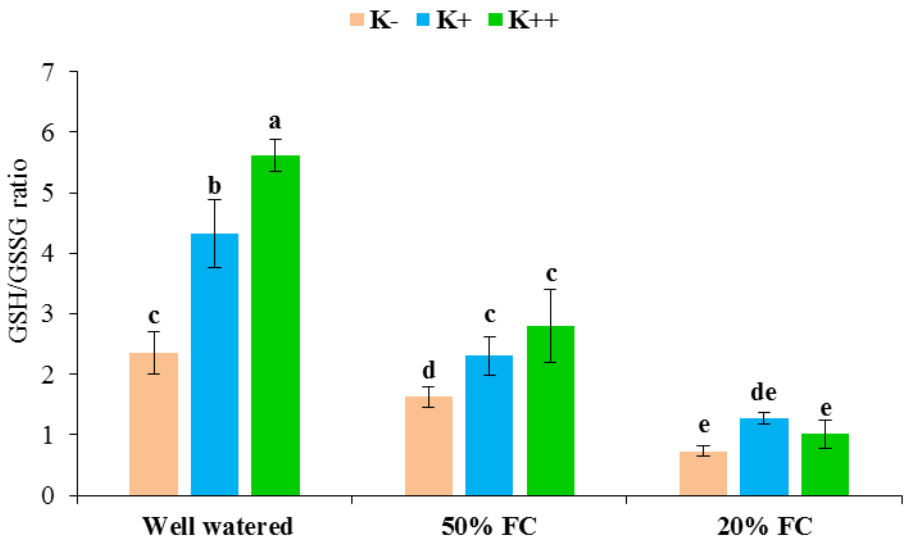

Figure 9. Effect of potassium treatment on GSH/ GSSG ratio of wheat seedlings under drought and control condition. Here the treatments, well watered= 100% field capacity (control), 50% FC, 20% FC and -K, +K and +2K indicate 0 mM potassium, 6 mM potassium and 12 mM potassium respectively. ($LSD_{0.05}$ =0.58)

GSH is another important nonezymatic antioxidant which involves in the reduction of most ROS (Noctor and Foyer, 1998). GSH also plays a key role by regenerating AsA via AsA-GSH cycle. To scavenge ROS, GSH is a substrate for GPX; as GPX is well known ROS scavenger enzyme (Noctor *et al.*, 2002). In this study, the endogenous GSH level increased under drought stress in wheat seedlings which in accordance with earlier studies Nahar *et al.* (2015). However plant treated with potassium further increased GSH levels which might be possible reason for quenching free radicals that's produced under various abiotic stress condition.

Reduced Glutathione is oxidized to GSSG and increased the level of GSSG during the scavenging reaction of ROS (Rahman *et al.*, 2015). The results of this study showed that drought stress markedly increase GSSG content. Similar reports were also found in Alam *et al.* (2013), Nahar *et al.* (2015). Potassium treated seedlings decreased GSSG content under drought stress conditions.

GSH/GSSG ratio has immense functions in cell redox potential and stress signaling processes (Forman *et al.*, 2009). Higher GSH/GSSG ratio is an indicator for higher stress-tolerances. Under drought stress condition, the GSH/GSSG ratio was greatly reduced compared with control. However potassium treated seedlings under drought stress showed increased GSH/GSSG ratio.

4.3.2.5 Ascorbate peroxidase (APX) activity

Increasing drought severity increased the APX activity in wheat seedlings compared to control. APX activity further increased in drought-stressed seedlings with Potassium application. However, the highest APX activity 70% was observed 50% FC with 12 mM K treatment compared to control (0 mM K). Similar increasing trend was followed in 20% FC with 17% and 35% increase in 6 mM and 12 mM K apply respectively (Figure 10).

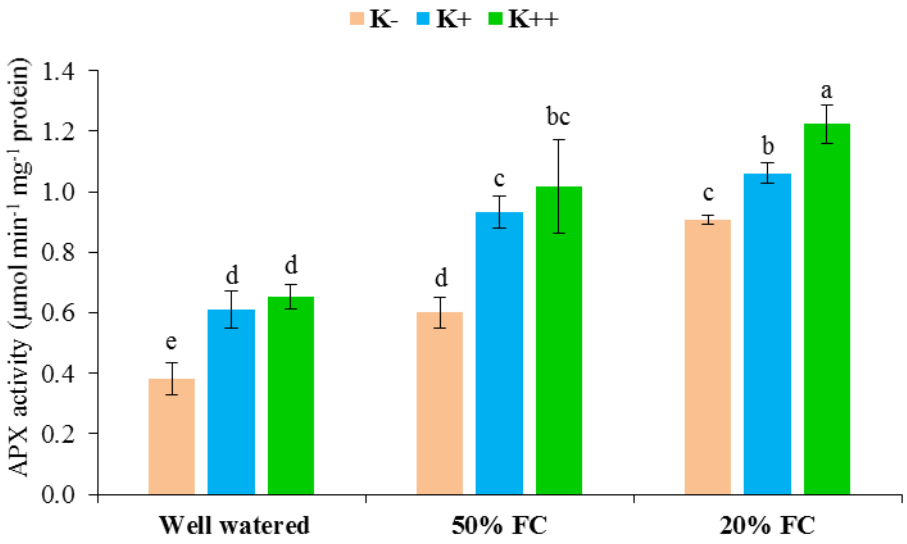

Figure 10. Effect of potassium treatment on APX activity of wheat seedlings under three water regimes. Here the treatments, well watered= 100% field capacity (control), 50% FC, 20% FC and -K, +K and +2K indicate 0 mM potassium, 6 mM potassium and 12 mM potassium respectively. ($LSD_{0.05}$ = 0.11)

APX is one of the most important enzymes of AsA-GSH cycle which converts H_2O_2 into water. APX used AsA as a reducing equivalent during conversion of H_2O_2 into water. In this study, under drought stress APX activity increased compared to control seedlings. Similar trends were also observed by Alam *et al.* (2013) and Nahar *et al.* (2016). APX activity increased under drought stress might be due to higher H_2O_2 and lower AsA content. However APX activity further increased while drought stressed seedlings treated with potassium. These results are supported by Hameed *et al.* (2013), Islam *et al.* (2015).

4.3.2.6 Mono dehydroascorbate reductase (MDHAR) activity

Deficit water condition reduced MDHAR activity in drought stressed seedlings compared to control (well watered) condition. However, drought plant treated with potassium showed increased activity of MDHAR enzyme. Although in 50% FC the enzyme activity was increased by 18% to 34% with 6 mM and 12 mM K supply, accordingly. On the contrary, in 20% FC this trend reduced while plant treated with 12 mM K. in 20% FC, 6 mM K increased MDHAR activity by 30% where 12 mM K increased only 9% activity which was non-significant compared to control (Figure 11).

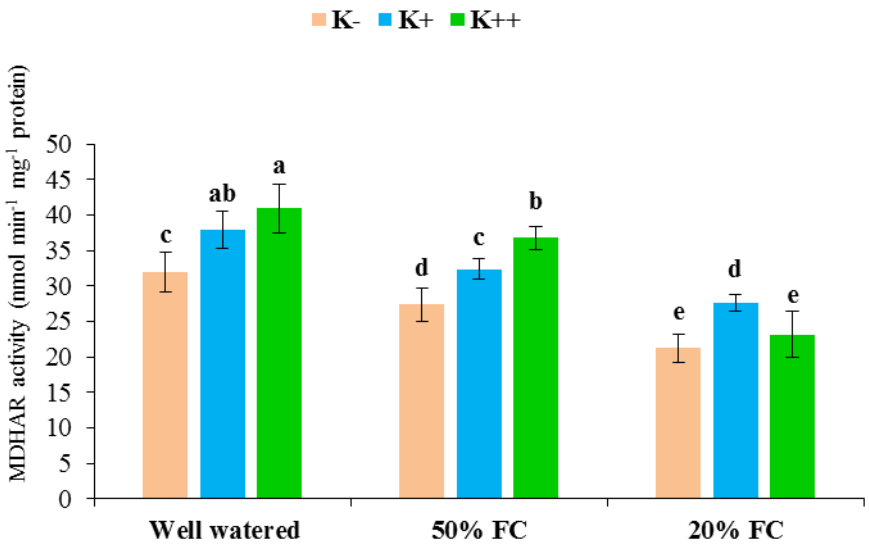

Figure 11. Effect of potassium supply on MDHAR content of wheat seedlings under three water regimes. Here the treatments, well watered= 100% field capacity (control), 50% FC, 20% FC and -K, +K and +2K indicate 0 mM potassium, 6 mM potassium and 12 mM potassium respectively. ($LSD_{0.05}$ = 3.34)

Monodehydroascorbate reductase (MDHAR) is another important enzyme of AsA-GSH cycle. The univalent oxidation of AsA leads to the formation of MDHA. If MDHA is not reduced again to AsA by MDHAR, it will spontaneously disproportionate into AsA and DHA. The regeneration of AsA could be regulated in this cycle mainly by NADPH-dependent MDHAR activity (Mittova *et al.*, 2000) and thus it is crucial for AsA regeneration and essential for maintaining a reduced pool of AsA (Martínez and Araya, 2010) . Although there are also a few reports about MDHAR activity in other physiological processes that are related to oxidative stress, research on different crops under environmental stresses revealed the regulatory role of MDAHR during oxidative stress tolerance and acclimation (Hasanuzzaman *et al.*, 2012a).

4.3.2.7 Dehydroascorbate reductase (DHAR) activity

To adjust the level of AsA and its redox state under stress condition, DHAR is an equally important enzyme as MDHAR (Eltayeb *et al.*, 2007). DHAR activity was notably reduced while plant exposed to drought at different magnitude comparing with control (well watered) condition. Potassium treatment increased DHAR activity in a significant manner. The highest increase was observes at 20% FC with 12 mM K supply but not significantly different from the increase with 6 mM K compared to their control (0 mM K). However at 50% FC, the increase rate was 45% and 55% with 6 and 12 mM K apply, respectively comparing their control (Figure 12).

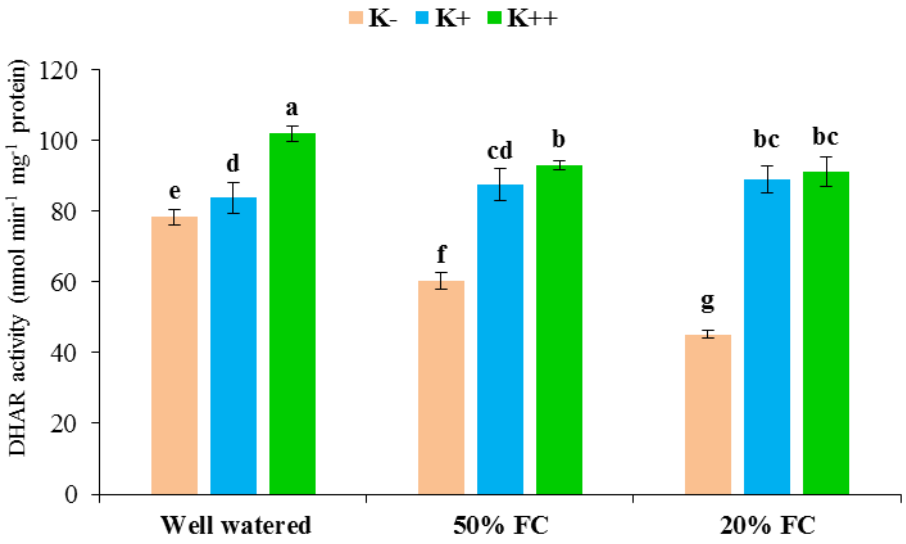

Figure 12. Effect of potassium supply on DHAR activity of wheat seedlings under three water condition. Here the treatments, well watered= 100% field capacity (control), 50% FC, 20% FC and -K, +K and +2K indicate 0 mM potassium, 6 mM potassium and 12 mM potassium respectively. ($LSD_{0.05}$ =4.61)

4.3.2.8 Glutathione reductase (GR) activity

Reduced GR activity compared to control was highly visible in drought stressed seedlings. Potassium apply increased GR content in drought stressed plant to an

extent. At 50% FC, 6 mM and 12 mM K increased GR content by 28% and 31% respectively without no significant differences compared to control (0 mM K). Whereas in 20% FC, 6 mM K increased GR level by 59% but 12 mM K increased only 38% GR content compared to control (Figure 13).

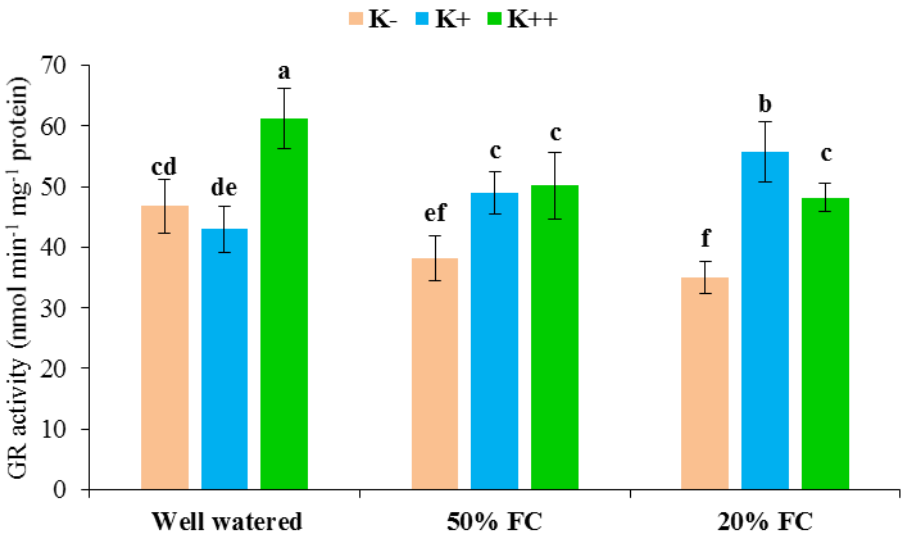

Figure 13. Effect of potassium supply on GR activity of wheat seedlings under three water condition. Here the treatments, well watered= 100% field capacity (control), 50% FC, 20% FC and -K, +K and +2K indicate 0 mM potassium, 6 mM potassium and 12 mM potassium respectively. ($LSD_{0.05}$ = 5.17)

Glutathione reductase (GR) is an important NADPH-dependent enzyme in AsA-GSH cycle because it plays a vital role in defense system against overproduction of ROS in various stress conditions (Romero-Puertas *et al.*, 2006). In plants GR catalyses the NADPH-dependent reduction of disulphide bond of GSSG into GSH, thus it is essential for maintaining the GSH status, GSH/GSSG ratio and for accelerating the H_2O_2 scavenging pathways under abiotic stress conditions (Pang and Wang, 2010).

4.3.2.9 Catalase (CAT) activity

Catalase is considered as a vital enzyme for ROS detoxification. CAT activity has been reduced under increasing drought stress compared to control seedlings (well watered). Potassium treatment increased CAT activity both in 50% and 20% FC. Although the increase has no significant difference in 50% FC with 6 mM and 12 mM K application. But in 20% FC it showed the highest increase of CAT activity with 6 mM K supply which is 59% compared to the respective control (0mM K (Figure 14).

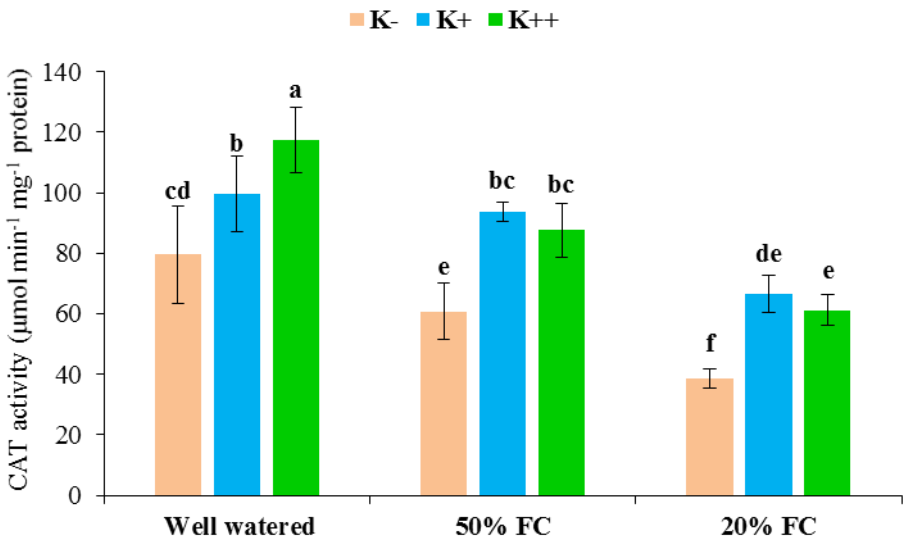

Figure 14. Effect of potassium supply on CAT activity of wheat seedlings under three water condition. Here the treatments, well watered= 100% field capacity (control), 50% FC, 20% FC and -K, +K and +2K indicate 0 mM potassium, 6 mM potassium and 12 mM potassium respectively. ($LSD_{0.05}$ = 2.34)

Catalase is one of the major enzymes that convert excess H_2O_2 into water. Several earlier reports suggested that higher activity of this enzyme helps to reduce excess level of H_2O_2 (Hasanuzzaman *et al.*, 2012a). In this study, under exposure to drought stress CAT activity decreased. Increased production of H_2O_2 or ineffective synthesis of enzyme under drought stress may result decreased activity of CAT (Gupta *et al.*, 2009). However, potassium treated drought

stressed seedlings showed enhance CAT activity than those under drought stress without. This trend was supported by Ahanger and Agarwal (2016) and Sharma *et al.* (2006) who reported that potassium treatment enchance the CAT activity under abiotic stress conditions.

4.3.2.10 Glutathione peroxidase (GPX) activity

Compared with the control seedlings GPX activity decreased in the drought stressed seedlings. However seedlings which were treated with potassium showed an increase in GPX activity both in 50% and 20% FC with 6 mM and 12 mM K. However the highest increase 64% was observed at 20% FC with 6 mM K. At 50% FC with K supply no significant difference was observed (Figure 15).

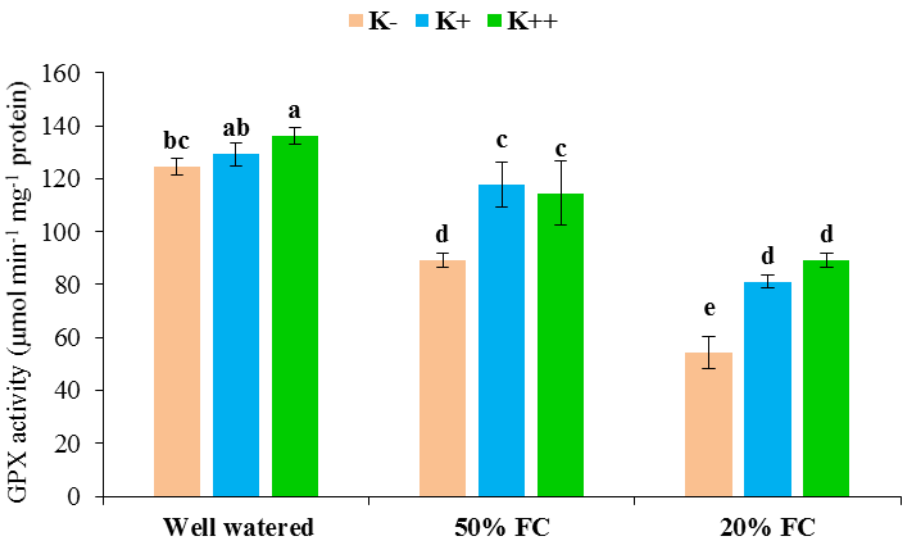

Figure 15. Effect of potassium supply on GPX activity of wheat seedlings under three water condition. Here the treatments, well watered= 100% field capacity (control), 50% FC, 20% FC and -K, +K and +2K indicate 0 mM potassium, 6 mM potassium and 12 mM potassium respectively. ($LSD_{0.05}$ = 4.74)

The GPX is another vital enzyme of antioxidant defense system and due to substrate specifications and stronger affinity for H_2O_2 it can efficiently scavenge,

especially, H_2O_2 and thus provide protection against stress (Hasanuzzaman and Fujita, 2011b). Drought stress significantly decreased GPX activities in wheat seedlings, compared to untreated control seedlings. Similar observations of increased GPX under drought stress were reported by several researchers (Liu *et al*., 2010). But compared to drought stress potassium supplemented drought treatment improved GPX activity further. Similarly, it was found that application of potassium increased GPX activity in Brassica sp. under drought Blackgram and mungbean under drought stress (Singh *et al*., 2002; Asgar *et al*., 2006).

4.3.3 Glyoxalase system and methylglyoxal detoxification

The Glyoxalase system, comprising the Gly I and Gly II enzymes, effectively eliminates cytotoxic MG**.**

4.3.3.1 Methylglyoxal content

Plant exposed to drought stress significantly elevated the MG content compared to control seedlings. However, plant treated with potassium drastically reduced the MG content in stressed plant. At 50% FC and 20% FC, 6 mM K reduced MG content by 15% and 27% accordingly, compared to their control (0 mM K). On the contrary, with 12 mM K supply at both water stress condition reduced MG content by 20% individually, compared to control (Figure 16).

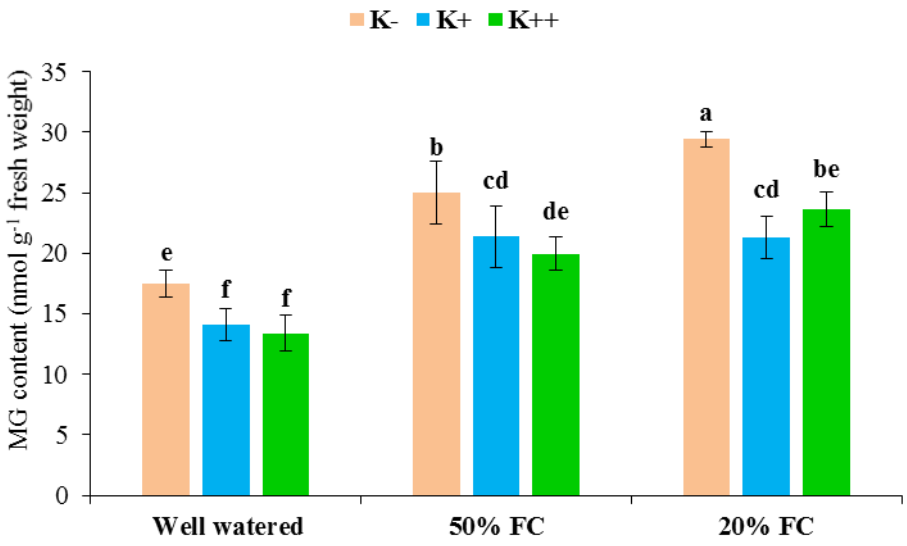

Figure 16. Effect of potassium supply on MG content of wheat seedlings under three water condition. Here the treatments, well watered= 100% field capacity (control), 50% FC, 20% FC and -K, +K and +2K indicate 0 mM potassium, 6 mM potassium and 12 mM potassium respectively. ($LSD_{0.05}$ = 3.08)

4.3.3.2 Gly I activity

Gly I activity was considerably reduced under drought stress condition compared with control seedlings. However, potassium treatment increased Gly I activity in 20% FC condition while in 50% it slightly reduced the Gly I content. Furthermore, no significant difference was observed due to K supply at 50% FC compared to control. (Figure 17).

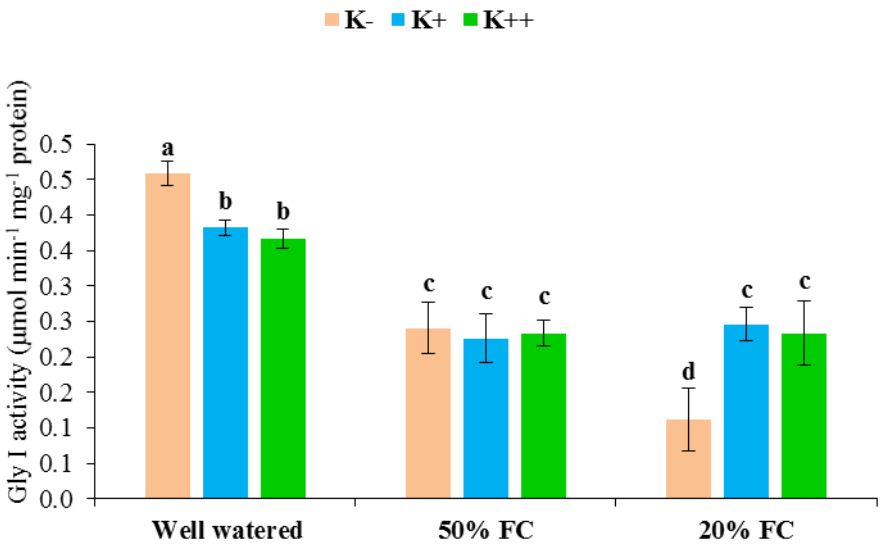

Figure 17. Effect of potassium supply on Gly I content of wheat seedlings under three water condition. Here the treatments, well watered= 100% field capacity (control), 50% FC, 20% FC and -K, +K and +2K indicate 0 mM potassium, 6 mM potassium and 12 mM potassium respectively. ($LSD_{0.05}$ = 0.04)

4.3.3.2 Gly II activity

Gly II activity was reduced while plant exposed to drought stress compared to control. Potassium supplementation further decreased or increased the Gly II content in different stressed plant. In 50% FC, 6 mM and 12 mM K further decreased the Gly II content to 10% and 18% respectively compared to control. However in 20% FC, 6 mM K reduced 15% Gly II content but 12 mM K increased the content 32% in a significant manner (Figure 18).

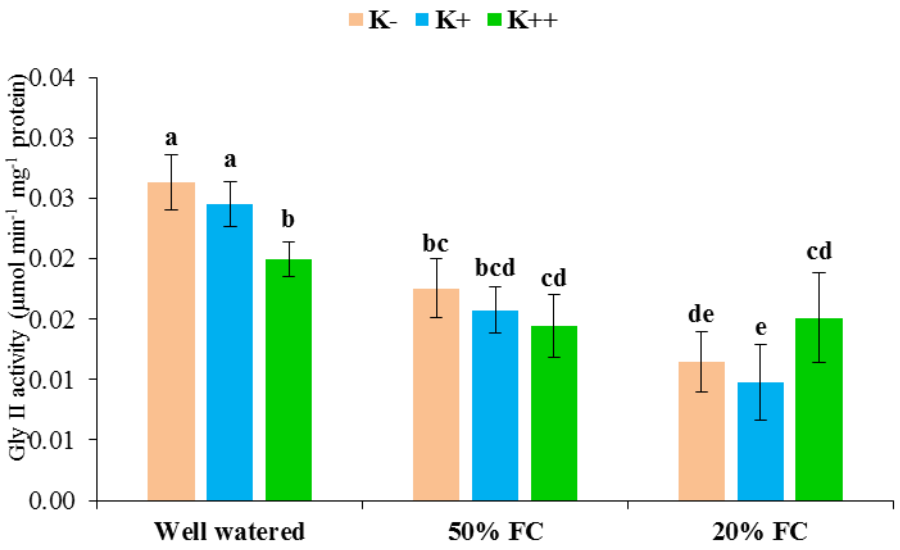

Figure 18. Effect of potassium supply on Gly II content of wheat seedlings under three water condition. Here the treatments, well watered= 100% field capacity (control), 50% FC, 20% FC and -K, +K and +2K indicate 0 mM potassium, 6 mM potassium and 12 mM potassium respectively. ($LSD_{0.05}$ = 2.83)

Under abiotic stress condition cytotoxic MG production aggrevated which can enhance the ROS production. Rapid accumulation of endogenous MG has been observed in plants under various abiotic and biotic stresses condition. MG detoxification is one of the potential systems for actuating tolerance in plants against various abiotic and biotic stresses (Yadav *et al.*, 2005). To detoxify MG, Glyoxalase system plays pivotal roles which is includes Gly I and Gly II enzymes. Upregulation of Gly I and Gly II can limit the over accumulation of MG. In our study, it was observed that drought stress increased MG production but decreased activity of Gly I and Gly II. Similar results were reported by Alam *et al.* (2013) and. However, stressed plant treated with potassium considerably further reduces the activities of Gly I and Gly II under drought stress condition.

CHAPTER 5

SUMMARY AND CONCLUSION

The present study was conducted in the Laboratory of Plant Stress Responses, Kagawa University, Japan, to evaluate the relative effectiveness of the three potassium doses (0mM, 6 mM and 12 mM) in alleviating the detrimental effects caused by drought stress on wheat seedlings. The experiments were arranged in completely randomized design (CRD) with three replications. Seedlings were grown in controlled environment of greenhouse where drought stress was imposed by withholding water at 50% FC and 20% FC and the Data were taken by sampling the leaves of 9 days stressed seedlings after 21 days of normal growing period. Different data of growth, physiology and biochemical parameters were measured. Plant height, fresh weight and dry weight were measured.

Physiological parameters includes leaf chl contents, relative water content (RWC) where biochemical parameters includes lipid peroxidation (MDA content), H_2O_2, pro, AsA, GSH, GSSG, MG and antioxidant enzyme like CAT, APX, MDHAR, DHAR, GR, Gly I and Gly II activities were observed thoroughly to investigate the drought effect and ameliorative role of K during drought in wheat seedlings.

In the present study drought stress significantly reduced plant height, fresh and dry weight plant^{-1}. However, exogenous K application improved the above mentioned growth parameters. Among three doses of potassium, 12 mM K at 50% FC was the most effective in alleviating the drought induced growth reduction and in promoting seedling growth under water stress conditions.

Drought stress results in significant reduction of leaf RWC and chl content. However, exogenous supplementation of potassium restored the tissue water

status as well as the photosynthetic pigments. Here, 6 mM and 12 mM showed almost the similar effects in restoring the water status and Chl contents under 50% FC while in 20% FC, 6 mM K showed the best result in storing leaf pigments and cell water content.

Drought induced oxidative damage result in a drastic increase in MDA and H_2O_2 and proline contents. Potassium application reduced the oxidative damage by reducing the MDA, H_2O_2 and Proline contents was studied in drought stressed wheat seedlings. This proved that potassium possess some adaptive mechanisms against drought.

Drought stress also found to enhance MG content, decreased GSH/GSSG ratio and alters the antioxidant defense system in wheat seedlings which we can describe that drought leds to a severe oxidative damage due to inappropriate induction of ROS and MG detoxification systems. Antioxidant defense system comprises non-enzymatic antioxidants such as AsA and GSH and enzymatic antioxidants such as CAT, APX, MDHAR, DHAR, GPX etc. Most importantly, potassium treatment modulated the activities of CAT, APX, MDHAR, DHAR, GR, GPX, Gly I and Gly II and higher GSH/GSSG ratio with an associated decrease in oxidative stress parameter like MDA and H_2O_2 as compared to the seedlings subjected to drought stress without K treatment. Finally, we may conclude with remarks that potassium as an essential plant nutrient works as a plant protectant that impart a diverse ranges of beneficial effect in plant while exposed to drought. Application of K during drought stress recovered plant growth and reduces water loss and thus increases the photosynthesis rate. Potassium also imparts drought stress tolerance by reducing ROS and MG production through upregulationg antioxidant and glyoxalase system, respectively. At 50% water stress, plant treated with 12 mM potassium showed better plant physiological performances than control and other treatment combination by reducing ROS production and lipid peroxidation while upregulating antioxidant defense and glyoxalase system.

REFERENCES

Abass, S. M. and Mohamed, H. I. (2011). Alleviation of adverse effects of drought stress on common bean (*Phaseolus vulgaris* L.) by exogenous application of hydrogen peroxide. *Bang. J. Bot.* **41**: 75–83.

Abbaspour, H., Saeidi-Sar, S. and Afshari, H. (2011). Improving drought tolerance of *Pistacia vera* L. Seedlings by arbuscular mycorrhiza under greenhouse conditions. *J. Med. Plants Res.* **5**: 7065–7072.

Abbate, P. E., Dardanellib, J. L., Cantareroc, M. G., Maturanoc, M., Melchiorid, R. J. M. and Sueroa, E. E. (2004). Climatic and water availability effects on water use efficiency in wheat. *Crop Sci.* **44:**474–483.

Abdoli, A., Saeidi, M., Jalali-Honarmand, S., Mansourifar, S. and Ghobad, M. (2013). Effect of post-anthesis water deficiency on storage capacity and contribution of stem reserves to the growing grains of wheat cultivars. *Plant Knowl. J.* **2**(1): 99107.

Addinsoft. (2018). XLSTAT v. 2018: Data analysis and statistics software for Microsoft Excel. Addinsoft, Paris, France.

Agarwal, M., Hao, Y., Kapoor, A., Dong, C. H., Fujii, H., Zheng, X. and Zhu, J. K. (2006). A R2R3 type MYB transcription factor is involved in the cold regulation of CBF genes and in acquired freezing tolerance. *J. Biol. Chem.* **281**(49): 37636-37645.

Ahanger, M. A. and Agarwal, R. M. (2016). Potassium improves antioxidant metabolism and alleviates growth inhibition under water and osmotic stress in wheat (*Triticum aestivum* L.). *Protoplasma* DOI: 10.1007/s00709–016–1037-0.

Ahmad, P. and Prasad, M. N. V. (2012). Abiotic stress responses in plants: metabolism, productivity and sustainability. Springer, New York.

Ahmed, B. C., Ben-Rouina, B., Sensoy, S., Boukhriss, M. and Ben-Abdullah, S. F. (2010). Exogenous proline effects on photosynthetic performance and antioxidant defense system of young olive tree. *J. Agric. Food. Chem.* **58**(7): 4216-4222.

Ahmed, S. M. and Meisner, C. A. (1996). Wheat Research and Development in Bangladesh. Dhaka: International Maize and Wheat Improvement Center (CIMMYT).

Akram, M. (2011). Growth and yield components of wheat under water stress of different growth stages. *Bangladesh J. Agric. Res.* **36:** 455–468.

Al Hassan, M., Chaura, J., Donat-Torres, M.P., Boscaiu, M. and Vicente, O. (2017). Antioxidant responses under salinity and drought in three closely related wild monocots with different ecological optima. *AoB PLANTS*. **9**: plx009.

Alam, M., Hasanuzzaman M., Nahar, K. and Fujita, M. (2013). Exogenous salicylic acid ameliorates short-term drought stress in mustard (*Brassica juncea* L.) seedlings by upregulating the antioxidant defense and glyoxalase system. *Aust. J. Crop Sci*. **7**: 1053-1063.

Al-Ghamdi, A. A. (2009). Evaluation of oxidative stress tolerance in two wheat (*Triticum aestivum*) cultivars in response to drought. *Int. J. Agric. Biol.* **11**: 7–12.

Ali, A., Ali, N., Ullah, N., Ullah, F., Adnan, M. and Ahmed, Z. (2013). Effect of drought stress on the physiology and yield of the Pakistani wheat germplasms. *Int. J. Ad. Res. Tech.* **2**(7): 419-430.

Amtmann, A., Armengaud, P. and Volkov, V. (2004). Potassium nutrition and salt stress. In: Membrane Transport in Plants, Blatt M.R., ed. (Oxford: Blackwell Publishing), pp. 293–339.

Apel, K. and Hirt, H. (2004) Reactive oxygen species: metabolism, oxidative stress, and signal transduction. *Annu. Rev. Plant Biol.* **55:** 373–399.

Araus, J. L., Slafer, G. A., Reynolds, M. P. and Royo, C. (2002). Plant breeding and drought in C3 cereals: what should we breed for? *Ann. Bot*. **89**: 925-940

Arnon, D. T. (1949). Copper enzymes in isolated chloroplasts polyphenol oxidase in *Beta vulgaris*. *Plant Physiol*. **24**: 1-15.

Asada, K. and Takahashi, M. (1987). Production and scavenging of active oxygen in photosynthesis. **In:** Photoinhibition: Topics of photosynthesis.

D. J. Kyle, C. B. Osmond and C. J. Arntzen, (ed.). 9th edn, Elsevier, Amsterdam, The Netherlands. pp. 227-287.

Asgar, A., Nadeem, M. A., Muddassar, M. and Ejaz, M. (2006). Effect of different levels of potash on growth, yield and protein contents of mungbean varieties. *J. Agric. Res.* **44**(2): 121 -126.

Ashraf, M. and Bashir, A. (2003). Salt stress induced changes in some organic metabolites and ionic relations in nodules and other plant parts of two crop legumes differing in salt tolerance. *Flora*. **198**: 486- 498.

Assefa, Y., Staggenborg, S. A., and Prasad, V. P. V. (2010). Grain sorghum water requirement and responses to drought stress: a review. *Online Crop Manag.* doi: 10.1094/CM-2010-1109-01-RV.

Atreya, A., Vartak, V. and Bhargava, S. (2009). Salt priming improves tolerance to desiccation stress and extreme salt stress in *Bruguiera cylindrical. Int. J. Integr. Biol*. **6**(2): 68-73.

Banker, K. B., Gosavi, S. V. and Balsanen, V. K. (2008). Effect of different irrigation treatment on growth and yield of wheat crop varieties. *Intl. J. Agric. Sci.* **4**: 114-118.

Barrs, H. D. and Weatherley, P. E. (1962). A re-examination of the relative turgidity technique for estimating water deficits in leaves. *Aust. J. Biol. Sci.* **15**: 413-428.

Bates, L. S., Waldren, R. P. and Teari, D. (1973). Rapid determination of free proline for water stress studies. *Plant Soil* **39**: 205-207.

Bazzaz, M. (2013). Growth, production physiology and yield in wheat under variable water regimes. Ph. D. thesis. Dept. of Agronomy. Bangabandhu Shiekh Mujibur Rahman Agricultural University, Gazipur.

Beck, E. H., Fettig, S., Knake, C., Hartig, K. and Bhattarai T. (2007). Specific and unspecific responses of plants to cold and drought stress. *J. Biosci.* **32**: 501–510.

Berkowitz, G. A. and Peters, J. S. (1993). Chloroplast inner envelope ATPase acts as a primary H+ pump. *Plant Physiol.* **102:**261–267.

Blokhina, O., Virolainen, E. and Fagerstedt, K. V. (2003). Antioxidants, oxidative damage and oxygen deprivation stress: a review. *Ann. Bot.* **91**: 179–194.

Bradford, M. M. (1976). A rapid and sensitive method for the quantitation of microgram quantities of protein utilizing the principle of protein-dye binding. *Anal Biochem.* **72**(1-2): 248-254.

Bray, E. A., Bailey-Serres, J. and Weretilnyk, E. (2000). Responses to abiotic stresses. In: Buchanan BB, Gruissem W and Jones RL (eds) Biochemistry and molecular biology of plants. Amer. Soc. *Plant Physiol.* Rockville, pp. 1158-1203.

Bukhsh, M. A. A. H., Ahmad, A. R., Iqbal, J., Maqbool, M. M., Ali, A., Ishaque, M. and Hussain, S. (2012). Nutritional and physiological significance of potassium application in maize hybrid crop production. *Pak. J. Nutr.* **11**(2): 187-202.

Cakmak, I. (2005). The role of potassium in alleviating detrimental effects of abiotic stresses in plants, *J. Plant Nutri. Plant Sci.* **168**: 521–530.

Cattivelli, L., Rizza, F., Badeck, F.W., Mazzucotelli, E., Mastrangelo, A.M., Francia, E., Mare, C., Tondelli, A. and Stanca, A.M. (2008). Drought tolerance improvement in crop plants: an integrated view from breeding to genomics. *Field Crops Res.* **105**(1-2): 1-14.

Ceccarelli, S., Grando, S., Maatougui, M., Michael, M., Slash, M., Haghparast, R., Rahmanian, M., Taheri, A., Al-Yassin, A., Benbelkacem, A., Labdi Mimoun, H. and Nachit. M. (2010). Plant breeding and climate changes. *J. Agric. Sci.* **148**: 627-637.

Chandler, S. S. and Singh, T. K. (2008). Selection Criteria for Drought Tolerance in Spring Wheat (*Triticum aestivum* L.). In: Appels, R., Eastwood, R., Lagudah, E., Langridge, P. and Mackay, M. (eds) The 11th International Wheat Genetics Symposium Proceedings, Series: Coping with Wheat in a Changing Environment Abiotic Stresses, Sydney University Press, Lynne. pp. 1-3.

Chaves, M. M. and Oliveira, M. M. (2004). Mechanisms underlying plant resilience to water deficits: prospects for water-saving agriculture. *J. Exp. Bot*. **55**: 2365–2384.

Chaves, M. M., Pereira, J. S. and Maroco, J. (2003). Understanding plant response to drought from genes to the whole plant. *Funct. Plant Biol.* **30**(3): 239-264.

Choudhury, S., Panda, P., Sahoo, L. and Panda, S. K. (2013). Reactive oxygen species signaling in plants under abiotic stress. *Plant Signal. Behav.* **8**: e23681.

Cochard, H., Coll, L., Roux, X. L. and Amegilo, T. (2002). Unraveling the effects of plant hydraulics on stomatal closer during water stress in walnut. *Plant Physio.* **128**(1): 282–290.

Collins, N. C., Tardieu, F. and Tuberosa, R. (2008). Quantitative trait loci and crop performance under abiotic stress: where do we stand? *Plant Physiol.* **147**: 469-486.

Comas, L. H., Becker, S. R., Cruz, V. M. V., Byrne, P. F. and Dierig, D. A. (2013). Root traits contributing to plant productivity under drought. *Front. Plant Sci.* **4**: 1–16.

Cornic, G. and Massacci, A. (1996). Leaf photosynthesis under drought stress, in: Baker N.R., (Ed.), Photosynthesis and the Environment, Kluwer Academic Publishers, The Netherlands.

Cruz de Carvalho, M. H. (2008). Drought stress and reactive oxygen species: Production, scavenging, and signaling. *Plant Signal. Behav.* **3**(3): 156-65.

DAE (Department of agriculture extension) (2018). http://dae.portal.gov.bd/ (Accessed 12 May 2018)

Damon, P. M. and Rengel, Z. (2007). Wheat genotypes differ in potassium efficiency under glasshouse and field conditions. *Aust. J. Agric. Res.* **58**(8): 816-823.

De Campos, M. K. F., de Carvalho, K., de Souza, F. S., Marur, C. J., Pereira, L. F. P., Filho, J. C. B. and Vieira, L. G. E. (2011). Drought tolerance and

antioxidant enzymatic activity in transgenic ‘Swingle’citrumelo plants over-accumulating proline. *Environ. Exp. Bot.***72**: 242–250.

Demirevska, K., Simova-Stoilova, L., Fedina, I., Georgieva, K. and Kunert, K. (2010). Response of oryzacystatin I transformed tobacco plants to drought, heat and light stress. *J. Agron. Crop Sci.* **196**(2): 90–99.

Desclaux, D. and Roumet, P. (1996). Impact of drought stress on the phenology of two soybean (*Glycine max* L. Merr) cultivars. *Field Crops Res*. **46**: 61–70.

Dey, N. C., Alam, M. S., Sajjan, A. K., Bhuiyan, M. A., Ghose, L., Ibaraki, Y. and Karim, F. (2011). Assessing Environmental and Health Impact of Drought in the Northwest Bangladesh. *J. Environ. Sci. Nat. Res.* **4**(2): 89-97.

Dhanda, S. S. and Sethi, G. S. (2002). Tolerance to drought stress among selected Indian wheat cultivars. *J. Agric. Sci.* **139**: 319–326.

Din, J., Khan, S. U., Ali, I. And Gurmani, A. R. (2011). Physiological and agronomic response of canola varieties to drought stress. *J. Anim. Plant Sci.* **21**(1): 78–82.

Egilla, J. N., Davies, F. T. and Boutton, T.W. (2005). Drought stress influences leaf water content, photosynthesis, and water-use efficiency of *Hibiscus rosa-sinensis* at three potassium concentrations. *Photosynthetica*. **43**(1): 135-140.

Ehdaie, B., Alloush, G. A. and Waines, J. G. (2008). Genotypic variation in linear rate of grain growth and contribution of stem reserves to grain yield in wheat. *Field Crops Res.* **106**(1): 34–43.

Ehdaie, B., Layne, A. P. and Waines, J. G. (2012). Root system plasticity to drought influences grain yield in bread wheat. *Euphytica.* **186**(1): 219-232.

Elia, A. C., Galarini, R., Taticchi, M. I., Dorr, A. J. M. and Mantilacci, L. (2003). Antioxidant responses and bioaccumulation in *Ictalurus melas* under mercury exposure. *Ecotoxicol. Environ. Saf.* **55**(2): 162–167.

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

Eltayeb, A. E., Kawano, N., Badawi, G., Kaminaka, H., Sanekata, T., Shibahar, T., Inanaga, S. and Tanaka, K. (2007). Overexpression of monodehydroascorbate reductase in transgenic tobacco confers enhanced tolerance to ozone, salt and polyethylene glycol stresses. *Planta*. **225**(5): 1255-1264.

Faize, M., Burgos, L., Faize, L., Piqueras, A., Nicola, E., Barba-Espin, G., Clemente-Moreno, M. J., Alcobendas, R., Artlip, T. and Hernández, J. A. (2011). Involvement of cytosolic ascorbate peroxidase and Cu/Zn-superoxide dismutase for improved tolerance against drought stress. *J. Exp. Bot.* doi: 10.1093/jxb/erq432.

Fang, J., Wu, F., Yang, W., Zhang, J. and Cai, H. (2012). Effects of drought on the growth and resource use efficiency of two endemic species in an arid ecotone. *Acta Ecol. Sinica.* **32**: 195–201.

FAO (2014). http://www.fao.org/faostat/en/#data/QC (accessed on 22 August, 2017).

FAO, Food outlook (2016) biannual report on global food markets

FAO, Food outlook (2017) biannual report on global food markets

FAOStat (Food AND Agriculture Organization of The United Nations), (2013).

FAOStat, (2018). http://www.fao.org/faostat/en/#data/QC Retrieved 18 April 2018.

FAOStat. Food and Agriculture Organization of The United Nations. (2016). Food and agricultural commodities production. http://faostat3.fao.org.

Farooq, M., Irfan, M., Aziz, T., Ahmad, I. and Cheema, S. (2013). Seed priming with ascorbic acid improves drought resistance of wheat. *J. Agron. Crop Sci.* **199**: 12-22.

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

Fayez, K. A. and Bazaid, S. A. (2014). Improving drought and salinity tolerance in barley by application of salicylic acid and potassium nitrate. *J. Saudi Society Agric. Sci.* **13**: 45-55.

Fernando, M., Kulpa, J., Siddiqi, M. Y. and Glass, A.D. (1990). Potassium-Dependent Changes in the Expression of Membrane-Associated Proteins in Barley Roots: I. Correlations with K+ (86Rb+) Influx and Root K+ Concentration. *Plant physiol.* ***92***(4):1128-1132.

Forman, H. J., Zhang, H. and Rinna, A. (2009). Glutathione: overview of its protective roles, measurement, and biosynthesis. *Mol. Asp. Med*. **30**: 1-12.

Foyer, C.H., Vanacker, H., Gomez L.D. and Harbinson, J. (2002). Regulation of photosynthesis and antioxidant metabolism in maize leaves at optimal and chilling temperatures: Review. *Plant Physiol. Biochem.* **40**(6-8): 659-668.

Gao, J. M., Xiao, Q., Ding, L. P., Chen, M. J., Yin, L., Li, J. Z., Zhou, S. Y. and He, G. Y. (2008). Differential responses of lipid peroxidation and antioxidants in *Alternanthera philoxeroides* and *Oryza sativa* subjected to drought stress. *Plant Growth Regul.* **56**(1): 89–95.

Geerts, S., Raes, D., Garcia, M., Mendoza, J. and Huanca, R. (2008). Crop water use indicators to quantify the flexible phenology of quinoa (*Chenopodium quinoa* Willd.) in response to drought stress. *Field Crops Res.* **108:** 150–156.

Ghosh, N., Adak, M. K., Ghosh, P. D., Gupta, S., Sen Gupta, D. N. and Mandal, C. (2011). Differential responses of two rice varieties to salt stress. *Plant Biotechnol. Rep.* **5**: 89–103.

Gill, S. S. and Tuteja, N. (2010). Reactive oxygen species and antioxidant machinery in abiotic stress tolerance in crop plants. *Plant Physiol. Biochem.* **48**: 909–930.

GOB (Government of Bangladesh) (2016) Bangladesh Economic Review – 2016. Dhaka: Ministry of Finance, http://www.mof.gov.bd/ (accessed March 08, 2017).

Gupta, A. S., Berkowitz, G. A. and Pier, P.A. (1989). Maintenance of photosynthesis at low leaf water potential in wheat: role of potassium status and irrigation history. *Plant Physiol.* ***89*****(**4): 1358-1365.

Gupta, M., Sharma, P., Sarin, N. B. And Sinha, A. K. (2009). Differential response of arsenic stress in two species of *Brassica juncea* L. *Chemosphere.* **74**(9): 1201–1208.

Hameed, A., Goher, M. and Iqbal, N. (2013). Drought induced programmed cell death and associated changes in antioxidants, proteases, and lipid peroxidation in wheat leaves. *Biol. Plant*. **57**: 370-374.

Harb, A., Krishnan, A., Ambavaram, M. M. R. and Pereira, A. (2010). Molecular and physiological analysis of drought stress in Arabidopsis reveals early responses leading to acclimation in plant growth. *Plant Physiol.* **154**: 1254–1271.

Hasan, M. K. (2006). Yield gap in wheat production: A perspective of farm specific efficiency in Bangladesh. Ph.D. dissertation, Dept. of Agricultural Economics, BAU, Mymensingh.

Hasanuzzaman, M. and Fujita, M. (2011a). Selenium pretreatment upregulates the antioxidant defense and methylglyoxal detoxification system and confers enhanced tolerance to drought stress in rapeseed seedlings. *Biol. Trace Elem. Res.* **143(**3): 1758–1776.

Hasanuzzaman, M. and Fujita, M. (2013). Exogenous sodium nitroprusside alleviates arsenic-induced oxidative stress in wheat (*Triticum aestivum* L.) seedlings by enhancing antioxidant defense and glyoxalase system. *Ecotoxicology*. **22**(3): 584–596.

Hasanuzzaman, M., Alam, M. M., Nahar, K., Mahmud, J. A., Ahamed, K. U. and Fujita, M. (2014a). Exogenous salicylic acid alleviates salt stress-induced oxidative damage in *Brassica napus* by enhancing the antioxidant defense and glyoxalase systems. *Aust. J. Crop Sci.* **8**: 631-639.

Hasanuzzaman, M., Bhuyan, M. H. M., Nahar, K., Hossain, M. S., Mahmud, J. A., Hossen, M. S., Masud, A. A. C. and Fujita, M. (2018). Potassium: A Vital Regulator of Plant Responses and Tolerance to Abiotic Stresses. *Agronomy*. **8**(3): 31.

Hasanuzzaman, M., Hossain, M. A. And Fujita, M. (2011b). Nitric oxide modulates antioxidant defense and the methylglyoxal detoxification system and reduces salinity-induced damage of wheat seedlings. *Plant Biotechnol. Rep.* **5**(4): 353–365.

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

Hasanuzzaman, M., Nahar, K., Alam, M. M. and Fujita, M. (2012b). Exogenous nitric oxide alleviates high temperature induced oxidative stress in wheat (*Triticum aestivum*) seedlings by modulating the antioxidant defense and glyoxalase system. *Aust. J. Crop Sci.* **6**(8): 1314–1323.

Hasanuzzaman, M., Nahar, K., Alam, M. M. and Fujita, M. (2014b). Modulation of antioxidant machinery and the methylglyoxal detoxification system in selenium-supplemented *brassica napus* seedlings confers tolerance to high temperature stress. *Biol. Trace Elem. Res.* **161**: 297–307.

Hasanuzzaman, M., Nahar, K., Gill, S. S. and Fujita, M. (2013). Drought stress responses in plants, oxidative stress, and antioxidant defense. In: Tuteja, N. and Gill, S. S. (eds) Climate Change and Plant Abiotic Stress Tolerance. Weinheim: Wiley-VCH Verlag GmbH & Co. KGaA. pp. 209-250.

Hasanuzzaman, M., Nahar, K., Hossain, M. S., Mahmud, J. A., Rahman, A., Inafuku, M., Oku, H. and Fujita, M. (2017). Coordinated actions of glyoxalase and antioxidant defense systems in conferring abiotic stress tolerance in plants. *Int. J. Mol. Sci.* **18**(1): 200. doi:10.3390/ijms18010200

Heath, R. L. and Packer, L. (1968). Photo-peroxidation in isolated chloroplasts: I. Kinetics and stoichiometry of fatty acid peroxidation. *Arch. Biochem. Biophys.* **125**: 189–198.

Hoque, M.A., Banu, M.N.A., Nakamura, Y., Shimoishi, Y. and Murata, Y. (2008). Proline and glycinebetaine enhance antioxidant defense and methylglyoxal detoxification systems and reduce NaCl-induced damage in cultured tobacco cells. *J Plant Physiol.* **165**(8): 813–824.

Hossain, M. A. and Fujita, M. (2009). Exogenous glycinebetaine and proline modulate antioxidant defense and methylglyoxal detoxification systems and reduce drought induced damage in mung bean seedlings (*Vigna radiata* L.). In: Proceedings of the 3rd international conference on integrated approaches to improve crop production under drought-prone environments, 11–16 Oct 2009. Shanghai, China, pp 138

Hossain, M. A., Nakano, Y. and Asada, K. (1984). Monodehydroascorbate reductase in spinach chloroplasts and its participation in the regeneration of ascorbate for scavenging hydrogen peroxide. *Plant Cell Physiol.* **25**(3): 385–395.

Hou, X., Xie, K., Yao, J., Qi, Z. and Xiong, L. (2009). A homolog of human ski-interacting protein in rice positively regulates cell viability and stress tolerance. *Proc. Natl. Acad. Sci.* USA **106**: 6410–6415.

Huang, C., He, W., Guo, J., Chang, X., Su, P. and Zhang, L. (2005). Increased sensitivity to salt stress in an ascorbate-deficient Arabidopsis mutant. *J. Exp. Bot.* **56** (422): 3041–3049.

Hussain, M., Malik, M. A., Farooq, M., Ashraf, M. Y. and Cheema, M. A. (2008). Improving drought tolerance by exogenous application of glycinebetaine and salicylic acid in sunflower. *J. Agron. Crop Sci.* **194**: 193–199.

Hussain, M., Malik, M. A., Farooq, M., Khan, M. B., Akram, M. and Saleem, M. F. (2009). Exogenous glycinebetaine and salicylic acid application improves water relations, allometry and quality of hybrid sunflower under water deficit conditions. *J. Agron. Crop Sci.* **195**: 98–109.

Ibrahim, H. M. (2014). Selenium pretreatment regulates the antioxidant defense system and reduces oxidative stress on drought-stresses wheat (*Triticum aestivum* L.) plants. *Asian. J. Plant Sci.* **13**: 120-128.

IPCC, (2007). Fourth assessment report: synthesis. http:// www.ipcc.ch/pdf/assessment

Islam, F., Yasmeen, T., Ali, S., Ali, B., Farooq, M. A. and Gill, R. A. (2015). Priming induced antioxidative responses in two wheat cultivars under saline stress. *Acta. Physiol. Plant.* **37**: 1-12.

Jaleel, C. A., Manivannan, P., Wahid, A., Farooq, M., Somasundaram, R., and Panneerselvam, R. (2009). Drought stress in plants: a review on morphological characteristics and pigments composition. *Int. J. Agric. Biol.* **11**(1): 100–105.

Jin, S. H., Huang, J. Q., Li, X. Q., Zheng, B. S., Wu, J. S., Wang, Z. J., Liu, G. H. and Chen, M. (2011). Effects of potassium supply on limitations of photosynthesis by mesophyll diffusion conductance in *Carya cathayensis*. *Tree Physiol.* **31**: 1142–1151.

Jones, D. P. (2006). Redefining oxidative stress. *Antioxid. Redox Signal.* **8**: 18651879.

Kang, G., Li, G., Xu, W., Peng, X., Han, Q., Zhu, Y. and Guo, T. (2012). Proteomics reveals the effects of salicylic acid on growth and tolerance to subsequent drought stress in wheat. *J. Proteome Res.* **11**: 6066-6079.

Kavas, M., Baloğlu, M. C., Akça, O., Köse, F. S. and Gökçay, D. (2013). Effect of drought stress on oxidative damage and antioxidant enzyme activity in melon seedlings. *Turk. J. Biol.* **37**: 491-498.

Kaya, D. M., Okcu, G., Atak, M., Cikili, Y. and Kolsarici, O. (2006). Seed treatments to overcome salt and drought stress during germination in sunflower (*Helianthus annuus* L.). *Eur. J. Agron*. **24**: 291–295.

Keyvan, S. (2010). The effects of drought stress on yield, relative water content, proline, soluble carbohydrates and chlorophyll of bread wheat cultivars. *J. Anim. Plant Sci*. **8**(3): 1051-1060.

Khan, H. R., Paull, J. G., Siddique, K. H. M. and Stoddard, F.L. (2010). Faba bean breeding for drought-affected environments: A physiological and agronomic perspective. *Field Crop Res* **115**(3): 279–286

Khosravifar S., Yarnia M., Benam, M. B. K. and Moghbeli, A. H. H. (2008). Effect of potassium on drought tolerance in potato cv. Agria. *J. Food Agric. Environ.* **6:**236-241

Kirkham, M. B. (2005). Principles of Soil and Plant Water Relations. Elsevier, The Netherlands.

Kulkarni, M. and Deshpande, U. (2007). In Vitro screening of tomato genotypes for drought resistance using polyethylene glycol. *Afr. J. Biotechnol*. **6**: 691-696.

Kuşvuran, Ş., Daşgan, Y. H. and Abak, K. (2011). Responses of different melon genotypes to drought stress. Yüzüncü Yıl University. *J. Agric. Sci.,* **21**: 209–219.

Lafitte, H. R., Yongsheng, G., Yan, S. and Li, Z. K. (2007). Whole plant responses, key processes, and adaptation to drought stress: the case of rice. *J. Exp. Bot.* **58**:169–175.

Lanceras, J., Pantuwan, G., Jongdee, B. and Toojinda, T. (2004). Quantitative trait loci associated with drought tolerance at reproductive stage in rice. *Plant Physiol.* **135**:384–399.

Larcher, W. (2006). Physiological Plant Ecology (4rd Edn), Rima, São Carlos, pp. 550

Liu, F., Andersen, M. N. and Jensen, C. R. (2003). Loss of pod set caused by drought stress is associated with water status and ABA content of reproductive structures in soybean. Funct. *Plant Biol.* **30:** 271–280.

Liu, F., Jensen, C. R. and Andersen, M. N. (2004). Drought stress effect on carbohydrate concentration in soybean leaves and pods during early reproductive development: its implication in altering pod set. *Field Crops Res.* **86**:1–13.

Liu, J. X., Liao, D. Q., Oane, R., Estenor, L., Yang, X. E., Li, Z. C. and Bennett, J. (2006). Genetic variation in the sensitivity of anther dehiscence to drought stress in rice. *Field Crops Res.* **97**: 87–100.

Liu, Z. J., Guo, Y. K. and Bai, J. G. (2010). Exogenous hydrogen peroxide changes antioxidant enzyme activity and protects ultrastructure in leaves of two cucumber ecotypes under osmotic stress. *J. Plant Growth Regul*. **29**: 171-183.

Lobell, D. B. and Gourdji, S. M. (2012). The influence of climate change on global crop productivity. *Plant Physiol*. **160**(4): 1686-1697.

Logan, B. (2005). Reactive oxygen species and photosynthesis. **In**: Smirnoff, N. (ed) Antioxidants and reactive oxygen species in plants. Blackwell, Oxford. pp. 250-267.

Lonbani, M. and Arzani, A. (2011) Morpho-physiological Traits Associated with Terminal Drought Stress tolerance in Triticale and Wheat. *Agric. Res*. **9**: 315329.

Lu, Z., Lu, J., Pan, Y., Lu, P., Li, X., Cong, R. and Ren, T. (2016). Anatomical variation of mesophyll conductance under potassium deficiency has a vital role in determining leaf photosynthesis. *Plant Cell Environ.* **39**(11): 2428–2439.

Macedo, A. F. (2012). Abiotic Stress Responses in Plants: Metabolism to Productivity. **In**: Abiotic stress responses in plants: Metabolism, productivity and sustainability. P. Ahmed, and M.N.V. Prasad, (ed.). Springer, New York. pp. 41 -61.

Mahmood, M., Bidabadi, S. S., Ghobadi, C. and Gray, D. J. (2012). Effect of methyl jasmonate treatments on alleviation of polyethylene glycol mediated water stress in banana (*Musa acuminata* cv. 'Berangan', AAA) shoot tip cultures. *Plant Growth Regul.* **68**: 161–169.

Mahouachi, J. (2007). Growth and mineral nutrient content of developing fruit on banana plants (*Musa acuminata* AAA, 'Grand Nain') subjected to water stress and recovery. *J. Hort. Sci. Biotechnol.* **82**:839-844.

Majid, S. A., Asghar, R. and Murtaza, G. (2007). Yield stability analysis conferring adaptation of wheat to pre- and post-anthesis drought conditions. *Pak. J. Bot.* **39**:1623–1637.

Malik, R. S., Dhankar, J. S. and Turner, N. C. (1979). Influence of soil water deficits on root growth of cotton seedlings. *Plant Soil.* **53**: 109–115.

Mantri, N., Patade, V., Penna, S., Ford, R. and Pang, E. (2012). Abiotic stress responses in plants: present and future. In: Ahmad P, Prasad MNV (eds) Abiotic stress responses in plants: metabolism, productivity and sustainability. Springer, New York, pp 1–19.

Marschner, H. (2002). *Mineral Nutrition of Higher Plants*, London, Academic Press.

Marschner, P. (2012). Marschner's Mineral Nutrition of Higher Plants, 3rd ed.; Academic Press: London, UK. pp. 178–189.

Martineau, E., Domec, J. C., Bosc, A., Denoroy, P., Fandino, V. A., Lavres Jr, J. and Jordan-Meille, L. (2017). The effects of potassium nutrition on water use in field-grown maize (Zea mays L.). *Environ. Exp. Bot.* **134**: 62–71.

Martínez, J. P. and Araya, H. (2010). Ascorbate–glutathione cycle: enzymatic and non-enzymatic integrated mechanisms and its biomolecular regulation. In: Anjum, N. A., Chan, M. T. and Umar, S. (eds.) Ascorbate glutathione pathway and stress tolerance in plants. Springer, Dordrecht, pp. 303–322.

McMaster, G. S. and Wilhelm, W. W. (2003). Phenological responses of wheat and barley to water and temperature: improving simulation models. *J. Agric. Sci.* **141**: 129–147.

Mengel, K. (2007). Potassium. In: Barker AV, Pilbeam DJ (Eds) Handbook of Plant Nutrition (1 st Edn), CRC Taylor and Francis, NY, pp 91-120

Mengel, K. and Kirkby E. A. (2001). Principles of Plant Nutrition. 5th ed., Kluwer Academic Publishers, Dordrecht.

Mhamdi, A., Queval, G., Chaouch, S., Vanderauwera, S., Van Breusegem, F. and Noctor, G. (2010). Catalase function in plants: a focus on Arabidopsis mutants as stress-mimic models. *J. Exp. Bot.* **61**(15): 4197-4220.

Mishra, A. K. and Singh, V. P. (2010). A review of drought concepts. J. *Hydrol.* **391**: 202–216

Mishra, S., Jha, A. B. and Dubey R. S. (2011). Arsenite treatment induces oxidative stress, upregulates antioxidant system, and causes phytochelatin synthesis in rice seedlings. *Protoplasma*. **248**(3): 565-577.

Mittler, R. (2002). Oxidative stress, antioxidants and stress tolerance. *Trends Plant Sci.* **7:** 405–410.

Mittler, R. and Blumwald, E. (2010). Genetic engineering for modern agriculture: challenges and perspectives. *Annu. Rev. Plant Biol.* **61**: 443–462.

Mittler, R., Vanderauwera, S., Suzuki, N., Miller, G., Tognetti, V. B., Vandepoele, K., Gollery, M., Shulaev, V., Van-Breusegem, F. (2011). ROS signaling: the new wave? *Trends Plant Sci.* **16**: 300–309.

Mittova, V., Volokita, M., Guy, M. and Tal, M. (2000). Activities of SOD and the ascorbate-glutathione cycle enzymes in subcellular compartments in leaves

and roots of the cultivated tomato and its wild salt-tolerant relative *Lycopersicon pennellii*. *Physiol. Plant.* **110**: 42–51.

Miyake, C. (2010). Alternative electron flows (Water cycle and cyclic electron flow around PSI) in photosynthesis: molecular mechanisms and physiological functions. *Plant Cell Physiol.* **51**(12): 1951-1963.

Mohammadkhani, N. and Heidari, R. (2007). Effects of drought stress on protective enzyme activities and lipid peroxidation in two maize cultivars. *Pak. J. Biol. Sci*. **10**: 3835– 3840

Moller, I. M., Jensen, P. E. and Hansson, A. (2007). Oxidative modifications to cellular components in plants. *Annu. Rev. Plant Biol*. **58**: 459–481.

Morgan, J. M. (1992). Osmotic components and properties associated with genotypic differences in osmoregulation in wheat. *Aust. J. Plant Physiol.* **19**(1): 67–76.

Mottaleb, K. A. and Krupnik, T. J. (2015) Uptake of Scale-Appropriate Agricultural Machinery in Bangladesh: Policy Insights from Historical

and Census Survey Analyses. Research Report No. 3. Cereal Systems initiative for South Asia Mechanization and Irrigation (CSISA-MI) project. Dhaka: CIMMYT, Bangladesh.

Mottaleb, K. A., Kruseman, G., and Erenstein, O. (2017). Wheat production and consumption dynamics in an Asian rice economy: The Bangladesh case. *European J. Dev. Res.* **30**(2): 252-275.

Mpelasoka, B. S., Schachtman, D. P., Treeby, M. T. and Thomas, M. R. (2003). A review of potassium nutrition in grapevines with special emphasis on berry accumulation. *Aus. J. Grape Wine Res.* **9**(3): 154–168.

Nahar, K., Hasanuzzaman, M. and Fujita, M. (2016) Roles of osmolytes in plant adaptation to drought and salinity. In: Iqbal *et al*. (eds.), Osmolytes and plants acclimation to changing environment. Springer, India, doi:10.1007/978-81-322-2616-1_4.

Nahar, K., Hasanuzzaman, M., Alam, M. M. and Fujita, M. (2013). Exogenous glutathione induced drought stress tolerance in *Vigna radiata* seedlings through enhanced antioxidant defense and methylglyoxal system. *Interdrought IV Conference* September 02-September 09, 2013, Perth, Australia.

Nahar, K., Hasanuzzaman, M., Alam, M. M. and Fujita, M. (2015). Exogenous spermidine alleviates low temperature injury in mung bean (*Vigna radiata* L.) seedlings by modulating ascorbate-glutathione and glyoxalase pathway. *Int. J. Mol. Sci*.**16**: 30117-30132.

Nakano, Y. and Asada, K. (1981). Hydrogen peroxide is scavenged by ascorbate-specific peroxidase in spinach chloroplasts. *Plant Cell Physiol.* **22**: 867–880.

Nelson, W.L. (1980). Soil test summaries and their interpretation. *Better Crops Plant Food.* **63**: 6–10.

Nguyen, G. N., Hailstones, D. L., Wilkes, M. and Sutton, B. G. (2009). Drought induced oxidative conditions in rice anthers leading to a programmed cell death and pollen abortion. *J. Agron. Crop. Sci.* **195**: 157–164.

Noctor, G. and Foyer, C. (1998). Ascorbate and glutathione: keeping active oxygen under control. *Annu. Rev. Plant Physiol. Plant Mol. Biol.* **49**: 249–279.

Noctor, G., Gomez, L., Vanacker, H. and Foyer, C. H. (2002). Interactions between biosynthesis, compartmentation, and transport in the control of glutathione homeostasis and signaling. *J. Exp. Bot*. **53**(372): 1283-1304.

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**: 596–604.

Nouri-Ganbalani, A., Nouri-Ganbalani, G. and Hassanpanah, D. (2009). Effects of drought stress condition on the yield and yield components of advanced wheat genotypes in Ardabil, Iran. *J. Food Agric. Environ*. **7**(3-4): 228-234.

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.

Ozkur, O., Ozdemir, F., Bor, M. and Turkan, I. (2009). Physiochemical and antioxidant responses of the perennial xerophyte *Capparis ovata* Desf. to drought. *Environ. Exp. Bot.* **66**: 487–492.

Pandey, H. C., Baig, M. J. and Bhatt, R. K. (2010). Drought stress induced changes in lipid peroxidation and antioxidant system in genus Avena. *J. Environ. Biol.* **31**: 435–440.

Pandey, H. C., Baig, M. J. and Bhatt, R. K. (2012). Effect of moisture stress on chlorophyll accumulation and nitrate reductase activity at vegetative and flowering stage in *Avena* species. *Agric. Sci. Res. J.* **2**: 111–118

Pandey, V. and Shukla A. (2015). Acclimation and tolerance strategies of rice under drought stress. *Rice Sci.* **22**(4): 147–161.

Pang, C.H. and Wang, B.S. (2010). Role of ascorbate peroxidase and glutathione reductase in Ascorbate–Glutathione cycle and stress tolerance in plants.

In: AscorbateGlutathione pathway and stress tolerance in plants. N.A. Anjum, M.T. Chan and S. Umar, (ed.). Springer, Dordrecht. pp. 91–112.

Paradiso, A., Berardino, R., de Pinto, M., di Toppi, L. S., Storelli, F. T. and de Gara, L. (2008). Increase in ascorbate-glutathione metabolism as local and precocious systemic responses induced by cadmium in durum wheat plants. *Plant Cell Physiol.* **49**: 362–374.

Parida, A. K. and Das, A. B. (2005). Salt tolerance and salinity effects on plants. *Ecotoxicol. Environ. Saf.* **60**(3): 324–349.

Patade, V. Y., Bhargava, S. and Suprasanna, P. (2009). Halopriming imparts tolerance to salt and PEG induced drought stress in sugarcane. *Agri. Ecosyst. Environ.* **134**: 24-28.

Patakas, A. (2012). Abiotic stress-induced morphological and anatomical changes in plants. In: Ahmad, P. and Prasad, M. N. V. (eds) Abiotic stress responses in plants: Metabolism, productivity and sustainability. Springer, New York, pp 21-39.

Pennisi, E. (2008). The blue revolution, drop by drop, gene by gene. *Science.* **32**: 171–173.

Pervez, M. A., Ayub, C. M., Khan, H. A., Shahid, M. A. and Ashraf, I. (2009). Effect of drought stress on growth, yield and seed quality of tomato (*Lycopersicon esculentum* L.). *Pak. J. Agric. Sci.* **46**: 174–178.

Pettigrew, W.T. (2008). Potassium influences on yield and quality production for maize, wheat, soybean and cotton. *Physiol. Plant.* **133**: 670–681.

Pier, P. A. and Berkowitz, G. A. (1987). Modulation of water stress effects on photosynthesis by altered leaf K+. *Plant Physiol **85***(3): 655-661.

Prasad, P. V. V. and Staggenborg, S. A. (2008). Impacts of drought and/or heat stress on physiological, developmental, growth, and yield processes of crop plants. In: Response of Crops to limited water: understanding and modeling water stress effects on plant growth processes, advances in agricultural systems modeling series 1, Soil Science Society of America, Madison, WI, pp. 301–355.

Pratap, V. and Sharma, V. K. (2010). Impact of osmotic stress on seed germination and seedling growth in black gram (*Phaseolus mungo*). *J. Environ. Biol.* **31**: 721–726.

Premachandra, G.S., Saneoka, H., Fujita, K. and Ogata, S. (2008). Water stress and potassium fertilization in field grown maize (*Zea mays* L.): Effects on leaf water relations and leaf rolling. *J Agro. Crop Sci.***170**:195-201.

Principato, G. B., Rosi, G., Talesa, V., Govannini, E. and Uolila, L. (1987). Purification and characterization of two forms of glyoxalase II from rat liver and brain of Wistar rats. *Biochem. Biophys. Acta.* **911**: 349–355.

Rahman, A., Mostofa, M. G., Nahar, K., Alam, M. M., Hasanuzzaman, M. and Fujita, M. (2015). Calcium mitigates arsenic toxicity in rice seedlings by reducing arsenic uptake and modulating the antioxidant defense and glyoxalase systems and stress markers. *Biomed. Res. Int.* **2015**: 340812.

Rahman, A., Nahar, K., Hasanuzzaman, M. and Fujita, M. (2016). Calcium supplementation improves Na+/K+ ratio, antioxidant defense and glyoxalase systems in salt-stressed rice seedlings. *Front. Plant Sci.* 7: 609. Doi: 10.3389/fpls.2016.00609.

Ramezanpoor, M. and Dastfal, M. (2004). Evaluation bread and durum wheat cultivars tolerance to water stress. Paper presented at the 8rd Iran agronomy and plant breeding conference, University of Rasht, Gilan, Iran.

Rapacz, M., Kos´cielniak, J., Jurczyk, B., Adamska, A. and Wo´ jcik, M. (2010). Different patterns of physiological and molecular response to drought in seedlings of malt- and feed-type barleys (*Hordeum vulgare*). *J .Agron. Crop Sci*. **196**: 9–19.

Rashid, M. A., and Hossain, T. M. (2016). Adoption of wheat varieties in Bangladesh: expert elicitation approach. *Bangladesh J. Agric. Res.* **41**(3): 491-505.

Razaji, A., Farzanian, M. and Sayfzadeh, S. (2014). The effects of seed priming by ascorbic acid on some morphological and biochemical aspects of

rapeseed (*Brassica napus* L.) under drought stress condition. *Int. J. Biosci.* **4**: 432-442.

Reddy, A. R., Chaitanya, K. V. and Vivekanandan, M. (2004). Drought-induced responses of photosynthesis and antioxidant metabolism in higher plants. *J. Plant Physiol.* **161**:1189–1202.

Reguera, M., Peleg, Z. and Blumwald, E. (2012). Targeting metabolic pathways for genetic engineering abiotic stress-tolerance in crops. *Biochem. Biophys. Acta.* **1819**: 186–194.

Restrepo-Diaz, H., Benlloch, M. and Fernández-Escobar, R. (2008). Plant water stress and K+ starvation reduce absorption of foliar applied K+ by olive leaves. *Scientia Hort*. **116**(4): 409-413

Reynolds, M., Dreccer, F. and Trethowan, R. (2007). Drought-adaptive traits derived from wheat wild relatives and landraces. *J. Exp. Bot.* **58**(2): 177–186.

Romero-Puertas, M. C., Corpas, F. J., Sandalio, L. M., Leterrier, M., Rodriguez Serrano, M., del Rio, L. A. and Palma, J. M. (2006). Glutathione reductase from pea leaves: response to abiotic stress and characterization of the peroxisomal isozyme. *New Phytol.* **170**: 43–52.

Sangakkara, U. R., Frehner, M. Nosberger, J. (2001). Influence of soil moisture and fertilizer potassium on the vegetative growth of mungbean (*Vigna radiata* L. Wilczek) and cowpea (*Vigna unguiculata* L. Walp). *J. Agron. Crop Sci.* **186**(2): 73-81.

Saraswathi, S. G. and Paliwal, K. (2011). Drought induced changes in growth, leaf gas exchange and biomass production in Albizia lebbeck and Cassia siamea seedlings. *J. Environ. Biol.* **32**: 173–178.

Schneekloth, J., Bauder, T. and Hansen, N. (2012). Limited Irrigation Mnagement: Principles and Practices.

Selote, D. S. and Khanna-Chopra, R. (2010). Antioxidant response of wheat roots to drought acclimation. *Protoplasma.* **245:** 153–163.

Shahbaz, M., Ashraf, M., Akram, N. A., Hanif, A., Hameed, S., Joham, S. and Rehman, R. (2011). Salt-induced modulation in growth, photosynthetic

capacity, proline content and ion accumulation in sunflower (*Helianthus annuus* L.). *Acta Physiol. Plant.* **33:** 1113-1122.

Shanker, A. and Venkateswarlu, B. (2011). Abiotic stress Response in Plants-Physiological. Biochemical and Genetic Perspectives. Intech Publishers.

Sharma, G. L., Agarwal, R. M. and Singh, R. P. (2006). Potassium induced changes in certain aspects of nitrogen metabolism in chickpea (*Cicer arietinum* L.). *Physiol. Mol. Biol. Plants.* ***12***(2): 157–162.

Sharp, R. E. and Davies, W. J. (1979). Solute regulation and growth by roots and shoots of water-stressed maize plants. *Planta.* **147**: 43–49.

Shehab, G. G., Ahmed, O. K. and El-Beltagi, H. S. (2010). Effects of various chemical agents for alleviation of drought stress in rice plants (*Oryza sativa* L.). *Not. Bot. Hort. Agrobot. Cluj*. **38**: 139–148.

Shi, J. F., Mao, X. G., Jing, R. L., Pang, X. B., Wang, Y. G. and Chang, X. P. (2010). Gene expression profles of response to water stress at the jointing stage in wheat. *Agric. Sci. China.* **9**(3): 325-330.

Siddique, K. H. M., Regan, K. L., Tennant, D. and Thomson, B. D. (2001). Water use and water use efficiency of cool season grain legumes in low rainfall Mediterranean-type environments. *Eur. J. Agron.* **15**: 267–280.

Silva, E.C., Nogueira, R. J. M. C., Azevedo-Neto, A. D. and Santos, V. F. (2003). Comportamento estomático e potencial da água da folha em três espécies lenhosas cultivadas sob estresse hídrico. *Acta Bot. Bras.* **17**:231-246.

Singh, K. and Pandey, S. N. (2011). Effect of nickel-stress on uptake, pigments and antioxidative responses of water lettuce, *Pistia stratiotes* L. *J. Environ. Biol.* **32**: 391–394.

Singh, R. K., Athokpam, K., Singh, H. S. and Nandini, L. N. (2002). Effect on nitrogen, phosphorus and potassium on the growth, yield and nutrient uptake of blackgram (*Vigna mungo* L). *J. Agric. Sci. Soc. North-East India.* **15**(2): 175-177.

Smirnoff, N. (2000). Ascorbic acid: metabolism and functions of a multi-facetted molecule. *Curr. Opin. Plant Biol.* **3**(3): 229-235.

Sorkheha, K., Shirana, B., Rouhia, V., Khodambashia, M. and Sofob, A. (2011) Regulation of the ascorbate–glutathione cycle in wild almond during drought stress. *Russ. J. Plant Physiol.* **58**: 76-84.

Statista (2017). From https://www.statista.com/topics/1668/wheat/ Topic: Wheat. Retrieved 20 December 2017,

Statista, (2018). Largest wheat producing countries worldwide in 2016/2017 Retrieved 10 January 2018, from https://www.statista.com/

Subramanian, K. S., Santhanakrishnan, P. and Balasubramanian, P. (2006). Responses of field grown tomato plants to arbuscular mycorrhizal fungal colonization under varying intensities of drought stress. *Sci. Hort.* **107**: 245–253.

Taiz, L. and Zeiger, E. (2006). Plant Physiology (4th Edn), Sinauer Associates, Massachusetts, 690 pp

Taiz, L. and Zeiger, E. (2010). Plant Physiology, 5th edn. Sinauer Associates Inc. Publishers, Massachusetts.

Tan, J., Zhao, H., Hong, J., Han, y., Li, H. and Zhao, W. (2008). Effects of exogenous nitric oxide on photosynthesis, antioxidant capacity and proline accumulation in wheat seedlings subjected to osmotic stress. *World J. Agric. Sci.* **4**: 307-313.

Thameur, A., Lachiheb, B. and Ferchichi, A. (2012) Drought effect on growth, gas exchange and yield, in two strains of local barley Ardhaoui, under water deficit conditions in southern Tunisia. *J. Environ. Manag.* **113**: 495–500.

Tian, X. R. and Lei, Y. B. (2007). Physiological responses of wheat seedlings to drought and UV-B radiation. Effect of exogenous sodium nitroprusside application. *Russ. J. Plant Physol.* **54**: 676-682.

Tuma, J., Skalicky, M., Tumova, L., Bláhová, P. and Rosulkova, M. (2004). Potassium, magnesium and calcium content in individual parts of *Phaseolus vulgaris* L. plant as related to potassium and magnesium nutrition. *Plant, Soil Environ*. **50**(1):18-26.

Tuteja, N., Gill, S. S. and Tuteja, R. (2012). Improving Crop Productivity in Sustainable Agriculture, Wiley-VCH Verlag GmbH, Weinheim.

USDA, United States department of agriculture. (2017). Bangladesh: Grain and Feed Annual | Foreign Agricultural Service. Retrieved 20 December 2017.

USDA, United States department of agriculture. (2018). World Agricultural Production.
.https://www.fas.usda.gov/psdonline/circulars/production.pdf

Uzilday, B., Turkan, I., Sekmen, A. H., Ozgur, R. and Karakaya, H. C. (2012). Comparison of ROS formation and antioxidant enzymes in *Cleome gynandra* (C4) and *Cleome spinosa* (C3) under drought stress. *Plant Sci.* **182**: 59– 70.

Wahid A. and Rasul E. (2005). Photosynthesis in leaf, stem, flower and fruit, in: Pessarakli M. (Ed.), Handbook of Photosynthesis, 2nd ed., CRC Press, Florida, pp. 479–497.

Waraich, E. A., Ahmad, R., Ashraf, M. Y., Saifullah, and Ahmad, M. (2011). Improving agricultural water use efficiency by nutrient management in crop plants. *Acta Agric. Scandinavica*. **61**(4): 291-304.

Wei, J., Li, C., Li, Y., Jiang, G., Cheng, G. and Zheng, Y. (2013). Effects of external potassium (K) supply on drought tolerances of two contrasting winter wheat cultivars. *PloS one*, **8**(7): p.e69737.

Wild, R., Ooi, L., Srikanth, V. and Münch, G. (2012). A quick: convenient and economical method for the reliable determination of methylglyoxal in millimolar concentrations: the N-acetyl-L-cysteine assay. *Anal. Bioanal. Chem.* **403**: 2577-2581.

Xu, Z. Z., Zhou, G. S. and Shimizu, H. (2009). Are plant growth and photosynthesis limited by pre-drought following rewatering in grass? *J. Exp. Bot.* **60**: 3737–3749.

Xu, Z., Zhou, G. and Shimizu, H. (2010). Plant responses to drought and rewatering. *Plant Signal. Behav.* **5**: 649–654.

Yadav, R. S., Hash, C. T., Bidinger, F. R., Devos, K. M. and Howarth, C. J. (2004). Genomic regions associated with grain yield and aspects of post flowering drought tolerance in pearl millet across environments and tester background. *Euphytica* **136**: 265–277.

Yadav, S. K., Singla-Pareek, S. L., Ray, M., Reddy, M. K. And Sopory, S. K. (2005). Methylglyoxal levels in plants under salinity stress are dependent on glyoxalase I and glutathione. *Biochem. Biophys. Res. Commun.* **337**(1): 61–67.

Yan, H. F., Mao, P.S., Sun, Y. and Li, M. L. (2015). Impacts of ascorbic acid on germination, antioxidant enzymes and ultrastructure of embryo cells of aged *Elymus sibiricus* seeds with different moisture contents.*Int. J. Agric. Biol.* **18**(1): 176-183.

Yokota, A., Kawasaki, S., Iwano, M., Nakamura, C., Miyake, C. and Akashi, K. (2002). Citrulline and DRIP-1 Protein (ArgE Homologue) in Drought Tolerance of Wild Watermelon, *Ann. Bot.* **89**: 825–832.

Yu, C. W., Murphy, T. M. and Lin, C. H. (2003). Hydrogen peroxide induced chilling tolerance in mung beans mediated through ABA-independent glutathione accumulation. *Func. Plant Biol.* **30**(9): 955–963.

Zhang, B., Li, F. M., Huang, G., Cheng, Z. Y. and Zhang, Y. (2006). Yield performance of spring wheat improved by regulated deficit irrigation in an arid area. *Agric. Water Manage.* **79**: 28-42.

Zima, T., Fialová, L., Mestek, O., Janebová, M., Crkovská, J., Malbohan, I., Stípek, S., Mikulíková, L. and Popov, P. (2001). Oxidative stress, metabolism of ethanol and alc ohol-related diseases. *J. Biomed. Sci.* **8**: 59-70.

APPENDICES

Appendix I. Mean square values of plant height, shoot fresh weight, shoot dry weight, chl a, chl b, chl (a+b) of BARI Wheat-21 seedlings as influenced by K treatment at 0 mM, 6 mM and 12 mM under three (well watereded, 50% FC and 20% FC) water regimes

		Mean square values of					
Sources of variation	**df**	**Plant height**	**Shoot FW**	**Shoot DW**	**Chl a**	**Chl b**	**Chl (a+b)**
Replication	2	24.412	0.000	2.51×10^{-04}	0.001	0.003	0.008
Treatment	8	68.699	0.087	2.16×10^{-03}	0.125	0.035	0.287
Error	16	0.391	0.003	3.59×10^{-05}	0.002	0.001	0.002

Appendix II. Mean square values of MDA, H_2O_2, Proline, MG (Methyl Glyoxal), GLY I, GLY II of BARI Wheat-21 seedlings as influenced by K treatment at 0 mM, 6 mM and 12 mM under three (well watereded, 50% FC and 20% FC) water regimes

		Mean square values of					
Sources of variation	**df**	**MDA**	**H_2O_2**	**Proline**	**MG**	**GLY I**	**GLY II**
Replication	2	24.711	0.497	0.002	0.438	0.002	5.538×10^{-06}
Treatment	8	840.894	20.632	0.137	79.625	0.033	9.264×10^{-05}
Error	16	18.090	0.569	0.004	3.175	0.001	6.512×10^{-06}

Appendix III. Mean square values of antioxidant enzymes (CAT, APX, MDHAR, DHAR, GR, GPX) of BARI Wheat-21 seedlings as influenced by K treatment at 0 mM, 6 mM and 12 mM under three (well watereded, 50% FC and 20% FC) water regimes

		Mean square values of					
Sources of variation	**df**	**CAT**	**APX**	**MDHAR**	**DHAR**	**GR**	**GPX**
Replication	2	12.680	0.011	22.395	32.795	80.677	46.870
Treatment	8	1736.420	0.223	136.781	949.010	198.911	2178.550
Error	16	95.860	0.004	3.745	7.116	8.932	33.760

Appendix IV. Mean square values of AsA, DHA, AsA/DHA, GSH, GSSG, GSH/GSSG of BARI Wheat-21 seedlings as influenced by K treatment at 0 mM, 6 mM and 12 mM under three (well watereded, 50% FC and 20% FC) water regimes

		Mean square values of					
Sources of variation	**df**	**AsA**	**DHA**	**AsA/ DHA**	**GSH**	**GSSG**	**GSH/ GSSG**
Replication	2	102347	975695	0.029	17.840	55.880	0.167
Treatment	8	2001032	2610466	0.250	2804.210	1251.420	7.794
Error	16	89063	133124	0.009	95.170	11.090	0.114

Appendix V. Mean square values of Shoot and Root mineral content of BARI Wheat-21 seedlings as influenced by K treatment at 0 mM, 6 mM and 12 mM under three (well watereded, 50% FC and 20% FC) water regimes

		Mean square values of					
Sources of variation	**df**	**Shoot**			**Root**		
		K	**Ca**	**Mg**	**K**	**Ca**	**Mg**
Replication	2	1961.80	558.17	57.70	303.50	861	258.25
Treatment	8	31872.30	6694.43	8490.86	10346.90	6523618	1596.5
Error	16	761.10	486.59	132.46	469.50	22063	102.64

df: degree of freedom

Plates 2. Different phases of plant growth before stress treatment

Step 1. Seed germination on pot filled with sand

Step 2. Greenhouse controlled environment

Step 3. Seedlings at vegetative stage after first nutrient supply

Step 4. Drought imposed in wheat seedlings with K as a protectant