SCREENING OF MUNGBEAN GENOTYPES UNDER PEG INDUCED DROUGHT STRESS CONDITION

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December, 2017

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A Thesis
Submitted to the Institute of Seed Technology,
Sher-e-Bangla Agricultural University, Dhaka,
in partial fulfillment of the requirements
for the degree of

MASTER OF SCIENCE (MS)

IN

SEDD TECHNOLOGY

SEMESTER: JULY-DECEMBER, 2017

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CERTIFICATE

This is to certify that the thesis entitled " ***Screening of Mungbean Genotypes Under PEG Induced Drought Stress Condition"*** *submitted to the Faculty of Agriculture, Sher-e-Bangla Agricultural University, Dhaka, in partial fulfiLment of the requirements for the degree of* ***MASTER OF SCIENCE (MS)*** *in* ***SEED TECHNOLOGY,*** *embodies the results of a piece of bona fide research work carried out by* ***ABDULLAH ALL IMTIAZ,*** *Registration. No.* ***11-04559*** *under my supervision and guidance. No part of this 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:
Dhaka, Bangladesh

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ACKNOWLEDGEMENTS

All praises to the almighty ***Allah*** *for her kindness and infinite mercy in all the endeavors the author to let him successfully complete the research work and the thesis leading to Master of Science.*

The author would like to express his heartiest respect, deep sense of gratitude and sincere, profound appreciation to his supervisor, ***Dr. Md. Abdullahil Baque****, Department of Agronomy, Sher-e-Bangla Agricultural University, Dhaka for his sincere guidance, scholastic supervision, constructive criticism and constant inspiration throughout the course and in preparation of the manuscript of the thesis. Likewise grateful appreciation is conveyed to Co-supervisor,* ***Dr. A. K. M. Ruhul Amin****, Department of Agronomy, Sher-e-Bangla Agricultural University, Dhaka, for his constant encouragement, cordial suggestions, constructive criticisms and valuable advice to conduct the research work as well as preparation of the thesis.*

The author would like to express his deepest respect and boundless gratitude to all the respected teachers of the Department of Seed technology, Sher-e-Bangla Agricultural University, Dhaka, for their scholarly suggestions, constructive criticism, support, encouragement and sympathetic consideration during the course of studies and for providing unforgettable help at the time of preparing the thesis.

The author appreciates the assistance rendered by the stuffs of Seed Technology Department and all stuff of Sher-e-Bangla Agricultural University farm, for their valuable and sincere help in carrying out the research work.

The author wishes to extend his special thanks to Mashud, Keya, Munmun, Shithi, Habib, Kauser and all other friends and all well wishers for their help during experimentation, active encouragement and inspiration to complete this study.

Finally the author is very glad to express his gratefulness and deepest appreciation to his family members and other relatives for their great sacrifices, endless prayers, blessings and support to reach his at this level of higher education.

The Author

Screening of Mungbean Genotypes Under PEG Induced Drought Stress Condition

Abstract

Ensuring food security for over growing population is a major challenge to the agriculturist. More over phenomena like abiotic stresses (especially drought) exert more challenges to fulfill the task. So, screening of drought tolerant crop could be a feasible means to fight the battle. Mungbean is a popular pulse crop in Bangladesh. Screening of drought tolerant mungbean genotypes is time demanding issues as the drought prone areas are increasing exponentially with times. To screen drought tolerance mungbean genotypes, germination, seedling growth, root shoot ratio, coefficient of germination, vigor index and water relation behavior were used as screening criteria. Sixteen mungbean genotypes *viz.* V_1 = BARI Mung 3, V_2 = BARI Mung 4, V_3 = BARI Mung 5, V_4 = BARI Mung 6, V_5 = BARI Mung 7, V_6 = BARI Mung 8, V_7 = BINA Mung 5, V_8 = BINA Mung 6, V_9 = BINA Mung 8, V_{10} =$BMXK_1$-09015-6, V_{11} =BMX-08011-2, V_{12} = BMX-08011-8, V_{13} =$BMXK_1$ -09015-2, V_{14} =$BMXK_1$-09012-1, V_{15} =PM-5 and V_{16} =IPM-02-03 were tested under 5 different PEG concentrations (0%, 5%, 10%, 15% and 20%) at central laboratory, Sher-e-bangla Agricultural University, Dhaka-1207, during February to March, 2018. The experiment was conducted with a complete randomize design (CRD) with 5 replications. The results of the experiment revealed that, germination, seedling growth and water relation behavior of mungbean genotypes varied significantly under different PEG concentration (Drought inducer). A marked reduction of germination, seedling growth and water relation behavior was observed with the increasing of PEG concentration for most of the mungbean genotypes except BINA Mung-6, BARI Mung-4 and BINA Mung-5. The maximum germination percentage (98.12%), root shoot ratio (0.55), relative water content (94.78), water retention capacity (24.98), coefficient of germination (22.27) and vigor index (233.90) were recorded from BINA Mung 6 at 0% PEG concentration. The minimum germination percentage (28.22%), relative water content (25.55), water retention capacity (3.08), coefficient of germination (6.06) and vigor index (13.45) were recorded from IPM-02-03 advanced lines of mungbean at 20% PEG concentration. The minimum root shoot ratio (0.20) at 20% PEG concentration was recorded from BARI Mung 5. BINA Mung 6, BARI Mung 4 and BINA Mung 5 showed consistently better performance against drought stress and there were slow linear reduction were observed with the increasing of PEG concentration from 0% to 20%. So considering the above results, BINA Mung 6, BARI Mung 4 and BINA Mung 5 could be promising drought stress genotypes against moderate drought stress condition.

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LIST OF ACRONYMS

%	= Percent
^{0}C	= Degree Celsius
AEZ	= Agro-Ecological Zone
BARI	= Bangladesh Agricultural Research Institute
BBS	= Bangladesh Bureau of Statistics
BINA	= Bangladesh Institute of Nuclear Agriculture
CG	= Coefficient of germination
CV%	= Percentage of coefficient of variance
cv.	= Cultivar
et al.	=And others
FAO	= Food and Agriculture Organization
LSD	= Least Significant Difference
Max	= Maximum
mg	= Milligram(s)
mm	= Millimeter
Min	= Minimum
NS	= Not significant
PEG	= Poly Ethylene Glycol
RWC	= Relative water content
SAU	= Sher-e-Bangla Agricultural University
VI	= Vigor index
WRC	= Water retention capacity
WSD	= Water saturation deficit
Wt.	= Weight

CHAPTER I

INTRODUCTION

Multiple stresses in natural habitats or fields can affect the plants (Ishag and Mohamed, 1996). Abiotic stresses are constantly creating problems during our efforts for crop improvement. Food productivity has emerged as a major concern worldwide owing to the detrimental effects of abiotic factors. This has necessitated study of stress and minimizing its loss has become essential to ensure the same. Stresses such as drought, salinity, heat and low temperature are responsible for creating considerable loss in world food supply (Farooq *et al.,* 2009). Among abiotic stresses, drought is a major abiotic factor that extensively limits the not only the plant growth and development but also limits the crop productivity (Poltronieri *et al.,* 2011; Dicken and Wright, 2008; Araus *et al.,* 2008; Ramachandra *et al.,* 2004 and Boyer, 1982).

Drought is a meteorological term and is commonly defined as a period without significant rainfall. It strongly determines the natural distribution of plant species. Drought aggravates the impact of the other abiotic or biotic stresses to which plants are exposed. Exposure to this stress reduces germination rate and seedlings growth with significant variations from crop to crop (Hamidi and Safarnejad, 2010). Drought stress either temporarily or permanently but adversely affects a number of morphological, physiological and biochemical processes in crop plants. Deleterious effects of drought stress on crops may include altered plant metabolism in higher plants and maize (Chimenti *et al.,* 2006 and Lawlor and Cornic, 2002), impaired enzyme activities in rice and maize (Xu *et al*., 2008 and Hong and Ji-yun, 2007), reduction in plant biomass of *Brassica* species and pea plants (Arshad *et al.,* 2008 and Ashraf and Mehmood, 1990), reduced solute accumulation in wheat (Khan *et al.,* 1999) or a combination of all these factors. A better understanding of how drought alters plant physiology, biochemistry and gene regulation is vital for improving management practices and breeding efforts in agriculture (Chaves *et al*., 2003). Plant tolerance to drought is a complex phenomenon that includes morphological, physio-biochemical, cellular and molecular responses that facilitate retention or acquisition of water under water deficit (Rampino *et al*., 2006). Osmotic adjustment has been considered to be one of the most crucial processes in a plant's response to water levels

Osmoprotectants including proline, soluble sugars, mannitol, trehalose, ononitol and glycine betaine can decrease the cell's osmotic potential under dehydration (Morgan 1984). It is generally acknowledged that water deficit stress can increase the production in plants of reactive oxygen species [ROS; mainly superoxide anion (O_2^-) and hydrogen peroxide (H_2O_2)] which then leads to oxidative stress, a serious imbalance between production of ROS and antioxidant defenses. The double-bonds of the unsaturated fatty acids in the cell membrane endure the most damage from ROS. Oxidative damage to poly unsaturated fatty acids creates the harmful secondary end product malondialdehyde (MDA), which can be measured as an index of general lipid peroxidation (Moskova *et al.*, 2009). They can be extremely reactive with several cellular constituents such as proteins, lipids and nucleic acids (Hasanuzzaman *et al*., 2013 and Cruz de Carvalho, 2008), which in turn result in negative effects on metabolism and cellular structures (França *et al*., 2007 and Bartels and Sunkar, 2005). To scavenge ROS during stress, plants have evolved a highly efficient antioxidant defense system that includes both non-enzymatic and enzymatic constituents, such as ascorbate (ASC), reduced glutathione (GSH), ascorbate peroxidase (APX), glutathione peroxidase (GPX), superoxide dismutase (SOD), catalase (CAT), and peroxidase (POD). Within the plant, the balance between ROS production and antioxidant defense determines the extent of oxidative damage (Moller *et al.*, 2007). Many factors tilt this balance, including plant genotype and stress intensity and duration.

Plants show simultaneously multigenic responses to tolerate abiotic stresses, which are very difficult to manage. Genes responsible for stress tolerance are either up regulated or synthesized in plants could be helpful to develop drought tolerant plant. Plants transfer signals from roots to other parts for activation of defensive genes (Siopongco *et al.*, 2008; Chaves and Oliveira, 2004 and Ramachandra *et al.*, 2004), including the synthesis and accumulation of substances/ions such as Ca^{+2}, salicylic acid, abscisic acid, jasmonic acid, ethylene that play specific roles in signaling cascades (Liu *et al.*, 2010). Jasmonic acid or abscisic acid signaling pathways activated in plants to upstream the stress responsive genes that lead to production of defense proteins. These proteins activate the defense system of plants for abiotic as well as biotic stresses (Hughes *et al.*, 2009). Proteins like dehydrins and late embryogenesis abundant proteins (LEA proteins) activated in drought stressed plants

for the assimilation of drought tolerance. Only tolerant varieties can accumulate these types of defense proteins in their cells (Hu *et al.*, 2010).

Mungbean [*Vigna radiata* (L.) Wilczek] is one of the most valuable and popular crops of the world, an important component of many major cropping systems (Lambrides and Godwin, 2006). It is also known as green gram or golden gram mainly cultivated in India and others Asian countries. It is highly consumed in sprouts or dry seed form because of its high protein content (Khattak *et al*., 2006). Presence of negligible amount of carbohydrates (4-6 g), fat free and other important vitamins (A, B, C and E) in it makes it popular among vegetarians (Bhatty *et al*., 2000). Mungbean has a major impact on immunity; its regular diet can enhance the immune power. Germinated seeds of mungbean contain anti-carcinogenic, antibacterial and antifungal properties which neutralize the toxicity. Since, it is an important ingredient in several protein supplements and nutraceutical formulations. Therefore, it can be used for the welfare of human beings. It can also be used as a whole or may be processed to bread, noodles, porridge, soups, snacks or even ice-cream (Mogotsi, 2006 and Bhatty *et al*., 2000). It has a fantastic property to fix the atmospheric nitrogen to soil ranging from 30 to 251 kg ha^{-1} (Devendra *et al.*, 2001 and Hoorman *et al*., 2009) by forming symbiotic relation with *Rhizobium* bacteria which also beneficial for the crop succeeding (Ali and Gupta, 2012). Therefore, it also enhances the productivity of soil which promotes the cropping system. It can be used as intercrop or a cover crop in-between two cereal crops due to its short growing period (80-90 days) (Ashour *et al*., 1991). It can be grown under limited amounts of water and poor soil fertility. It is also a valuable green manure, can produce a huge biomass (7.16 t ha^{-1}) (FAO, 2012) and Mungbean straw and byproducts are fairly good and valuable feed for sheep and goats (Khatik *et al.*, 2007), cattle (Vaidya, 2001), poultry (Robinson *et al.*, 2001) and fish (Eusebio and Coloso, 2000).

The production of mungbean is continuously threatened with the expanding drought stressed zones. Drought problems for Mung beans are worsening with the rapid expansion of water stressed areas of the world including 3 billion people by 2030 (Postel, 2000). Crop yield of Mung bean is more dependent on an adequate supply of water than on any other single environmental factor (Kramer and Boyer 1997). One strategy to reduce the effect of water stress on mungbean production is to use

drought tolerant genotypes. This assertion was supported by Siddique *et al.* (2000), who reported that for the purpose of crop production, yield improvement and yield stability under water stress conditions, development of drought tolerant varieties is the best faceable means to overcome this phenomena. Crop plants are usually under stress at one time or another and plant species able to withstand such stresses have great economic potential (Bibi *et al*., 2010). This encourages researchers to know more about mungbean drought tolerance which thereby necessitates a detailed screening of the morphological differences lying beneath. Plants and their response to drought are generally monitored by analysing the basic morphological and physiological parameters, already known as potential indicators of drought tolerance in a huge number of studies germination, seedling growth and water relation behavior (Xu, and Zhou, 2008). Application of polyethylene glycol (PEG), a non-toxic and non penetrating, osmotically active polymer, is a successful laboratory simulation of drought stress (Landjeva *et al*., 2008 and Kocheva *et al*., 2005). In these laboratory tests, the osmotic potential of the medium can be easily controlled and much of the environmental noise associated with field experiments can be avoided (Kocheva *et al*. 2009). Considering this the present work was conducted to screening the drought tolerance mungbean genotypes with the following objectives:

- To study the germination, seedling growth and water relation behavior of mungbean genotypes
- To screen the drought tolerant genotypes of mungbean
- To determine tolerance level to which the tolerant genotypes of mungbean performed better.

CHAPTER II

REVIEW OF LITERATURE

Mungbean is invariable important pulse crop in Bangladesh. Due to its higher protein content it supplements protein requirement to the poor who have no capability to uptake animal protein. Drought stress is one of the major threats for crop production and sustainable agriculture. So, attaining potential production under such abiotic stress (especially drought) condition is ample challenge to the crop producers. To met the national food demand agriculturist must choose some alternate option. Screening of drought stress tolerance genotypes is one of the options that could be a useful option to combat with the adverse situation. Literatures regarding screening of drought stress tolerance mungbean genotypes are scares. Available literatures, pertinent to this study, on different legumes as well as other crops and polyethylene glycol are, therefore, presented below:

2.1 Drought stress impact on crops

Drought, the condition of low water availability to plants is responsible for many detrimental effects that range from physiological to molecular levels. Some of drought stress effects and the extent to which they can affect the plants are discussed below:

Plant water status:

Water is an essential element of plant, which is essential at all stages of plant growth and development. Any reduction in water status of plants resulted in drought stress. Water relations *i.e.* water potential, osmotic potential and turgor potential are important parameters to evaluate drought tolerance in plants. Merah (2001) has characterized relative water content (RWC) an indicator of drought stress in wheat leaves. Water deficit condition highly influenced water relations and resulted in sever drop of plant water status. Grover *et al.,* 2004 stated that, the reduction in relative water content and minimize in water relations immediately after abiotic stress development in plants.

Among water relations, the leaf water potential has reported as a reliable parameter with regard of plant response to drought stress. Water potential significantly

diminished during drought stress in *Brassica* seeds (Singh *et al.,* 1990). Payam (2011) and Sinclair and Ludlow (1985) revealed that, relative water content negatively affected by stresses especially by drought stress is considered as better indicator of water status as compared to water potential attained by plants under drought stress mitigation.

Yürekli *et al.* (2001) has concluded that, the drought stress induced decline in leaf RWC. Similar result also reported by Egilla *et al.* (2005) in rose plants (*Hibiscus rosa-sinensis)*. Decline stomatal conductance in wheat and rice plants has reported to be responsible for maintaining plant water status under drought stress (Abbate *et al.,* 2004 and Siddique *et al.*, 2001).

Photosynthesis:

Drought negatively affects the photosynthetic activity of plant cell that leads to curtail the grain yield. The reduction of photosynthesis in crop plants is one of the reasons for food scarcity (Wahid and Rasul, 2005). During drought stress, the stomatal conductance of plant cell decline that leads to limited CO_2 availability to plant. Due to small concentration of CO_2, rubisco, the key enzyme of photosynthesis acts as oxygenase rather than carboxylase and for consequence over production of reactive oxygen species (ROS). ROS with lots of adverse effects; act as signaling molecules to trigger antioxidant defense systems (Griffiths and Parry, 2002).

Photosynthetic pigments *i.e.* chlorophyll and caratinoides has reported to decline under drought stress. Jaleel *et al.* (2009) reported that, the amount of chlorophyll in plants correlates with the grain yield under stress. Reduction of photosynthesis also outcome in over production of ROS due to which antioxidant shielding system of plants is arrested (Reddy *et al.,* 2004).

Stomatal conductance:

Drought stress adversely affects the photosynthesis a basic process of plant growth and development (Chaves and Oliveira, 2004). The closer of stomata is the prime effect of drought stress that leads to decline transpiration rate and reduced CO_2 influx, which ultimately retarded the Calvin-cycle even at intermediate drought stress level (Horton *et al.,* 1996). Shangguan *et al.* (1999) and Graan and Boyer (1990) revealed

that, stomatal and non-stomatal limitations, both phenomena are accountable for the declining of photosynthesis. Limited carbon uptake in leaves under drought stress is the principle feature of plants that occurs due to stomatal closure (Cornic and Massacci, 1996 and Chaves, 1991).

Concomitant decline in RWC and water potential with reduced photosynthetic rate has revealed in higher plants (Lawlor and Cornic, 2002). Cornic (2000) stated that, generally, reduced stomatal conductance is considered as a prime detrimental effect of reduction of photosynthesis under drought stress. Stomatal closure has found as a character in response to either a decreased leaf turgor and/or water potential in plants, or to a lowered humid environment (Maroco *et al.*, 1997).

Wilkinson and Davies (2002) and Turner *et al.* (2001) reported that, during drought stress, the closer of stomata is a defense mechanism of plant. Plant hormones like abscisic acid and cytokinin are liable for the opening and closing of stomata. However, Yokota *et al.,* (2002) concluded that, due to stomatal closure, stomatal conductance across cell declines and leads to arrest photosynthetic activity.

Reduction of RWC has reported to induce stomatal closure with parallel reduction in photosynthetic rate (Cornic, 2000). Nevertheless, stomata are more efficiently responding to dehydrated roots as compared to plant water status. Dehydrating roots activate the abscisic acid signaling for stomata closure (Davies and Zhang, 1991). Finally, Farquhar *et al.* (2001) and Hubbard *et al*. (2001) concluded that, photosynthesis and stomatal opening has a high degree of correlation.

Oxidative Stress:

Oxidative stress caused by over production of ROS *i.e.* superoxide radical (O^{-}_{2}), singlet oxygen, hydrogen peroxide (H_2O_2) etc. during drought stress is a common phenomena. Singh *et al.* (2012) reported that, ROS produced during drought stress are quenched by the antioxidant defense system of plants. ROS injure the macromolecules (DNA, RNA, Proteins and lipids) and finally impair the plant cell function (Foyer and Fletcher, 2001). Johnson *et al.* (2003) and Asada (1999) also reported that, the detrimental effects of ROS on macromolecules include DNA damage, lipid peroxidation and oxidation of amino acid and proteins.

The damaged macromolecules are either repaired or replaced by de-novo synthesis. However, under severe stresses, massive damage to macromolecules hinders their function that cannot be fixed and finally cell death occurs. During water stress conditions, many scientists (Chaitanya *et al*., 2002; Mano, 2002 and Ramachandra *et al*., 2000) have found development of oxidative stress in plants.

Drought stress induces oxidative stress by excess production of ROS. Although, ROS are normally produce in chloroplast during photosynthesis (Reddy *et al.,* 2004), in mitochondria during electron transport chain (Moller, 2001), in glyoxylate cycle of peroxisomes (Fazeli *et al.,* 2007), and in the plasma membrane (Sairam *et al.,* 2005) but their intensity within cell rise during stress like drought.

Osmotic stress:

Osmotic stress causes in rapid reduction in growth of most of the plants (Ashraf, 2004 and Flowers, 2004). Osmotic stress results in decrease of leaf chlorosis, antioxidants, plant growth, development, and hormonal imbalance (Ashraf *et al*., 2010; Iqbal and Ashraf, 2010; Mittler, 2002 and Munns, 2002) but decrease in growth depends upon the duration and level of stress and plant tissue types (Meloni *et al*., 2003 and Cony and Trione, 1998). Similarly, many scientists have reported the reduced growth of leaves and stems, leaf area, number of tillers, development of new leaves, lateral buds, branches formation and continued root growth under osmotic stress (Munns and Tester, 2008; Munns *et al.,* 2006 and Taiz and Zeiger, 2006). Ashraf and Harris (2004) reported that, decrease in growth usually occurs due to altered biochemical and physiological responses of plants. Reduced plant biomass with parallel decrease in chlorophyll contents, gaseous exchange characteristics and water potential has reported by Noreen *et al.* (2010) in pea plants. Moreover, reduced growth with reduced relative water content, phenolics and malondialdehyde, leaf osmotic potential and antioxidant activities has also reported by Noreen *et al.,* (2010) in turnip. Meloni *et al.,* (2008); Munns and Tester, (2008) and Zhu, (2001) finally concluded that, the ionic imbalance accomplishes the reduced leaf water potential in plants.

Cell membrane injury and lipid peroxidation:

Drought stress severely affects the cell membranes. Cell membrane loses their stability and integrity under drought stress. One measure of cell membrane injury is

the level of lipid peroxidation product MDA that alarmingly rises during drought stress. Raised lipid peroxidation under stress condition crops can be reduced by seed increment treatments from which most economical is seed priming (Yang *et al.,* 2009 and Farooq *et al.,* 2006).

Menconi *et al*. (1995); Baisak *et al*. (1994); Pastori and Trippi (1992) and Dhindsa *et al.* (1981) reported that, drought stress induced decrease in cell membrane stability that indicates the lipid peroxidation actually occurs due to ROS. Decreased cell membrane stability index of leaves with increasing extent of drought stress has reported by Sairam and Saxena (2000).

Germination and yield:

Kaya *et al.* (2006) and Harris *et al.* (2002) reported that, drought stress can result into drastic reduction of germination and growth in crop plants. Drought stress causes many alterations in plant cell development and growth (Hussain *et al.,* 2008 and Kaya *et al.,* 2006). Drought stress has reported to result in reduction of germination and growth in pea (Okcu *et al.,* 2005), alfalfa (Zeid and Shedeed, 2006) and rice seedlings (Manikavelu *et al.,* 2006). Decrease in turgor pressure of cell results in retardant of cell growth under drought stress (Taiz and Zeiger, 2006).

Blum (2009); Munns and Tester (2008); Reynolds and Tuberosa (2008); Bouman (2007) and Rehman *et al.* (2005) reported that, yield decline in crop plants has reported as the major factor of abiotic stresses. About 17 % yield losses in crop plants due to drought stress have recorded by Ashraf *et al.* (2008) and Rehman *et al.* (2005). Drought stress severely affects the yield depending upon stress level, time duration and causes of stress development that result into reduced plant yield (Plaut, 2003). Reduced dry weight in wheat (Wardlaw and Willenbrink, 2000), reduced grain yield and spikes numbers per plant in maize (Cattivelli *et al.,* 2008) and reduced grain yield in soybean, cotton and pearl millet (Pettigrew, 2004; Yadav *et al.,* 2004 and Frederick *et al.,* 2001) have observed under drought stress.

2.2 Polyethylene glycol (PEG) as an inducer of drought/osmotic stress

Osmotic stress is also one of the outcomes of drought stress, usually responded by over cumulation of salts within cell that diminish water status of plant cell. For a

consequence, osmotic stress alters the whole biochemistry of plant cells. Pei *et al.* (2010) reported that, poly ethylene glycol (PEG) is most commonly used to create osmotic stress in plants because it is not naturally produced in the plant tissue nether penetrate into cell from the media. PEG eventually destroys the normal emergence, growth, biochemical attributes and yield of plants including wheat.

Drought is the major constrains faced in rainfed areas so screening of genotypes for drought tolerance is the need of the day, by using physiological traits as a parameter. An investigation was designed out in-vitro and in-vivo conditions by Krishna *et al.* (2018) who used PEG_{6000} as a drought inducer to screen 17 genotypes of mungbean based on seed vigour 7 best genotypes were selected for in-vivo. Polyethylene glycol (PEG) could be used for evaluation of germination potential under variable water conditions since it stops the intake of water molecules and provides a controlled way to impose a physiological drought. Polyethylene glycol (PEG) compounds used to induce osmotic stress in Petri dish (*in vitro*) for plants to maintain uniform water potential during the experimental period. *In vitro* screening for drought tolerance has been proven to be a suitable method to effectively screen large sets of germplasm with good accuracy (Kulkarni and Deshpande, 2007). PEG was used for drought stress induction in mungbean seedling stage and tolerant genotypes were selected (Krishna *et al*., 2018).

Polyethylene glycol (PEG_{-6000}) generates osmotic stress which reduces photosynthetic rate. PEG is mainly used for the determination of the drought stress related information's from the plants (Landjeva *et al*., 2008 and Turkan *et al*., 2005). It is known that PEG does not enter the cell wall space (Rubinstein, 1982) and PEG molecules with a molecular weight greater than 3000 are apparently not absorbed (Tarkow *et al*., 1996). In an investigation conducted by Meher *et al.* (2018), PEG_{-6000} was used as drought inducer osmotic agent. Simulation of drought stress by polyethylene glycol (PEG) induces drought stress on the plants (Jiang *et al*., 1995). It was reported that PEG induced significant water stress in plants and not having any toxic effects (Emmerich and Hardegree, 1990).

Polyethylene glycol widely used to induce water stress in plants is a non-ionic water soluble polymer which is not expected to penetrate into cells (Djibril *et al.,* 2005). Selection for drought tolerance at early stage of seedlings is most frequently practiced

using poly ethylene glycol (PEG $_{6000}$) in the medium (Rana *et al*., 2017 and Rauf *et al.*, 2006).

6The PEG was first time used as an inducer and identifier to screen and select drought resistant tobacco cell lines. Chinese researchers used to do cotton drought evaluation and identification by repeated drought induction method. It is still in the experimental stage to use PEG solution for the identification. PEG$_{-6000}$ was used to establish a rapid and effective cotton-drought tolerance evaluation system for selection and breeding of the drought-tolerant cotton genotypes (Michel and Merrill, 1973). Earlier germination studies have been carried out with aqueous solutions of polyethyleneglycol-$_{6000}$ (PEG-$_{6000}$). Performance of cotton genotypes for drought tolerance using PEG water stress at germination, bud-stage, cotyledon stage and real-leaf stage revealed that at 17% PEG-$_{6000}$ treatment the seedlings growth rate showed inhibition. Physiological quality of cotton cultivar seeds were evaluated in laboratory by the simulation of water potentials with PEG$_{-6000}$ (0.0; -0.2; -0.4; -0.6; -0.8; -1.0 MPa), at 25 ºC using germitest paper as substrate Megha *et al.* (2017).

An investigation was carried out by Vijayakumari and Puthur (2015) to study the ɣ-aminobutyric acid (GABA) priming enhances the osmotic stress tolerance in black piper (*Piper nigrum*) plants subjected to PEG-induced stress. In their study, they used PEG (poly ethylene glycol$_{6000}$; 10 % w/v) as osmotic stress inducer to screen out the drought stress tolerance black piper.

Muscolo *et al.* (2014) and Hohl and Schopfer (1991) stated that, exposure to polyethylene glycol (PEG$_{-6000}$) solutions has been effectively used to mimic drought stress with limited metabolic interferences as those associated to the use of low molecular weight osmolytes that can be taken up by the plant.

Shitole and Dhumal (2012) used PEG$_{-6000}$ as drought stress inducer. Different concentration of PEG$_{-6000}$ (-0.1 bars to -2.0 bars) were used for seed treatment on seed germination and seedling growth of *Cassia angustifolia*.

In 1961 a paper published in 'Science' (Lagerwerff *et al*., 1961) indicated that PEG can be used to modify the osmotic potential of nutrient solution culture and thus induce plant water deficit in a relatively controlled manner, appropriate to experimental protocols. During the 1970's and 1980's PEG of higher molecular

weight (4000 to 8000) was quite commonly used in physiological experiments to induce controlled drought stress in nutrient solution cultures. Several papers also reported theoretical or measured concentration-osmotic potential relations for PEG of different molecular weights (Money, 1989 and Michel, 1983).

2.3 Improvement of drought tolerance capability

Plants perform many of the functions to cope drought stress at physiological, morphological and biochemical grounds. Drought tolerance is the phenomena of better plant growth and development under limited water supply. Plants maintain water relations in proper way (Zhou *et al.,* 2007), shed their leaves to reduce transpiration (DaMatta, 2004), develop extensive and prolific root system to extract water from depth of soils under limited water supply (Kavar *et al.,* 2007) to establish drought tolerance. Screening of drought stress tolerance genotypes could be a feasible means to fight with the current adverse situations. For lab test, germination, seedling growth and water relation behaviors could be prime indicators against drought stress to screening out tolerance genotypes of mungbean. Some of the literatures of different scientist regarding this aspect are discussed below:

Germination percentage (GP)

Krishna *et al*. (2018) carried out an investigation at the field experimentation centre, Department of Genetics and Plant Breeding, Sam Higginbottom University of Agriculture, Technology and Sciences, Allahabad, U.P. during *zaid*-2018 to study the physiological characterization of mungbean genotypes for drought tolerance. There were 17 genotypes of mungbean (*Vigna radiata* L) such asKM-1401, KM-1404, KM-1405, KM-1406, KM-1408, KM-1409, KM-1410, KM-1413, KM-1414, KM-1415, KM-1422, KM-1423, KM-2195, KM-2241, T-44, IPM 02-3, and IPM 02-14. They revealed that, with an increase in water stress (0 – 10%), there was a gradual depletion in rate of water uptake by mungbean seeds of all genotypes thus reduce the germination percentages. The maximum germination percentage (70%) was recorded from KM-1415 and T-44 genotypes where as the minimum germination percentage (10%) was recorded from KM-1408 mungbean genotypes.

The present experiment was performed by Bharadwaj *et al*. (2018) with the objective of studying the morpho-physiological differences in seven mungbean genotypes

namely, PDM 54 (V_1), PDM39 (V_2), IPM99-125 (V_3), PDM11 (V_4), IPM2-14 (V_5), IPM2-3 (V_6) and Pratap (V_7) under water deficit environment. Two treatments had been arranged for each genotype: One set of plants were maintained as control plants with regular watering. T_1 -treatment plants where watering was withdrawn for 10 consecutive days during the vegetative stage which is almost 21 days after germination.T_2-treatment plants where watering was withdrawn for 10 consecutive days during reproductive stage. They reported that, control plants of all the mungbean genotypes maintained a higher plant height as compared to the treated ones. Once water was withdrawn, the genotypes showed a significant though linear decline in plant height during both vegetative and reproductive stages. However, the decline was highest and lowest in genotypes V_3 and V_7 respectively.

Kaur and Bains (2017) conducted a study to screening of mungbean genotypes for drought tolerance using different water potential levels. Twenty five genotypes of summer mungbean *viz.,* SML-1003, SML-837, SML-1136, SML-1361, SML-1002, SML-859, SML-1414, SML-1086, SML-1206, SML-843, SML-1427, SML-1412, SML-668, SML-1073, SML-1205, SML-1164, SML-1165, SML-1178, SML-1023, SML-1360, SML-1411, SML-829, SML-1077, SML-1018 and SML-971constituted the material for the study. Germination test was conducted in Petridishes moistened with each of five water potential treatments viz. 0.0 (control), -0.4, -0.6 and -0.8MPa of Poly Ethylene Glycol (PEG_{-6000}). The result of the experiment revealed that, the extent of reduction in germination varying with the genotypes in the individual treatments. In the lowest water potential i.e. -0.4MPa, five genotypes viz. SML-837, SML-1361, SML-1002, SML-1360, and SML-1411 registered germination above 90 per cent and the rest of the genotypes recorded germination between 47.5 and 87.0 per cent. In reduced water potentials lower than -0.4MPa, significant differences were observed in the germination of genotypes. In -0.6MPa, the germination varied from 25% (SML-837) - to 61% (SML-1411). It was thus, apparent that germination significantly decreased in highly reduced water potentials or increased moisture stress.

Swathi *et al*. (2017) conducted an experiment to screening of mungbean genotypes against water stress mediated through polyethylene glycol. The genotypes were screened for drought tolerance under laboratory conditions in CRD using PEG_{6000} at

different concentrations of -0.3 MPa, -0.6MPa and -0.9MPa. The results of the experiments showed that, the maximum germination percentage (100%) was recorded from most of the genotypes at control (no water stress) where as the minimum germination percentage (0%) was recorded from KM-122, EC- 396117, MH-3-18 and PM110 mungbean genotypes at -0.9MPa water stress.

Laboratory experiment was conducted by Moliehi *et al.* (2017) to screening of common bean cultivars (*Phaseolus vulgaris* L.) for drought tolerance at the National University of Lesotho, Faculty of Agriculture, in the Department of Crop Science. The result of the investigation revealed that, the highest germination percentage was obtained where control (0 PEG) was employed having 95%, followed by 80% where 39g (-0.5bars) PEG was applied. The lowest germination percentage of 37.62% was exhibited in a PEG concentration of 117g (-1.5 bars).

Research conducted by Rana *et al*. (2017) to study the performance of twenty wheat genotypes under Polyethylene Glycol (PEG) induced water stress during germination and early seedling growth stages. The wheat genotypes were tested under three levels of water potential i) Control (Tap water), ii) -2 bars and iii) -4 bar at the Crop Physiology and Ecology Laboratory of Hajee Mohammad Danesh Science and Technology University, Dinajpur during September, 2014 to October, 2014. They reported that, germination percentage was significantly influenced by the interaction effect of water potential levels and wheat genotypes during germination. Germination percentge was higher at control (with a range from 82.66 in BAW 1170 to 98.66 in Satabdi and BARI Gom 27 and a mean of 93.28), moderate at moderate stress (with a range from 77.00 in BAW 1140 to 95.66 in E 34 and a mean of 88.36) and lower at higher water deficit stress (with a range from 63.33 in BAW 1140 to 91.33 in E 34 and a mean of 81.44).

Laboratory experiment was conducted by Megha *et al.* (2017) during 2016 at Agriculture Research Station, Crop physiology division, Dharwad, to evaluation of *Hirsutum* cotton genotypes for water stress using peg_{-6000} by slanting glass plate technique. Study consisted of 19 *Gossypium hirsutum* varieties with two checks. The genotypes were subjected to different osmotic potentials (0.0 MPa (0 bar), - 0.140 MPa (-1 bar) and -0.39 MPa (-3.9 bar)) by slanting glass plate technique. Experimental results showed that final germination percent of *Hirsutum* varieties

significantly affected by PEG_{6000}. The final germination percent of cotton decreased by the increasing of osmotic potential. In distilled water (Control), percentage of seed germination was highest. As the concentration of PEG_{-6000} increases seed germination is restricted. The seed germination percentage decreased as the PEG_{6000} concentration increases from 0% to 20%. Among the PEG concentrations, control (0.00 concentration) recorded significantly higher germination percent (87.6), which was significantly differed with 10 % (76.4) and 20 % (19.0). Whereas the genotypes, Sahana recorded highest (86.7) germination percentage followed by BS-37, LRA-5166, GBHV-177, CCH-12-3 and BS-39 the genotypes such as, RAH-806 recorded less germination percent (20.0) followed by TSH-04/115, CNH-1110, NDLH-1943, NDLH-1938 and RAH-100. The genotype Sahana, BS-37, LRA-5166, ARBH-1357, BS-39 and CCH-12-3 are germinated well under all the PEG concentrations, hence these genotypes were considered as an osmotic stress tolerant.

An experiment was carried out by Rajabpoor and Hajihashemi (2017) with the aim of determining the effect of polyethylene $glycol_{6000}$ (PEG; 0, 3%, 6% and 9% w/v) treatment on six cultivars of alfalfa (*Medicago sativa*) *viz.* Isfahani, Hamedani, Bami, Baghdadi, Yazdi and Ghare-Medicago. The result of the investigation revealed that, the PEG treatment decreased germination percentage significantly in Hamedani and Ghare-Medicago, while PEG treatment had no significant effect on other cultivars. The lowest germination percentage in Hamedani and Ghare-Medicago was observed at 9% of PEG.

Experiment carried out by Hanen and Ahmad (2016) to study the to evaluate the impact of water stress on the germination of the henna plant (*Lawsonia inermis* L.). Seeds were germinated under stress of aqueous Polyethylene Glycol (PEG) solutions blended to create water potentials of 0, -0.2, -0.4, -0.6, - 0.8 and -1 MPa. Analysis of variance for germination percentage data after ten days for *L.inermis* revealed significant differences among different levels of osmotic potential (0 to -1MPa). Germination percentage over 10 days was highest in deionized water control (96%) and there was declined with a decrease in osmotic potential. Germination percentage did not change much under PEG (-0.2 and -0.4 MPa) and the plants showed highest values (88% and 80%, respectively). Polyethylene glycol (-0.6, -0.8 and -1MPa) induced water stress significantly reduced germination percentage by 66%, 84% and

94%, respectively. Inhibition of seed germination was greatest under the osmotic potential, -1MPa.

Muscolo *et al.* (2014) conducted an experiment to investigate the effect of PEG-induced drought stress on seed germination of four lentil genotypes. Seeds of four lentil genotypes (Castelluccio, Eston, Pantelleria, and Ustica) were subjected to five levels (0, 10, 15, 18, and 21%) of polyethylene glycol (PEG_{-6000}) in their study. The results revealed that, water stress reduced seed germination percentage in all cultivars to different extent. Germination was significantly affected by the osmotic potential and by cultivars. The final germination percentage of the control (0% PEG) reached 100% for each cultivar. An increase in PEG stress markedly decreased the germination percentage of all cultivars compared to their relative controls. The germination percentage of Castelluccio and Eston at the highest PEG concentrations (18% and 21%) was higher than that of Pantelleria and Ustica.

Two black gram (*Vigna mungo* L. Hepper) genotypes LBG20 and PU19 were selected Yadav *et al*. (2013) to study the impact of PEG induced drought stress on seed germination, metabolite concentration and activities of antioxidant enzymes. The results revealed that, PEG induced drought stress caused considerable decrease in germination and fresh weight of seedlings in PU 19 and LBG 20 which could be due to stress induced dormancy.

Shitole and Dhumal (2012) conducted an experiment to investigate effect of water stress by polyethylene $glycol_{6000}$ and sodium chloride on seed germination and seedling growth of *Cassia angustifolia*. The results of the experiment revealed that, decreasing osmotic potential (increasing PEG_{6000} concentration) caused reduction in seed germination percentage. The reduction in seed germination was proportional to the increasing concentration of PEG_{6000}. Maximum retardation was noted at highest PEG_{6000} (-2 bars). There was absolute (100%) inhibition of germination above this concentration.

Shoot length (SL)

Krishna *et al*. (2018) carried out an investigation at the field experimentation centre, Department of Genetics and Plant Breeding, Sam Higginbottom University of Agriculture, Technology and Sciences, Allahabad, U.P. during *zaid*-2018 to study the

physiological characterization of mungbean genotypes for drought tolerance. There were 17 genotypes of mungbean such as: KM-1401, KM-1404, KM-1405, KM-1406, KM-1408, KM-1409, KM-1410, KM-1413, KM-1414, KM-1415, KM-1422, KM-1423, KM-2195, KM-2241, T-44, IPM 02-3, and IPM 02-14. They revealed that, drought stress decreased the root length it may be due to declining vacuolar K^+ because its accumulation in newly formed vacuoles drives cell expansion (Walker *et al*., 1998). Same results found in in alfalfa (Safarnezad, 2008).

Kaur and Bains (2017) conducted a study to screening of mungbean genotypes for drought tolerance using different water potential levels. Twenty five genotypes of summer mungbean *viz.*, SML-1003, SML-837, SML-1136, SML-1361, SML-1002, SML-859, SML-1414, SML-1086, SML-1206, SML-843, SML-1427, SML-1412, SML-668, SML-1073, SML-1205, SML-1164, SML-1165, SML-1178, SML-1023, SML-1360, SML-1411, SML-829, SML-1077, SML-1018 and SML-971constituted the material for the study. Germination test was conducted in Petridishes moistened with each of five water potential treatments viz. 0.0 (control), -0.4, -0.6 and -0.8MPa of Poly Ethylene Glycol (PEG_{-6000}). The result of the experiment revealed that, the shoot length in different genotypes was found to be significantly different from one another. The mean shoot length of all genotypes measured 8.95cm and 1.14cm in the control -0.4MPa respectively. It could be seen from the above that there was a sudden fall in the length of shoot from 8.95cm in the control to 1.14cm in -0.4MPa and further reduction of water potential to -0.6MPa and -0.8MPa caused total inhibition in shoot growth in all the tested mungbean genotypes.

Swathi *et al*. (2017) conducted an experiment to screening of mungbean genotypes against water stress mediated through polyethylene glycol. The genotypes were screened for drought tolerance under laboratory conditions in CRD using PEG_{6000} at different concentrations of -0.3 MPa, -0.6MPa and -0.9MPa.The results of the experiments showed that, the maximum shoot length (19 cm) was recorded from ML-267 at control (no water stress) where as the minimum shoot length (0 cm) was recorded from LGG 460, LGG 50 and LGG 407 mungbean genotypes at -0.9MPa water stress.

Laboratory experiment was conducted by Moliehi *et al.* (2017) to screening of common bean cultivars (*Phaseolus vulgaris* L.) for drought tolerance at the National

University of Lesotho, Faculty of Agriculture, in the Department of Crop Science. The result of the investigation revealed that, the overall mean of shoot length for four PEG concentrations recorded 1.343mm with the highest and lowest being 2.182mm and 0.408mm where 10g PEG and 117g were added to the solution, respectively.

Research conducted by Rana *et al*. (2017) to study the performance of twenty wheat genotypes under Polyethylene Glycol (PEG) induced water stress during germination and early seedling growth stages. The wheat genotypes were tested under three levels of water potential i) Control (Tap water), ii) -2 bars and iii) -4 bar at the Crop Physiology and Ecology Laboratory of Hajee Mohammad Danesh Science and Technology University, Dinajpur during September, 2014 to October, 2014. They reported that, shoot length of 10 days old seedling was significantly influenced by the interaction effect of water potential levels and wheat genotypes. The shoot length was found to be higher at control (with a range from 15.43 cm in BAW 1151 to 23.78 cm in Sourav and a mean of 20.81 cm), moderate at moderate stress (ranging from 10.56 cm in BAW 1151 to 18.54 cm in BARI Gom 27 with a mean of 16.00 cm) and lower at higher water deficit stress (with a range from 7.32 cm in BAW 1140 to 15.54 cm in BAW 1138 and a mean of 11.11 cm). The shoot length was found to be reduced with the increment of water deficit stress but the degree of reduction was not similar for all wheat genotypes.

Laboratory experiment was conducted by Megha *et al.* (2017) during 2016 at Agriculture Research Station, Crop physiology division, Dharwad, to evaluation of *Hirsutum* cotton genotypes for water stress using peg$_{-6000}$ by slanting glass plate technique. Study consisted of 19 *Gossypium hirsutum* varieties with two checks. The genotypes were subjected to different osmotic potentials (0.0 MPa (0 bar), - 0.140 MPa (-1 bar) and -0.39 MPa (-3.9 bar)) by slanting glass plate technique. The genotypes were screened for germination percentage, root length, shoot length and seedling vigour traits. The shoot length decreased with the increase in PEG-$_{6000}$ concentrations from 0% to 20%. Under different PEG concentrations, the shoot length were recorded significantly highest in control (7.03 cm) followed by 10% (3.31 cm) and less shoot length was recorded in 20% (0.07 cm). The genotype Sahana recorded significantly higher shoot length (6.10 cm) which was followed by BS-37, LRA-5166, GBHV-177, CCH-12-3, BS-39 and ARBH-1352 (5.48, 5.11, 4.80, 4.65, 4.38 and 4.00

cm, respectively). Whereas, the genotype RAH-806 (0.57 cm) and TSH-04/115 (0.88 cm) followed by CNH-1110, NDLH-1943, NDLH-1938, GSHV-169 and RAH-100were recorded (1.57, 2.02, 2.45, 2.50 and 2.77 cm, respectively) significantly lower shoot length.

An investigation was carried out by Rajabpoor and Hajihashemi (2017) with the aim of determining the effect of polyethylene $glycol_{6000}$ (PEG; 0, 3%, 6% and 9% w/v) treatment on six cultivars of alfalfa (*Medicago sativa*) *viz.* Isfahani, Hamedani, Bami, Baghdadi, Yazdi and Ghare-Medicago. Analysis showed that the seedling length decreased in all cultivars with increasing PEG concentration. With increasing PEG concentration, the highest and lowest reduction in seedling length was observed in Ghare-Medicago and Yazdi cultivars, respectively.

In a study 17 mungbean genotypes were screened for drought tolerance Aslam *et al.* (2013) at seedling stage and to find out best selection criterion against drought conditions. There were three moisture levels *viz.* T_1 =80% of field capacity, T_2=50% of field capacity and T_3=30% of field capacity. They reported that, root length (RL) and shoot length (SL) at three treatment levels (T_1=80%FC; T_2=50%FC; T_3=30%FC) showed that RL and SL at T_1 is more than at T_2 and T_3. At T_3, RL is shorter than other treatments.

Shitole and Dhumal (2012) conducted an experiment to investigate effect of water stress by polyethylene $glycol_{6000}$ and sodium chloride on seed germination and seedling growth of *Cassia angustifolia*. The results of the experiment revealed that, maximum shoot length (5.90 cm) was recorded from control (no drought stress) treatment and the minimum one (0.86 cm) was recorded from highest PEG_{6000} (-2 bars) treatment.

Root length (RL)

Kaur and Bains (2017) conducted a study to screening of mungbean genotypes for drought tolerance using different water potential levels. Twenty five genotypes of summer mungbean *viz.*, SML-1003, SML-837, SML-1136, SML-1361, SML-1002, SML-859, SML-1414, SML-1086, SML-1206, SML-843, SML-1427, SML-1412, SML-668, SML-1073, SML-1205, SML-1164, SML-1165, SML-1178, SML-1023, SML-1360, SML-1411, SML-829, SML-1077, SML-1018 and SML-971constituted

the material for the study. Germination test was conducted in Petridishes moistened with each of five water potential treatments viz. 0.0 (control), -0.4, -0.6 and -0.8MPa of Poly Ethylene Glycol (PEG_{-6000}). The result of the experiment revealed that, the root length in individual genotype was found to be significantly different from one another in the individual treatments. The mean root length of all genotypes measured 8.82cm, 7.53cm, 1.92cm and 0.65cm in the control, -0.4MPa, -0.6MPa and -0.8MPa, respectively.

Swathi *et al*. (2017) conducted an experiment to screening of mungbean genotypes against water stress mediated through polyethylene glycol. The genotypes were screened for drought tolerance under laboratory conditions in CRD using PEG_{6000} at different concentrations of -0.3 MPa, -0.6MPa and -0.9MPa.The results of the experiments showed that, the maximum root length (17.10 cm) was recorded from MH-3-18 at control (no water stress) where as the minimum root length (0 cm) was recorded from LGG 460, LGG 50 and LGG 407 mungbean genotypes at -0.9MPa water stress.

Laboratory experiment was conducted by Moliehi *et al.* (2017) to screening of common bean cultivars (*Phaseolus vulgaris* L.) for drought tolerance at the National University of Lesotho, Faculty of Agriculture, in the Department of Crop Science. The result of the investigation revealed that, the root length had a grand mean of 5.897cm. The longest length of 7.632cm was obtained where PEG concentration is 39g while the shortest length of 2.396cm was found where 117g PEG was applied.

Research conducted by Rana *et al*. (2017) to study the performance of twenty wheat genotypes under Polyethylene Glycol (PEG) induced water stress during germination and early seedling growth stages. The wheat genotypes were tested under three levels of water potential i) Control (Tap water), ii) -2 bars and iii) -4 bar at the Crop Physiology and Ecology Laboratory of Hajee Mohammad Danesh Science and Technology University, Dinajpur during September, 2014 to October, 2014. They reported that, root length of seedling was significantly influenced by the interaction effect of water potential levels and wheat genotypes. The root length was found to be higher at control (with a range from 9.94 cm in BAW 1163 to 13.88 cm in BARI Gom 27 and a mean of 12.00 cm), moderate at moderate stress (ranging from 8.98 cm in BAW 1163 to 12.50 cm in BAW 1171 with a mean of 10.98 cm) and lower at higher

water deficit stress (with a range from 7.42 cm in BAW 1140 to 11.43 cm in E 34 and a mean of 9.46 cm). The root length was found to be reduced with the increment of water deficit stress but the degree of reduction was not similar for all wheat genotypes.

Laboratory experiment was conducted by Megha *et al.* (2017) during 2016 at Agriculture Research Station, Crop physiology division, Dharwad, to evaluation of *Hirsutum* cotton genotypes for water stress using peg_{-6000} by slanting glass plate technique. Study consisted of 19 *Gossypium hirsutum* varieties with two checks. The genotypes were subjected to different osmotic potentials (0.0 MPa (0 bar), - 0.140 MPa (-1 bar) and -0.39 MPa (-3.9 bar)) by slanting glass plate technique. The genotypes were screened for germination percentage, root length, shoot length and seedling vigour traits. Root length was increased with the increasing PEG_{-6000} concentrations up to 10% of PEG_{-6000} concentrations it declined thereafter. The root length differed significantly with respect to PEG_{-6000} concentration of 10% and 20%. Root length was significantly maximum at control, followed by 10 and 20% (5.21, 0.00 and 0.00 cm, respectively), and whereas the maximum root length was observed in 10% followed by control and 20% at (7.37, 6.58 and 0.83 cm, respectively). The root length differed significantly with respect to genotypes. Genotypes, Sahana, BS-37, LRA-5166, GBHV-177, CCH-12-3, BS-39, ARBH-1352 and PH-1060 recorded significantly higher root length (18.69, 17.11, 16.49, 16.03, 15.28, 15.15, 13.73 and 13.31 cm, respectively) than genotypes, RAH-806, TSH-04/115, CNH-1110, NDLH-1943, NDLH-1938, RAH-100 and AKH-09-5 recorded significantly lowest root length (5.34, 5.72, 7.10, 7.40, 7.93, 8.40 and 9.45 cm, respectively).

Muscolo *et al.* (2014) conducted an experiment to investigate the effect of PEG-induced drought stress on seed germination of four lentil genotypes. Seeds of four lentil genotypes (Castelluccio, Eston, Pantelleria, and Ustica) were subjected to five levels (0, 10, 15, 18, and 21%) of polyethylene glycol (PEG_{-6000}) in their study. The results revealed that, water stress reduced root length in all cultivars to different extent. The effects of drought stress and cultivars were also significant on root length. Root length decreased, with the increasing of water stress. By increasing PEG concentrations a different behavior among the cultivars was observed. Eston and Castelluccio showed a greater radicle elongation to Pantelleria and Ustica. The

greatest radicle reduction was observed in Ustica and Pantelleria in presence of PEG at the concentrations of 18 and 21%.

Shitole and Dhumal (2012) conducted an experiment to investigate effect of water stress by polyethylene $glycol_{6000}$ and sodium chloride on seed germination and seedling growth of *Cassia angustifolia*. The results of the experiment revealed that, maximum root length (2.50 cm) was recorded from control (no drought stress) treatment and the minimum one (0.43 cm) was recorded from highest PEG_{6000} (-2 bars) treatment.

Fifteen mungbean (*Vigna radiata* L. Wilczek) genotypes were screened for drought tolerance by Bera (2008) under laboratory condition using PEG_{6000}. He concluded that, shoot length, root length and total length of seedlings were found to reduce in response to moisture stress of -3.0 bar in all the genotypes studied.

Root shoot ratio

In a study 17 mungbean genotypes were screened for drought tolerance Aslam *et al.* (2013) at seedling stage and to find out best selection criterion against drought conditions. There were three moisture levels *viz.* T_1 =80% of field capacity, T_2=50% of field capacity and T_3=30% of field capacity. They reported that, Root shoot ratio decreased with the increase in stress level. AUM-18 and AUM-38 have lowest root shoot ratio at T_1, highest at T_2 and intermediate at T_3. AUM-19 exhibited very slight differences in value at T_1, T_2 and T_3.

Shoot dry weight (SDW)

The present experiment was performed by Bharadwaj *et al.* (2018) with the objective of studying the morpho-physiological differences in seven mungbean genotypes namely, PDM 54 (V_1), PDM39 (V_2), IPM99-125 (V_3), PDM11 (V_4), IPM2-14 (V_5), IPM2-3 (V_6) and Pratap (V_7) under water deficit environment. Two treatments had been arranged for each genotype: One set of plants were maintained as control plants with regular watering. T_1 -treatment plants where watering was withdrawn for 10 consecutive days during the vegetative stage which is almost 21 days after germination.T_2-treatment plants where watering was withdrawn for 10 consecutive days during reproductive stage. They reported that, dry biomass was recorded for all

the mungbean genotypes during drought and after drought recovery. Water stress led to significantly decreased rate of production of dry biomass with the highest decrease in genotype V_2 and the least in genotype V_6 at vegetative stage

Kaur and Bains (2017) conducted a study to screening of mungbean genotypes for drought tolerance using different water potential levels. Twenty five genotypes of summer mungbean *viz.*, SML-1003, SML-837, SML-1136, SML-1361, SML-1002, SML-859, SML-1414, SML-1086, SML-1206, SML-843, SML-1427, SML-1412, SML-668, SML-1073, SML-1205, SML-1164, SML-1165, SML-1178, SML-1023, SML-1360, SML-1411, SML-829, SML-1077, SML-1018 and SML-971constituted the material for the study. Germination test was conducted in Petridishes moistened with each of five water potential treatments *viz*. 0.0 (control), -0.4, -0.6 and -0.8MPa of Poly Ethylene Glycol (PEG_{-6000}). The result of the experiment revealed that, the dry weight of shoot in seedlings of all the tested genotypes ranged between 21mg (SML-859) to 34mg (SML-1086) under normal conditions. The dry weight of shoots decreased from 95.51% to 90.4% in different genotypes at -0.4MPa.

Research conducted by Rana *et al*. (2017) to study the performance of twenty wheat genotypes under Polyethylene Glycol (PEG) induced water stress during germination and early seedling growth stages. The wheat genotypes were tested under three levels of water potential i) Control (Tap water), ii) -2 bars and iii) -4 bar at the Crop Physiology and Ecology Laboratory of Hajee Mohammad Danesh Science and Technology University, Dinajpur during September, 2014 to October, 2014. They reported that, seedling dry weight was significantly influenced by the interaction effect of water potential levels and wheat genotypes (Table 4). The seedling dry weight was higher at control (from 83.66 mg in BARI Gom 28 to 150.66 mg in Satabdi), moderate at moderate stress (from 74.66 mg in BARI Gom 28 to 116.33 mg in Satabdi) and lower at higher water deficit stress (from 67.33 mg in E 24 to 100.00 mg in BARI Gom 25). The seedling dry weight was reduced with the increment of water deficit stress but the degree of reduction was not similar for all wheat genotypes.

An investigation was carried out by Rajabpoor and Hajihashemi (2017) with the aim of determining the effect of polyethylene $glycol_{6000}$ (PEG; 0, 3%, 6% and 9% w/v) treatment on six cultivars of alfalfa (*Medicago sativa*) *viz.* Isfahani, Hamedani, Bami,

Baghdadi, Yazdi and Ghare-Medicago. Analysis of data showed that the levels of dry weight of shoot and root decreased in all cultivars with increasing PEG concentration. Dry matter production showed a significant reduction in Hamedani and Ghare-Medicago cultivars at 9% of PEG treatment.

Laboratory experiment was conducted by Moliehi *et al.* (2017) to screening of common bean cultivars (*Phaseolus vulgaris* L.) for drought tolerance at the National University of Lesotho, Faculty of Agriculture, in the Department of Crop Science. The result of the investigation revealed that, shoot dry weight had a grand mean of 0.01982 with the highest weight of 0.03798 obtained where 39g PEG was dissolved in the solution. The lowest shoot dry weight was 0.000g where 117g PEG was added.

Shitole and Dhumal (2012) conducted an experiment to investigate effect of water stress by polyethylene $glycol_{6000}$ and sodium chloride on seed germination and seedling growth of *Cassia angustifolia*. The results of the experiment revealed that, maximum dry weight (29.00 mg) was recorded from control (no drought stress) treatment and the minimum dry weight (7.00 mg) was recorded from highest PEG_{6000} (-2 bars) treatment.

Root dry weight (RDW)

Kaur and Bains (2017) carried out an investigation to screening of mungbean genotypes for drought tolerance using different water potential levels. Twenty five genotypes of summer mungbean *viz.*, SML-1003, SML-837, SML-1136, SML-1361, SML-1002, SML-859, SML-1414, SML-1086, SML-1206, SML-843, SML-1427, SML-1412, SML-668, SML-1073, SML-1205, SML-1164, SML-1165, SML-1178, SML-1023, SML-1360, SML-1411, SML-829, SML-1077, SML-1018 and SML-971constituted the material for the study. Germination test was conducted in Petridishes moistened with each of five water potential treatments *viz.* 0.0 (control), -0.4, -0.6 and -0.8MPa of Poly Ethylene Glycol (PEG_{-6000}). The result of the experiment revealed that, the variation in root dry weight of seedlings of different genotypes under normal and stressed conditions was observed by the researchers. In controls, the range of root dry weight per seedling varied between 3mg (SML-837) to 8.0mg (SML-1427). At -0.4MPa of water potential the percent reduction in root dry weight varied between 36.66% (SML-1411) to 71.66% (SML-1136) in genotypes.

The magnitude of reduction in root dry weight further increased at -0.6MPa and -0.8MPa. Significant reduction in seedling growth in terms of length, fresh and dry weight of shoot and root among the genotypes might be attributed to their differential response in term of tolerance level to moisture stress.

Laboratory experiment was conducted by Moliehi *et al.* (2017) to screening of common bean cultivars (*Phaseolus vulgaris* L.) for drought tolerance at the National University of Lesotho, Faculty of Agriculture, in the Department of Crop Science. The result of the investigation revealed that, root dry weight had a grand mean of 0.0349g with the highest weight of 0.0558g and lowest weight of 0.0101g obtained from the PEG concentration of 39g and 117g, respectively.

Relative water content (RWC)

Krishna *et al*. (2018) revealed that, among the 17 mungbean genotypes, irrespective of the irrigation and moisture stress treatments, KM-1423 (65.36) and IPM 02-14 (61.76) recorded highest relative water content compared to other genotypes, IPM 02-3(60.50), KM-1422 (60.36), and KM-1409 (59.467) recorded moderate relative water content, whereas KM-1415 (56.96) and KM-2195 (57.400) recorded significantly low relative water content. Relative water content were significantly reduced due to imposition of stress 30% when compared to other treatments.

The present experiment was performed by Bharadwaj *et al*. (2018) with the objective of studying the morpho-physiological differences in seven mungbean genotypes namely, PDM 54 (V_1), PDM39 (V_2), IPM99-125 (V_3), PDM11 (V_4), IPM2-14 (V_5), IPM2-3 (V_6) and Pratap (V_7) under water deficit environment. Two treatments had been arranged for each genotype: One set of plants were maintained as control plants with regular watering. T_1 -treatment plants where watering was withdrawn for 10 consecutive days during the vegetative stage which is almost 21 days after germination.T_2-treatment plants where watering was withdrawn for 10 consecutive days during reproductive stage. They reported that, substantial reduction in RWC of leaves was recorded for all the genotypes under drought while, the highest percentage reduction (54%) was seen in genotype V_3 for both the stages. Significant differences were recorded between the genotypes under control and drought.

Meher *et al.* (2018) conducted an experiment to study the Effect of PEG_{-6000} imposed drought stress on RNA content, relative water content (RWC), and chlorophyll content in peanut leaves and roots. It was clearly evident that severe stress clearly affects the relative water content as compare to the control of same age group plant, the significant differences in RWC was observed as compare to control and stressed of leaf and root (40 days old). The sharp decrease in RWC with the increased PEG concentration was noted of same age group plants.

An experiment was conducted by Chowdhury *et al.* (2017) in a venyl house at the environmental stress site of Bangabandhu Sheikh Mujibur Rahman Agricultural University during September to December, 2012 to know the internal water status under drought stress in soybean genotypes, *viz*. Shohag, BARI Soybean-6, BD2331 (relatively stress tolerant) and BGM2026 (susceptible). The found that, water stress significantly reduced RWC at two sampling times (8:00am and 1:00 pm) across the genotypes at different growth stages in all the four soybean genotypes studied. BARI Soybean-6 had higher RWC than the rest of genotypes and genotype BGM2026 had the lowest RWC at all the three growth stages under both non-stress and stress condition. At 8.00 am, RWC of water stressed plants of Shohag decreased by 9.58, 10.32 and 10.94%, BARI Soybean-6 decreased 9.02, 9.84 and 10.65%, BD2331 decreased 8.90, 11.68 and 12.94%, and BGM2026 decreased 13.90, 15.31 and 16.21% compared to control plants at vegetative, flowering and pod development stages, respectively. At 1.00 pm, RWC of water stressed plants decreased by 11.21, 12.55 and 13.40% in Shohag, decreased 10.79, 11.60 and 13.10% in BARI Soybean-6, 12.48, 14.27 and 18.74 % in BD 2331and 19.22, 21.51 and 25.45% in BGM2026 at three growth stages, respectively. The higher reduction was found in BGM2026 at both the day time.

An investigation was carried out by Rajabpoor and Hajihashemi (2017) with the aim of determining the effect of polyethylene $glycol_{6000}$ (PEG; 0, 3%, 6% and 9% w/v) treatment on six cultivars of alfalfa (*Medicago sativa*) *viz.* Isfahani, Hamedani, Bami, Baghdadi, Yazdi and Ghare-Medicago. The result of the investigation revealed that, the relative water content of six analyzed cultivars significantly decreased with increasing PEG level from 3% to 9%. There were obvious differences among the cultivars in water content value. Comparing different cultivars under PEG treatments,

the highest and lowest water content were observed in Yazdi and Hamedani cultivars, respectively.

An investigation was carried out by Vijayakumari and Puthur (2015) to study the ɣ-aminobutyric acid (GABA) priming enhances the osmotic stress tolerance in black piper (*Piper nigrum*) plants subjected to PEG-induced stress. Two black pepper varieties *viz.*, Panniyur 1 (P_1) and Panniyur 5 (P_5) were selected for the study; P_1 is a known drought-susceptible and P_5 is a drought-tolerant variety (Vijayakumari and Puthur, 2014). They found that, on application of 10 % PEG, black pepper varieties showed visual symptoms of osmotic stress by 10 days of treatment. During further period of stress (15 days), P_1 was severely affected by stress; whereas P_5 showed signs of tolerance up to 15 days (concentration of PEG above 15 % induced severe stress in both varieties and the plants showed signs of death by 15 days. Treatment with PEG, decreased RWC in black pepper plants. The decrease of RWC over control was higher in P_1 (58 %) when compared to P_5 (50 %), on 15 days of stress treatment.

Muscolo *et al.* (2014) conducted an experiment to investigate the effect of PEG-induced drought stress on seed germination of four lentil genotypes. Seeds of four lentil genotypes (Castelluccio, Eston, Pantelleria, and Ustica) were subjected to five levels (0, 10, 15, 18, and 21%) of polyethylene glycol (PEG_{-6000}) in their study. The results revealed that, water stress reduced RWC in all cultivars to different extent. The increase in intensity of osmotic stress caused a gradual decrease in the RWC in each cultivar compared to controls. The presence of PEG at different concentrations differently affected the cultivars. Increasing the RWC decreased in Pantelleria and Ustica and increased in Eston and Castelluccio. The lowest RWC was detected at Ustica and Pantelleria in presence of PEG at the highest (18% and 21%) concentrations.

Water retention capacity (WRC)

An experiment was conducted by Chowdhury *et al.* (2017) in a venyl house at the environmental stress site of Bangabandhu Sheikh Mujibur Rahman Agricultural University during September to December, 2012 to know the internal water status under drought stress in soybean genotypes, *viz.* Shohag, BARI Soybean-6, BD2331 (relatively stress tolerant) and BGM2026 (susceptible). The found that, water stress

decreased the water retention capacity (WRC) significantly which was affected more at noon compared to that at morning. Among the genotypes, the WRC ranged from 6.6 to 7.2 and 6.0 to 7.0 at morning and noon, respectively under non-stress and from 6.0 to 6.3 and 5.1 to 5.5 at morning and noon, respectively under water stress condition. Genotype BGM2026 presented the highest WRC value under non-stress condition but the lowest under water stress condition and decreased considerably at morning (16.66%) and noon (27.14%) while Shohag and BARI Soybean-6 presented the lowest WRC values under non-stress condition. The reduction rate of WRC was minimal which are 7.57% for Shohag, 13.33% for BARI Soybean-6, and 7.57% for Shohag, and 13.11% for BARI Soybean-6 at morning and noon, respectively. The higher reduction in WRC for BGM2026 indicated a greater damage in cell structure due to water stress than Shohag and BARI Soybean-6.

Coefficient of germination (CG)

An investigation was carried out by Rajabpoor and Hajihashemi (2017) with the aim of determining the effect of polyethylene $glycol_{6000}$ (PEG; 0, 3%, 6% and 9% w/v) treatment on six cultivars of alfalfa (*Medicago sativa*) *viz.* Isfahani, Hamedani, Bami, Baghdadi, Yazdi and Ghare-Medicago. The result of the investigation revealed that, the coefficient of germination decreased with increasing PEG concentration. Among the analyzed cultivars, Hamedani and Ghare-Medicago cultivars showed the highest reduction in coefficient of germination at 9% of PEG.

Vigor index (VI)

Krishna *et al*. (2018) found that, mungbean genotypes KM-1409, KM-1415, KM-1423, KM-1422, KM-2195, IPM 02-3 and IPM 02-14 were more tolerant to the drought stress on the basis of seed vigour.

Kaur and Bains (2017) conducted a study to screening of mungbean genotypes for drought tolerance using different water potential levels. Twenty five genotypes of summer mungbean *viz.*, SML-1003, SML-837, SML-1136, SML-1361, SML-1002, SML-859, SML-1414, SML-1086, SML-1206, SML-843, SML-1427, SML-1412, SML-668, SML-1073, SML-1205, SML-1164, SML-1165, SML-1178, SML-1023, SML-1360, SML-1411, SML-829, SML-1077, SML-1018 and SML-971constituted the material for the study. Germination test was conducted in Petridishes moistened

with each of five water potential treatments *viz.* 0.0 (control), -0.4, -0.6 and -0.8MPa of Poly Ethylene Glycol (PEG_{-6000}). The result of the experiment revealed that, the tendency of the highly reduced water potential either to inhibit germination or suppressed the growth and development of seedlings was also noticed for vigour index calculated for different cultivars. The mean vigour index of all genotypes 602.20 in the control significantly decreased to 192.76, 88.76 and 19.28 in -0.4MPa, -0.6MPa and -0.8MPa, respectively. The cumulative vigour index was higher in SML-1411 (294.5).

Swathi *et al*. (2017) conducted an experiment to screening of mungbean genotypes against water stress mediated through polyethylene glycol. The genotypes were screened for drought tolerance under laboratory conditions in CRD using PEG_{6000} at different concentrations of -0.3 MPa, -0.6MPa and -0.9MPa. The genotypes varied significantly for vigour index in all the concentrations. In reduced water potential of -0.3 MPa the vigour index ranged between 476.40 (EC 396117) and 2088.70 (ML 267) with an average mean of 1214.32. Similarly, at -0.6MPa vigour index ranged between 324.00 (WGG 2) and 1393.60 (WGG 37) with an average mean of 804.67. At -0.9 MPa the vigour index index ranged between 136.65 (TM 96-2) and 1425.45 (ML 267) with an average mean of 564.87.

Research conducted by Rana *et al*. (2017) to study the performance of twenty wheat genotypes under Polyethylene Glycol (PEG) induced water stress during germination and early seedling growth stages. The wheat genotypes were tested under three levels of water potential i) Control (Tap water), ii) -2 bars and iii) -4 bar at the Crop Physiology and Ecology Laboratory of Hajee Mohammad Danesh Science and Technology University, Dinajpur during September, 2014 to October, 2014. They reported that, different levels of water potential interacted significantly to wheat genotypes in context of vigor index. Vigor index was found higher at control (ranging from 34.50 in BAW 1163 to 43.62 in BAW 1171 with a mean of 39.61) moderate at moderate stress (ranging from 29.00 in BAW 1163 to 40.86 in E 30 with a mean of 37.09) and lower at higher water deficit stress (with a range from 26.29 in BAW 1163 to 37.96 in BARI Gom 27 and a mean of 34.65).

Laboratory experiment was conducted by Megha *et al.* (2017) during 2016 at Agriculture Research Station, Crop physiology division, Dharwad, to evaluation of

Hirsutum cotton genotypes for water stress using peg_{-6000} by slanting glass plate technique. Study consisted of 19 *Gossypium hirsutum* varieties with two checks. The genotypes were subjected to different osmotic potentials (0.0 MPa (0 bar), - 0.140 MPa (-1 bar) and -0.39 MPa (-3.9 bar)) by slanting glass plate technique. The genotypes were screened for germination percentage, root length, shoot length and seedling vigour traits. The seedling vigour index decreased with the increase in PEG_{-6000} concentrations. The PEG concentrations and genotypes differ significantly with respect to seedling vigor index values. 10% PEG concentration were recorded (2311.8) significantly higher seedling vigor index than control and 20% (1535.3 and 39.25, respectively). The genotype, Sahana was recorded (2422.0) significantly higher seedling vigor index which was followed by BS-37, LRA-5166, GBHV-177, CCH-12-3 and BS-39 (2122.8, 2107.3, 2036.7, 1824.4 and 1790.8, respectively). Whereas, the genotype RAH-806 (87.0) records significantly less seedling vigor index followed by TSH-04/115, CNH-1110, NDLH-1943, NDLH-1938, RAH-100, (227.6, 499.9, 601.5, 718.9 and 832.8, respectively).

Shitole and Dhumal (2012) conducted an experiment to investigate effect of water stress by polyethylene $glycol_{6000}$ and sodium chloride on seed germination and seedling growth of *Cassia angustifolia*. The results of the experiment revealed that, maximum vigor index (624.29) was recorded from control (no drought stress) treatment and the minimum vigor index (53.43) was recorded from highest PEG_{6000} (-2 bars) treatment.

CHAPTER III

MATERIALS AND METHODS

The experiment was conducted during the period from February to March, 2018 to screening of drought tolerant capability of mungbean (*Vigna radiata*) genotypes under drought stress condition. The materials and methods describes a short description of the experimental site, climatic condition of the culture room, experimental materials, treatments and design, methods of the study, data collection procedure and data analysis. The detailed materials and methods that were used to conduct the investigation are presented below under the following headings:

3.1 Description of the experimental site

3.1.1 Location

The experiment was conducted at the Agronomy Laboratory, Department of Agronomy, Sher-e-Bangla Agricultural University (SAU), Sher-e-Bangla Nagar, Dhaka-1207. It was located in 24.090 N latitude and 90.260 E longitudes.

3.1.2 Conditions of laboratory room

The temperature and relative humidity of the laboratory room were recorded daily basis during the study period with a digital thermo hygrometer (TERMO, TFA, Germany). The average minimum and maximum temperature during the study period of the culture room was 25.10 ^{0}C to 28.92 ^{0}C, respectively and average minimum and maximum relative humidity was 56% and 72%, respectively.

3.2 Test crops

The nine mungbean varieties and seven advanced mungbean lines were tested for investigation. BARI Mung-3 to BARI Mung-8 and the advanced mungbean lines were collected from Bangladesh Agricultural Research Institute (BARI) and BINA Mug-5, 6 and 8 were collected from Bangladesh Institute of Nuclear Agriculture (BINA) research station, Mymensing. The collected mungbean varieties were free from any visible defects, disease symptoms and insect infestations and transported to the laboratory of the Department of Agronomy, SAU, Dhaka with careful handling to avoid disease and injury.

3.3 Experimental materials

Different equipments such as electric balance, Petri dish, filter paper, micro pipette, forcep, oven etc. were used for this study.

3.4 Chemicals for seed priming

Different chemicals such as Polyethylene glycol (PEG) ($H(OCH_2CH_2)nOH$) and distilled water were utilized during conducting the experiment. PEG was used inducing drought stress over the mungbean genotypes. 70% Ethanol (CH_3CH_2OH) was use as seed surface sterilization.

3.5 Experimental treatments and design

The experiment comprises of sixteen mungbean genotypes among them nine were released by BARI and BINA and the rest of the genotypes were advanced lines of mungbean. The list of the tested genotypes is given below:

i. V_1 = BARI Mung 3
ii. V_2 = BARI Mung 4
iii. V_3 = BARI Mung 5
iv. V_4 = BARI Mung 6
v. V_5 = BARI Mung 7
vi. V_6 = BARI Mung 8
vii. V_7 = BINA Mung 5
viii. V_8 = BINA Mung 6
ix. V_9 = BINA Mung 8
x. V_{10} =$BMXK_1$-09015-6
xi. V_{11} =BMX-08011-2
xii. V_{12} = BMX-08011-8
xiii. V_{13} =$BMXK_1$ -09015-2
xiv. V_{14} =$BMXK_1$-09012-1
xv. V_{15} =PM-5 and
xvi. V_{16} =IPM-02-03

There were five levels of PEG ($H(OCH_2CH_2)nOH$) concentrations such as 0%, 5%, 10%, 15%, and 20% used as drought inducer for the experiment.

The experiment was laid out in a Completely Randomized Design (CRD) with 5 replications.

3.6 Experimental details

3.6.1 Weight of seeds

Seeds were weighted 100 g from the total seed of for this experiment to reduce the unnecessary loss of seeds.

3. 6. 2 Surface treatments

Seeds were initially treated with 70% solution of Ethanol for 20min for surface sterilization. The sterilized seeds were rinsed 2 min with distilled water for 3 times to reduce the ethanol from the seed surface. Seeds were then dried in room temperature to regain the normal weight.

3.6.3 Preparation of PEG solutions

The PEG solutions were used as drought inducer.

0 g, 12.50 g, 25.00 g, 37.50 g, and 50.00 g of PEG crystal were dissolved in 250 ml of water to prepare 0%, 5%, 10%, 15% and 20% PEG solutions, respectively.

3.6.4 Distilled water

Distilled water was collected from the laboratory of Sher-e-Bangla Agricultural University (SAU).

3.6.5 Germination of seeds

The standard germination test was performed by placing randomly selected 30 seeds in 90-mm-diameter Petri dishes on whatman No.1. Petri dishes containing seeds were irrigated with distilled water and PEG solutions of as per treatments. Here whatman No.1 filter paper were used as growth media for germination. Experimental units (60 Petri dishes for each solution) were arranged in a completely randomized design with five replications. Seeds were kept at room temperature (25-28°C) under normal light to facilitate germination for 8 days. Germination was considered to have occurred when radicle was 2 mm long (Akbari *et al*., 2007). Germination progress was inspected and data were collected at every 24 h intervals and continued up to 8 days.

The seedlings with short, thick and spiral formed hypocotyls and stunted primary root were considered as abnormally germinated seeds (ISTA, 2003). These types of abnormal or dead seedlings were excluded during counting. At the end of germination test (8 days), 5 seedlings from each of the treatments were selected randomly and roots and shoots were cut from the cotyledons and were transferred to brown paper. Then these seedlings were dried in an oven at 75±2°C for 48 hours.

3.7 Procedure of recording data

3.7.1 Germination percentage (GP)

The number of sprouted and germinated seeds was counted daily commencing. Germination was recorded at 24 hrs interval and continued up to 8^{th}. More than 2 mm long plumule and radicle was considered as germinated seed.

The germination percentage was calculated using following formula:

$$\text{Germination percentage (\%)} = \frac{Total\ Number\ of\ germinated\ seeds}{Total\ seed\ placed\ for\ germination} \times 100$$

3.7.2 Shoot length (SL)

The shoot length of five seedlings from each Petri dish was measured finally at 8 DAS. Measurement was done using the unit millimeter (mm) by a meter scale.

3.7.3 Root length (RL)

The Root length of five seedlings from each Ptri dish was recorded finally at 8DAS. Measurement was done using a meter scale and unit was expressed in millimeter (mm).

3.7.4 Dry weight of shoot and root (SDW and RDW)

The dry weight of shoot and root of the five seedlings from each Ptri dish was measured at finally at 8DAS. Dry weight was recorded by drying the sample in an oven at 70°C till attained a constant weight. Then the weight was converted to gram (mg).

3.7.5 Root shoot ratio (Root: Shoot)

The root shoot ratio was recorded on dry weight basis by using the following formula:

Root shoot ratio= $\frac{Root\ dry\ weight\ of\ seedling}{Shoot\ dry\ weight of\ seedling}$

3.7.6 Relative water content (RWC)

Relative water content was measured using following formula:

Relative water content (%) = $\frac{Fresh\ weight - Dry\ weight}{Tugid\ weight - Dry\ weight} \times 100$

3.7.7 Water saturation deficit (WSD)

Water saturation deficit was recorded using following formula:

Water saturation deficit = 100- Relative water content

3.7.8 Water retention capacity (WRC)

Water retention capacity was measured following formula:

Water retention capacity =$\frac{Turgid\ weight}{Dry\ weight}$

3.7.9 Coefficient of germination (CG)

Co- efficient of Germination was calculated using the following formula

Germination Co- efficient (%) = $\frac{A1+A2+\cdots+Ax}{A1T1+A2T2+\cdots+AxTx} \times 100$

Where,

A= Number of seeds germinated

T= Time corresponding to A

x= Number of days to final count

3.7.10 Vigour index (VI)

Vigour index was calculated using following formula:

Vigour index = $\frac{Total\ germination \times Seedling\ length\ (mm)}{100}$

3.8 Statistical Analysis

Data recorded for different parameters were compiled and tabulated in proper form for statistical analysis. CRD analysis was done for statistical test. The data were analyzed using “Analysis of Variance (ANOVA)” technique with the help of computer package program “MSTAT-C” and mean separation among the treatments were done by Least Significance Differences (LSD) test at 1% level of probability as described by Gomez and Gomez (1984).

CHAPTER IV

RESULTS AND DISCUSSION

Present study was undertaken to screen of drought tolerant capability of mungbean genotypes under drought stress conditions. Data on germination, seedling growth and water relation behavior were recorded to find out the potential mungbean genotypes against drought stress condition induced by polyethylene glycol (PEG). The results of the experiment have been presented and discussed in this chapter.

4.1 Germination percentage (%)

Germination percentage of mungbean genotypes were significantly varied due to different concentrations of PEG solution (Table 1). Initially, at 0% PEG concentration most of the mungbean genotypes responded more or less similar in respect of germination percentage except some exceptional. With the advancement of PEG concentration the germination percentage was decreased. But the magnitude of reduction was varied among 16 mungbean genotypes. From 0% to 10% PEG concentration there were a gradual decrease occurred and with increasing concentration from 10% to 20% the reduction was rapid for drought susceptible mungbean genotypes. On the other hand, the drought tolerant genotypes showed more tolerance compare to that of susceptible ones and the magnitude of germination percentage decrease was lower from 0% to 20% PEG concentration. The result revealed that, the maximum germination percentage (98.12%) was recorded from BINA Mung 6 (V_8) which was statistically similar with BARI Mung 4 (V_2), BARI Mung 7 (V_5), BARI Mung 5 (V_3), BINA Mung 5 (V_7), PM-5 (V_{15}) and BARI Mung-6 (V_4) at 0% PEG concentration where as the minimum germination percentage (28.22%) was recorded from IPM-02-03 (V_{16}) mungbean genotype. BINA Mung6 (V_8), BARI Mung 4 (V_2) and BARI Mung 7 (V_5) mungbean genotypes showed consistently better results at all PEG concentrations compare to that of other drought susceptible mungbean genotypes. Germination is one of the most critical periods in the life cycle of plants (Ashraf and Mehmood, 1996). Water is one of the primary requirements in seed for successful germination and when the germinating seed is exposed to drought stress conditions; it compromised the seedling establishment (Albuquerque and Carvalho, 2003). Yang *et al.* (2010) reported that, seed germination

has been the critical stage for species survival. Water absorption is the first stage of germination (Fathi and Tari, 2016). The amount of water absorbed depends on the chemical composition of the seed. Proteins, mucilage and pectin are more hydrophilic colloid and absorb more water than starch (Rahmani, 2006). Drought stress can affect germination rate, however, sensitive to drought stress during different stages of germination and root initiation (Zareian, 2004). Under drought stress the accumulation of proline protected the cell by balancing the osmotic strength of cytosol; preserving protein structures and enzyme activities; scavenging hydroxyl and other free radicals; and regulating cytosol acidity induced by unfavorable environmental conditions which may result retardant of seed germination (Yang *et al.*, 2010). PEG was used by many researchers in inducing drought in seeds and seedling to simulate conditions that exist in the field under dry soil conditions or drought (Moliehi *et al*., 2017; Kuhad *et al*., 1987 and Heikal and Shaddad, 1982). Turhum (1997) emphasized that PEG caused osmotic stress and could be used as a drought inducer. Similar results were obtained by Hu and Jones (2004) and Smok *et al*. (1993) who used PEG to induced osmotic stress on germinating seed and seedlings of cotton, peas, wheat, beans and sorghum. Slower germination of wheat under water deficit stress was found due to lower surface contact of water with seed (Wuest *et al.*, 1999) which restricts the water availability to the seeds (Soltani *et al.*, 2002). Water deficit stress may also lead to degradation and inactivation of the essential hydrolytic and other group of enzymes required for germination (Pratap and Sharma, 2010). Differential degree of sensitivity in potential germination to different water potentials was also found in wheat genotypes. It may be due to genetic variability of wheat susceptible to drought stress. Noorka and Khaliq (2007); Khayantnezhad *et al.* (2010) and Singh *et al.* (2008) also found differential sensitivity in germination among different wheat genotypes in their studies. In a study on pea Okcu *et al*. (2005) stated that, drought stress impaired the germination of five cultivars tested. Fathi and Tari (2016) reported that, the inhibiting action of drought stress on the wheat germination was increased with PEG_{-6000} concentration increasing. Kaur *et al.* (2017) reported that, as far as the highly reduced water potential *i.e*. -0.8MPa was detected to impart huge detrimental effect on germination. The cumulative germination that ranged from 49.17% in SML-1023 to 72.15% in SML-1411 mungbean genotypes and the existence of significant differences for germination in the mungbean genotypes indicated that the physiological means of tolerance to drought stress varied with the

genotypes. Such differences to drought stress in the genotypes would be helpful in identification of genotypes tolerant to drought stress. Similar results also found by Swathi *et al.* (2017); Rana *et al*. (2017); Hanen and Ahmad (2016); Ahmad *et al.* (2015); (Shaban, 2013); Mbarek *et al.* (2013); Aslam *et al.* (2013); Kuar *et al.* (2011); Bibi *et al.* (2010); Jaouadi *et al*. (2010); Zeid and Shedeed (2006); Wang *et al.* (2002); Dirik (2000) and Siddique *et al.* (2000) in mungbean; Mouradi *et al*. (2016); Dutta and Bera (2008) in soyabean; Kosturkova(2008) in pea; Zheng *et al.* (2005) in lentils who reported that the genotypic differences within a species have remarkable potential for crop improvement under water stress conditions.

Table 1. Germination percentage of mungbean genotypes under polyethylene glycol concentrations

Treatments	Germination percentage (%) at different polyethylene glycol (PEG) concentrations				
	0%	5%	10%	15%	20%
V_1	86.23 b-e	80.97 c-f	73.31 e-g	62.52 ef	51.78 ef
V_2	97.75 a	95.91 a	93.89 a	89.07 a	84.84 a
V_3	97.16 a	93.81 ab	87.82 a-c	76.55 c	62.82 cd
V_4	90.32 a-d	85.52 b-e	79.42 c-f	66.35 de	55.46 de
V_5	97.69 a	95.43 a	92.46 ab	87.26 ab	81.76 a
V_6	85.30 c-e	79.76 d-f	75.68 d-g	63.50 e	52.89 ef
V_7	95.69 a	90.45 ab	85.74 a-d	73.75 cd	57.73 c-e
V_8	98.12 a	97.28 a	95.41 a	91.75 a	88.02 a
V_9	84.65 c-e	78.96 ef	70.40 fg	59.35 e-g	45.34 fg
V_{10}	83.43 c-e	77.39 ef	68.77 g	54.54 f-h	39.40 gh
V_{11}	95.29 a	89.74 a-c	85.37 a-d	73.19 cd	61.48 cd
V_{12}	97.16 a	93.78 ab	87.91 a-c	77.37 c	64.34 c
V_{13}	81.86 de	76.45 ef	67.24 g	52.15 gh	36.25 h
V_{14}	92.02 a-c	88.47 a-d	82.96 b-e	74.35 cd	65.08 c
V_{15}	94.49 ab	91.10 ab	85.64 a-d	78.99 bc	73.69 b
V_{16}	79.33 e	75.11 f	65.68 g	48.29 h	28.22 i
LSD $_{(0.01)}$	**8.91**	**9.36**	**10.56**	**8.28**	**7.80**
CV (%)	**4.38**	**4.82**	**5.82**	**5.24**	**5.88**

V_1 = BARI Mung 3, V_2 = BARI Mung 4, V_3 = BARI Mung 5, V_4 = BARI Mung 6, V_5 = BARI Mung 7, V_6 = BARI Mung 8, V_7 = BINA Mung 5, V_8 = BINA Mung 6, V_9 = BINA Mung 8, V_{10} =$BMXK_1$-09015-6, V_{11} =BMX-08011-2, V_{12} = BMX-08011-8, V_{13} =$BMXK_1$ -09015-2, V_{14} =$BMXK_1$-09012-1, V_{15} =PM-5 and V_{16} =IPM-02-03

4.2 Shoot length (mm)

Shoot length of mungbean genotypes significantly influenced by different PEG concentrations (Table 2). Shoot length of mungbean genotype gradually decreased

with the increasing of PEG concentration up to 10%, and there was a rapid reduction observed with increasing the PEG concentration except some drought tolerance genotypes. The result of the experiment revealed that the maximum shoot length (139.40 mm) was recorded from mungbean genotype BINA Mung 6 (V_8) with 0% PEG concentration whereas the minimum shoot length (31.17 mm) was recorded from mungbean genotype IPM-02-03 (V_{16}) with 20% PEG concentration. Mungbean genotype BARI Mung 4 (V_2) and BARI Mung 7 (V_5) performed similar with BINA Mung 6 (V_8) under most of the PEG concentrations but (V_{16}) poorly performed under all the PEG concentrations. The rest of the mungbean genotypes performed intermediate to poor against drought stress condition. Under drought stress, the tolerant plant roots can sign (warning) to send the air to show that they are stress and tension we (root) experience before the leaves. The sign (warning), ABA hormone that is produced as a result of stress in the root tip (Pournajaf, 2005). In this respect, there is general agreement that the most important plant hormone abscisic acid is a major role in the life cycle of plants and many important physiological processes, morphological and plant adaptation to the environment, as well as reactions to adjust the tension (Kafi and Damghani, 1999). Strong evidence indicating that stomatal closure by ABA is an effective means to reduce transpirational water loss. Stress reduction can be one of the major tasks of the ABA. ABA cis-trans, a hormone that is produced in the leaves. In case of lack of water in the root zone and reduce the turgor pressure in the cells of this region, ABA synthesis in roots and aerial parts of the plant quickly spread (Ghodsi *et al*., 1998). Due to the fast reaction of stomatal guard cells during stress (stomatal closure at noon the weather is warm, low water absorption and transpiration rate increases), many scientists believe that the ABA should take place in the near or inside the cells stomatal guard to be able to act quickly so the theory of trans to sis conversion (active form ABA) have raised. Then, ABA through stomatal closure down to the roots and shoots of various genes involved in the function of the genes related to known absisic acid (Ghodsi *et al*., 1998). The accumulation materials such as carbohydrates and amino acids in plant cells that are called compatible solutes are known to play a role under water shortage condition (Ourcut and Nilsson, 2000). Compatible soluble, low molecular weight compounds that interfere with cellular biochemical reactions normally do during osmotic stress, act as guards. In addition to the primary role in osmoregulation of these compounds may have an important role as protecting enzymes and membrane structure and eliminate reactive oxygen species

(ROS) produced as response of drought (Ourcut and Nilsson, 2000). Moisture reduction reactions such as protein degradation and accumulation of free amino acids in order to adjust the osmotic pressure of the cell followed (Bajji *et al*., 2001). In situations where moderate or severe stress, increases the concentration of proline, proline as a nitrogen storage tank or soluble cytoplasmic osmotic potential decrease in acts of plant stress tolerance assists (Ghodsi *et al*., 1998). To prevent water losses, the drought tolerance crop close the stomatal, reducing absorption or decreased transpirational water loss, or a combination of all plant leaves will reduce the amount of transpiration (Shekari, 2000). With increasing water shortages, crop species can clog stomatal pores with the accumulation of ABA. This reduces transpiration and especially when the stomatal are completely blocked and cuticular resistance is much truer. Chaves *et al*. (2003) concluded that, stomatal closure and leaf growth inhibition are the earliest responses to water deficit as the plant attempts to protect itself from extensive water loss. Aslam *et al*. (2013) and Bhatt and Rao, (2005) reported that, shoot length decreased with increase in drought stress level because of inhibition in cell enlargement due to reduced cell turgor; inhibited cellular enlargement results in impartment of shoot growth. Jiang *et al*. (2013) revealed that, the shoot length were decreased by 30% in PEG-treated grass pea compared to those of control, and were decreased by 41% and 57%, respectively, in pea compared to controls. Here, stomatal opening showed a remarkable decrease in both grass pea and pea, which serves to reduce the transpiration rate. Stomatal closure in response to water stress in pea has also been reported by Jackson *et al.* (1987). Kaur *et al*. (2017) stated that, the mean plumule length of all genotypes of mungbean measured 8.95cm and 1.14cm in the control -0.4MPa, respectively. There was a sudden fall in the length of plumule from 8.95cm in the control to 1.14cm in -0.4MPa and further reduction of water potential to -0.6MPa and -0.8MPa caused total inhibition in plumule growth in all the tested mungbean genotypes. These results were also in accordance with the findings of Bharadwaj *et al.* (2018); Swathi *et al.* (2017); Fathi and Tari (2016); Kuar *et al.* (2011) and Dutta *and Bera* (2008) in mungbean; Ranu *et al.* (2005) in mungbean and blackgram.

Table 2. Shoot length of mungbean genotypes under polyethylene glycol concentrations

Treatments	Shoot length (mm) at different polyethylene glycol (PEG) concentrations				
	0%	5%	10%	15%	20%
V_1	91.57 hi	85.70 h	78.82 fg	70.44 f-i	55.32 ef
V_2	137.0 ab	135.8 a	133.8 a	126.7 a	117.1 ab
V_3	121.4 cd	118.0 c	111.5 b	97.89 bc	71.63 d
V_4	106.0 e-h	101.4 d-g	95.04 c-e	81.89 d-g	55.35 ef
V_5	133.1 a-c	132.8 ab	128.5 a	120.4 a	109.8 b
V_6	92.60 g-i	88.32 gh	80.71 fg	69.77 g-i	45.77 fg
V_7	114.3 d-f	110.4 c-e	105.1 bc	95.58 bc	71.01 d
V_8	139.4 a	138.8 a	136.0 a	132.0 a	125.4 a
V_9	101.8 f-i	96.63 e-h	90.53 d-f	76.77 e-h	55.56 e
V_{10}	95.47 g-i	90.07 f-h	83.93 e-g	65.42 h-j	44.79 g
V_{11}	117.3 de	113.2 cd	105.8 bc	91.86 b-d	71.32 d
V_{12}	106.1 e-g	102.2 d-f	94.30 c-e	82.09 d-f	64.28 de
V_{13}	92.17 g-i	88.77 f-h	82.07 e-g	64.04 ij	41.89 g
V_{14}	112.2 d-f	108.4 c-e	101.5 b-d	86.12 c-e	62.57 de
V_{15}	123.4 b-d	119.7 bc	112.5 b	102.3 b	85.30 c
V_{16}	90.12 i	84.04 h	76.09 g	56.58 j	31.17 h
LSD (0.01)	**14.50**	**13.78**	**13.12**	**12.18**	**9.62**
CV (%)	**5.85**	**5.75**	**5.81**	**6.14**	**6.21**

V_1 = BARI Mung 3, V_2 = BARI Mung 4, V_3 = BARI Mung 5, V_4 = BARI Mung 6, V_5 = BARI Mung 7, V_6 = BARI Mung 8, V_7 = BINA Mung 5, V_8 = BINA Mung 6, V_9 = BINA Mung 8, V_{10} =$BMXK_1$-09015-6, V_{11} =BMX-08011-2, V_{12} = BMX-08011-8, V_{13} =$BMXK_1$ -09015-2, V_{14} =$BMXK_1$-09012-1, V_{15} =PM-5 and V_{16} =IPM-02-03

4.3 Root length (mm)

PEG concentration significantly affected the root length of mungbean genotypes (Table 3). The magnitude of reduction of root length was lower in BINA Mung-6, BARI Mung 4 and BARI Mung 7 under different drought stress level. The root length ranged from 99.07 mm in BINA Mung 6 to 54.64 mm in IPM-02-03 at 0% PEG concentration; 97.83 mm in BINA Mung 6 to 48.18 mm in IPM-02-03 at 5% PEG concentration; 95.38 mm in BINA Mung 6 to 40.59 mm in IPM-02-03 at 10 % PEG concentration; 91.70 mm in BINA Mung 6 to 28.31 mm in IPM-02-03 at 15% PEG concentration and finally, 85.78 mm in BINA Mung 6 to 16.50 mm in IPM-02-03 at 20% PEG concentration. Mungbean genotype BARI Mung 4 and BARI Mung 7 showed statistically similarity with BINA Mung 6 at 0% to 10% PEG concentrations; at 15% and 20% PEG concentrations BARI Mung 4 showed statistically at par with BINA Mung 6. On the other hand, IPM-02-03 showed consistently poor performance

against all levels of drought stress which was statistically at par with $BMXK_1$-09015-6 at 0% to 20% PEG concentrations and the rest of the genotypes performed intermediate to poor. With increasing severity of drought, the root length was reduced in millet (Fathi and Tari, 2016). Drought has affected many aspects of plant growth and retarded the root growth (Shekari, 2000). Jiang *et al.* (2013) found that, the root length was decreased by 30% in PEG-treated grass pea compared to those of control, and were decreased by 41% and 57%, respectively, in pea compared to controls. Kaur *et al.* (2017) reported that, the mean root length of all genotypes of mungbean was 8.82cm, 7.53cm, 1.92cm and 0.65cm in the control, -0.4MPa, -0.6MPa and -0.8MPa, respectively. So, it may be concluded that, for drought stress sensitive mungbean genotypes, the root length decreased with increasing the drought condition induced by PEG.

Table 3. Root length of mungbean genotypes under polyethylene glycol concentrations

Treatments	Root length (mm) at different polyethylene glycol (PEG) concentrations				
	0%	5%	10%	15%	20%
V_1	72.21 f-h	68.53 e-h	64.91 d-f	58.40 ef	47.78 de
V_2	97.23 ab	95.17 a	93.92 a	87.42 ab	81.35 ab
V_3	76.08 e-g	72.56 d-f	67.10 c-e	61.29 c-e	51.43 d
V_4	66.97 gh	62.62 gh	57.42 fg	46.12 gh	37.37 fg
V_5	93.37 a-c	90.71 ab	87.34 a	82.66 b	76.38 b
V_6	85.09 c-e	83.25 bc	77.75 b	69.07 c	58.08 c
V_7	68.48 gh	65.06 f-h	58.70 e-g	50.84 fg	38.31 fg
V_8	99.07 a	97.83 a	95.38 a	91.70 a	85.78 a
V_9	74.56 f-h	70.95 e-g	63.95 d-f	52.10 fg	36.05 f-h
V_{10}	64.84 hi	60.75 h	53.94 g	42.64 h	30.03 h
V_{11}	80.07 d-f	75.86 c-e	68.28 cd	61.06 de	47.28 de
V_{12}	79.37 d-f	75.22 c-e	66.03 d-f	53.69 efg	41.92 ef
V_{13}	55.19 ij	50.49 i	41.64 h	30.04 i	19.43 i
V_{14}	69.46 gh	64.83 f-h	58.08 fg	47.77 gh	33.67 gh
V_{15}	87.48 b-d	82.07 b-d	75.51 bc	68.88 cd	60.46 c
V_{16}	54.64 j	48.18 i	40.59 h	28.31 i	16.50 i
LSD $_{(0.01)}$	**9.90**	**9.79**	**8.93**	**7.97**	**6.63**
CV (%)	**5.79**	**6.01**	**5.97**	**6.12**	**6.23**

V_1 = BARI Mung 3, V_2 = BARI Mung 4, V_3 = BARI Mung 5, V_4 = BARI Mung 6, V_5 = BARI Mung 7, V_6 = BARI Mung 8, V_7 = BINA Mung 5, V_8 = BINA Mung 6, V_9 = BINA Mung 8, V_{10} =$BMXK_1$-09015-6, V_{11} =BMX-08011-2, V_{12} = BMX-08011-8, V_{13} =$BMXK_1$ -09015-2, V_{14} =$BMXK_1$-09012-1, V_{15} =PM-5 and V_{16} =IPM-02-03

4.4 Shoot dry weight (mg)

Dry matter is the plant materials dried to a constant weight. Dry matter accumulation could be an efficient indicator for growth of vigour plant which has the capability of storing optimum photosynthates from source to sink even under stress condition (Drought stress). PEG concentration significantly affected the shoot dry weight of mungbean genotypes (Table 4). BINA Mung-6, BARI Mung 4 and BARI Mung 7 mungbean genotypes showed consistently slower reduction for shoot dry weight with the increasing of drought stress induced by PEG concentration. Maximum shoot dry weight was reported from BINA Mung 6 genotypes followed by BARI Mung 4 and BARI Mung 7 at all the PEG concentrations whereas IPM-02-03, $BMXK_1$-09015-2 and BMX-08011-8 mungbean genotypes showed more sensitivity to drought stress condition and produced lowest shoot dry weight. Therefore, BINA Mung-6, BARI Mung 4 and BARI Mung 7 showed promising performance against drought stress condition in terms of shoot dry weight. Water shortage, with the disappearance of inflammatory cells, disrupted physiological processes, leaf growth, photosynthesis, stomatal closure, and changes in metabolism, drying and dying plants (Rahmani, 2006). The main possible causes of reduction of dry weight in vegetative stage under drought stress condition can be a real photosynthesis. Chlorophyll is one of the major chloroplast components for photosynthesis (Rahdari *et al*., 2012). The decrease in chlorophyll content under drought stress has been considered a typical symptom of pigment photo oxidation and chlorophyll degradation. Decrease of chlorophyll content during drought stress depending on the duration and severity of drought level (Zhang and Kirkham, 1996). A decrease of total chlorophyll content with drought stress implies a lowered capacity for light harvesting. Since the production of reactive oxygen species is mainly driven by excess energy absorption in the photosynthetic apparatus, this might be avoided by degrading the absorbing pigments (Mafakheri *et al*., 2010) and ultimately reduced the accumulation of phosynthates and dry weight of seedling. Aslam *et al.* (2013) and Lisar *et al.* (2012) reported that, low moisture stressed leaves become unable for gaseous exchange, stomatal conductance, photosynthesis, light interception, carbohydrate synthesis and food translocation which ultimately is responsible for reduced dry matter. Decline in leaf water potential and relative water contents are amongst the reasons for decrease in photosynthesis activity in plant leaves. Quantitative and qualitative changes in photosynthetic

pigments, reduced CO2 uptake, oxidative damage by reactive oxygen species (ROS) and poor rate of assimilates translocation are the factors responsible for inhibited photosynthesis and for consequence reduce the dry weight (Lisar *et al.*, 2012). Moliehi *et al.* (2017) narrated that, PEG absorbed more water since it had high osmotic potential. Osmotic stress delayed the emergence of radicle and further development of seedling. These results were comparable with the findings of Turhum (1997). They indicated that decline in the seedling development was due to osmotic potential of PEG or ionic effects or a combination of both. PEG reduced osmotic potential of the external medium and decreases water availability for germinating seed. Rana *et al*. (2017) stated that, water deficit stress developed by PEG reduced the shoot length and dry weight of wheat genotypes. Water stress also reduced the seed reserve utilization solubilization of sugars during germination which contributed to lower seedling dry weight of wheat (Soltani *et al*., 2006 and Harb, 2013). Jiang *et al.* (2013) reported that, the shoot dry weight in PEG-treated grass pea were also decreased by 30% compared to untreated controls, whereas they were more than 65% reduced in PEG-treated pea compared to PEG-treated grass pea. Kaur *et al.* (2017) revealed that, decreasing water potential by PEG caused a remarkable reduction in dry weight of plumule and radicle in mungbean. The dry weight of shoots decreased from 95.51% to 90.4% in different genotypes of mungbean at -0.4MPa. Again, Dutta and Bera (2008) reported that, Pusa-9531 and K-851 showed least magnitude of reduction on total seedling dry weight, respectively whereas maximum reduction in these parameters was observed in B-1 and PDM-84-139 mungbean genotypes, respectively under drought stress condition. Significant variation in seedling dry weight among the genotypes might be attributed to their differential response in terms of tolerance level to drought stress. Similar results were found by Bharadwaj *et al.* (2018); Fathi and Tari (2016); Almaghrabi (2012); Raza *et al.* (2012); Khakwani *et al.* (2011); Jajarmi (2009); Datta and Bera (2008) and Wullschleger (2005) who were of opinion that the genotypes which performed better under osmotic stress in terms of lesser reduction in various aspects of growth might be related to their drought tolerance.

Table 4. Shoot dry weight of mungbean genotypes under polyethylene glycol concentrations

Treatments	Shoot dry weight (mg) at different polyethylene glycol (PEG) concentrations				
	0%	5%	10%	15%	20%
V_1	13.60 c-e	11.85 f-h	9.43 fg	7.88 f-h	4.26 d-f
V_2	22.06 a	21.64 a	20.52 ab	20.08 ab	17.64 a
V_3	20.48 a	18.54 cd	16.31 c	14.43 c	8.74 c
V_4	15.36 bc	13.60 ef	11.20 de	9.43 de	4.01 ef
V_5	21.24 a	20.53 ab	19.08 b	18.59 b	16.4 b
V_6	14.16 cd	12.43 e-g	10.59 d-f	8.24 e-g	3.74 ef
V_7	16.30 b	14.19 e	11.64 d	8.58 ef	4.51 de
V_8	22.32 a	22.21 a	21.36 a	21.03 a	18.51 a
V_9	13.95 cd	11.70 f-h	9.66 e-g	7.47 f-h	3.34 f-h
V_{10}	12.44 d-f	10.69 gh	9.22 fg	7.05 gh	2.49 g-i
V_{11}	13.75 cd	11.40 gh	10.06 d-g	8.07 e-g	3.74 ef
V_{12}	12.70 d-f	11.35 gh	9.65 e-g	7.49 f-h	3.52 e-g
V_{13}	11.69 ef	10.24 h	8.54 g	6.51 h	2.33 hi
V_{14}	20.55 a	16.79 d	15.30 c	10.75 d	5.27 d
V_{15}	20.72 a	18.77 bc	15.63 c	12.95 c	8.03 c
V_{16}	11.48 f	10.20 h	8.363 g	6.38 h	2.21 i
LSD (0.01)	**1.99**	**1.99**	**1.72**	**1.52**	**1.05**
CV (%)	**5.42**	**6.02**	**5.97**	**6.20**	**6.93**

V_1 = BARI Mung 3, V_2 = BARI Mung 4, V_3 = BARI Mung 5, V_4 = BARI Mung 6, V_5 = BARI Mung 7, V_6 = BARI Mung 8, V_7 = BINA Mung 5, V_8 = BINA Mung 6, V_9 = BINA Mung 8, V_{10} =$BMXK_1$-09015-6, V_{11} =BMX-08011-2, V_{12} = BMX-08011-8, V_{13} =$BMXK_1$ -09015-2, V_{14} =$BMXK_1$-09012-1, V_{15} =PM-5 and V_{16} =IPM-02-03

4.5 Root dry weight (mg)

PEG concentration significantly influenced the root dry weight of mungbean genotypes (Table 5). The magnitude of reduction of root dry weight was lower in BINA Mung-6, BARI Mung 4 and BARI Mung 7 under different levels drought stress. The root dry weight ranged from 12.26 mg in BINA Mung 6 to 3.30 mg in IPM-02-03 at 0% PEG concentration; 11.64 mg in BINA Mung 6 to 3.02 mg in IPM-02-03 at 5% PEG concentration; 10.69 mg in BINA Mung 6 to 2.72 mg in IPM-02-03 at 10 % PEG concentration; 10.12 mg in BINA Mung 6 to 1.91 mg in IPM-02-03 at 15% PEG concentration and finally, 8.34 mg in BINA Mung 6 to 0.49 mg in IPM-02-03 at 20% PEG concentration. Mungbean genotype BARI Mung 4 showed statistically similarity with BINA Mung 6 at 0% PEG concentrations. On the other hand, IPM-02-03 showed consistently poor performance against all levels of drought stress which was statistically at par with $BMXK_1$ -09015-2 and $BMXK_1$-09015-6 at

0% to 20% PEG concentrations and the rest of the genotypes performed intermediate to poor. Fathi and Tari (2016) reported that, in alfalfa (*Medicago sativa*), root dry weights was reduced by polyethylene glycol-induced water deficit. Jiang *et al.* (2013) reported that treatment with 20% PEG (−0.53 M pa) for 5 days caused a drastic reduction of root weight in both grass pea and garden pea seedlings. The obtaining results of our study were in accordance with the findings of Kaur *et al.* (2017) and Dutta and Bera (2008) who concluded that, the root dry weight of drought stress sensitive mungbean genotypes was drastically reduced under drought stress condition but in case of comparatively drought stress tolerant ones the magnitude of reduction was slower.

Table 5. Root dry weight of mungbean genotypes under polyethylene glycol concentrations

Treatments	Root dry weight (mg) at different polyethylene glycol (PEG) concentrations				
	0%	5%	10%	15%	20%
V_1	4.10 f-h	3.74 gh	3.57 gh	3.15 f	1.49 ef
V_2	11.69 a	10.83 b	10.04 b	9.43 b	7.58 b
V_3	4.48 e-g	4.25 fg	3.84 e-g	3.32 ef	1.75 e
V_4	5.48 cd	5.32 d	3.94 e-g	3.02 fg	1.20 fg
V_5	10.56 b	9.83 c	8.77 c	7.99 c	6.47 c
V_6	5.83 c	4.59 ef	3.50 g-i	2.47 hi	1.00 gh
V_7	5.39 cd	5.24 de	3.84 e-g	2.83 f-h	1.09 gh
V_8	12.26 a	11.64 a	10.69 a	10.12 a	8.34 a
V_9	4.17 f-h	3.98 f-h	3.78 fg	2.49 hi	1.10 gh
V_{10}	3.49 hi	3.30 hi	3.09 h-j	2.56 g-i	0.90 gh
V_{11}	5.24 c-e	3.75 gh	3.13 h-j	2.18 ij	1.05 gh
V_{12}	4.51 e-g	4.40 fg	4.21 ef	2.33 h-j	0.99 gh
V_{13}	3.76 g-i	3.39 hi	2.93 ij	2.22 ij	0.83 hi
V_{14}	4.85 d-f	4.61 ef	4.42 e	3.75 e	1.52 ef
V_{15}	5.62 cd	5.42 d	5.28 d	4.52 d	2.95 d
V_{16}	3.30 i	3.02 i	2.72 j	1.91 j	0.49 i
LSD (0.01)	**0.80**	**0.70**	**0.60**	**0.50**	**0.35**
CV (%)	**6.04**	**5.73**	**5.52**	**5.54**	**6.58**

V_1 = BARI Mung 3, V_2 = BARI Mung 4, V_3 = BARI Mung 5, V_4 = BARI Mung 6, V_5 = BARI Mung 7, V_6 = BARI Mung 8, V_7 = BINA Mung 5, V_8 = BINA Mung 6, V_9 = BINA Mung 8, V_{10} =$BMXK_1$-09015-6, V_{11} =BMX-08011-2, V_{12} = BMX-08011-8, V_{13} =$BMXK_1$ -09015-2, V_{14} =$BMXK_1$-09012-1, V_{15} =PM-5 and V_{16} =IPM-02-03

4.6 Root shoot ratio

Root shoot ration of mungbean genotypes significantly influenced by PEG concentrations (Table 6). The result of the experiment revealed that, root shoot ratio

ranged from 0.55, 0.52, 0.50, 0.48 and 0.45 in V_8 to 0.22, 0.23, 0.24, 0.23 and 0.20 in V_3 were recorded at 0%, 5%, 10%, 15%, 20% PEG concentrations, respectively. Mungbean genotype V_8 was similar with V_2 at all the PEG concentrations; with V_5 at 0%, 5% and 20% PEG concentrations and V_3 was similar with V_{14}, V_{15}, V_{10} and V_{16} at 0%; V_{14}, V_{15} and V_{16} at 5% and V_{16}, V_7 and V_6 at 20% PEG concentrations. This findings was not coincide with the findings of Aslam *et al.* (2013) who reported that, root shoot ratio decreased with the increase in drought stress level. But in our study, mungbean responded variably to low moisture stress regarding root shoot ratio. Decrease in root shoot ratio under stress is the indication of stunted growth of roots whereas increase in root shoot ratio indicates elongation of roots more relative to shoots to explore deeper soil foils for water absorption. Studies correlated the increase in root shoot ratio with high ABA level of roots and shoots (Lisar *et al.*, 2012).

Table 6. Root shoot ratio of mungbean genotypes under polyethylene glycol concentrations

Treatments	Root shoot ratio at different polyethylene glycol (PEG) concentrations				
	0%	5%	10%	15%	20%
V_1	0.30 d-f	0.32 c-e	0.38 d	0.40 c	0.34 c-f
V_2	0.53 a	0.50 a	0.49 a	0.47 a	0.43 ab
V_3	0.22 g	0.23 f	0.24 i	0.23 j	0.20 i
V_4	0.36 b-d	0.39 b	0.35 e	0.32 f-h	0.30 d-g
V_5	0.50 a	0.48 a	0.46 b	0.43 b	0.40 a-c
V_6	0.41 b	0.37 b-d	0.33 fg	0.30 h	0.25 g-i
V_7	0.33 c-e	0.37 b-d	0.33 fg	0.33 efg	0.24 g-i
V_8	0.55 a	0.52 a	0.50 a	0.48 a	0.45 a
V_9	0.30 d-f	0.34 b-e	0.39 d	0.33 efg	0.33 c-f
V_{10}	0.28 e-g	0.31 de	0.33 e-g	0.36 d	0.36 b-d
V_{11}	0.38 bc	0.33 b-e	0.31 g	0.27 i	0.28 f-h
V_{12}	0.35 b-d	0.39 bc	0.44 c	0.31 gh	0.29 f-h
V_{13}	0.32 c-e	0.33 b-e	0.34 ef	0.34 d-f	0.36 b-e
V_{14}	0.24 fg	0.27 ef	0.29 h	0.35 de	0.29 e-h
V_{15}	0.27 e-g	0.29 ef	0.34 ef	0.35 de	0.37 b-d
V_{16}	0.29 d-g	0.30 ef	0.33 fg	0.30 h	0.22 hi
LSD (0.01)	**0.07**	**0.07**	**0.02**	**0.02**	**0.07**
CV (%)	**5.72**	**5.54**	**5.58**	**5.85**	**6.29**

V_1 = BARI Mung 3, V_2 = BARI Mung 4, V_3 = BARI Mung 5, V_4 = BARI Mung 6, V_5 = BARI Mung 7, V_6 = BARI Mung 8, V_7 = BINA Mung 5, V_8 = BINA Mung 6, V_9 = BINA Mung 8, V_{10} =$BMXK_1$-09015-6, V_{11} =BMX-08011-2, V_{12} = BMX-08011-8, V_{13} =$BMXK_1$ -09015-2, V_{14} =$BMXK_1$-09012-1, V_{15} =PM-5 and V_{16} =IPM-02-03

4.7 Relative water content (%)

RWC could be the perfect indicator of plant hydrologic condition as it denotes the physiological consequences of cellular water deficit and metabolic activity of leaf. Water potential that possess the energy status of plant water which is useful for the transportation of water in the soil-plant-atmosphere chain. A wide range of statistical difference was observed for the relative water content of wheat genotypes under different PEG solutions (Table 7). Corresponding water content followed the similar trend as the previous parameters of mungbean genotypes. The results of the experiment revealed that the relative water content ranged from 94.78% in BINA Mung 6 to 74.44% in IPM-02-03 at 0% PEG concentration; 91.71% in BINA Mung 6 to 67.10 % in IPM-02-03 at 5% PEG concentration; 88.84% in BINA Mung 6 to 54.18% in IPM-02-03 at 10% PEG concentration; 84.20% in BINA Mung 6 to 41.84% in IPM-02-03 at 15% PEG concentration and finally, 80.94% in BINA Mung 6 to 25.55% in IPM-02-03 at 20% PEG concentration. BINA Mung-6, BARI Mung 4 and BARI Mung 7 performed distinctly superior to IPM-02-03, $BMXK_1$-09015-2 and $BMXK_1$-09015-6 where rest of the genotypes gave the intermediate results under all PEG concentration. Parsons and Howe (1984) opined that among several methods used to characterize internal plant water status under drought conditions, RWC is an integrative indicator. Relative water content is the parameter that is affected by the drought resulting in decrease in cell size (Hayatu *et al*., 2014). The content of proline and sugar justifies the lower RWC in sensitive mungbean genotypes as the plant is unable to draw enough water from the soil. Plants grown under water stress conditions showed a lower RWC than those grown under non stress conditions. Schonfeld *et al.* (1988) reported that the cultivars that were resistant to drought had more RWC. Upreti *et al.* (2000) reported that sensitive pea genotypes were more affected by a decline in relative water content than tolerant ones under drought stress condition. This might be due to the comparatively tolerant genotypes a higher capability for soil water extraction under drought stress. Jiang *et al.* (2013) and Turner (1981) reported that, RWC correlates well with stress intensity, grass pea was able to maintain leaf RWC at78% after 5 days of 20% PEG treatment, while the leaf RWC in pea decreased to 62%, suggesting that grass pea encountered a less severe water deficit than pea. The reduction in RWC due to water stress was also reported by Chowdhury *et al*. (2017); Omae *et al.* (2007) and Omae *et al*. (2005).

Table 7. Relative water content of mungbean genotypes under polyethylene glycol concentrations

Treatments	Relative water content (%) at different polyethylene glycol (PEG) concentrations				
	0%	5%	10%	15%	20%
V_1	81.68 b-e	77.52 c-f	73.37 c-e	62.28 cd	51.88 b-d
V_2	92.90 a	89.85 ab	85.70 ab	82.06 a	78.70 a
V_3	86.24 a-d	82.00 a-e	74.84 c-e	64.35 cd	53.95 bc
V_4	79.85 b-e	74.17 d-g	67.11 e-g	57.25 de	43.17 ef
V_5	88.73 ab	85.10 a-c	81.63 a-c	78.41 ab	76.09 a
V_6	86.35 a-d	83.31 a-d	75.77 cd	64.42 cd	51.02 cd
V_7	86.44 a-d	81.22 b-e	71.72 de	59.39 de	46.05 de
V_8	94.78 a	91.71 a	88.84 a	84.20 a	80.94 a
V_9	86.55 a-d	80.18 b-e	70.78 d-f	57.68 de	45.82 de
V_{10}	79.74 b-e	73.12 e-g	61.00 g-i	51.22 ef	37.57 fg
V_{11}	78.84 c-e	75.73 c-g	66.67 e-g	56.40 de	41.81 ef
V_{12}	77.49 de	74.68 d-g	63.10 f-h	52.87 ef	37.76 fg
V_{13}	74.56 e	68.91 fg	56.72 hi	47.26 fg	33.95 g
V_{14}	83.19 b-e	78.53 c-f	71.28 d-f	62.46 cd	48.65 c-e
V_{15}	87.42 a-c	84.76 a-c	78.25 b-d	70.19 bc	59.36 b
V_{16}	74.44 e	67.10 g	54.18 i	41.84 g	25.55 h
LSD (0.01)	**9.18**	**9.88**	**8.51**	**8.24**	**7.66**
CV (%)	**4.91**	**5.58**	**5.34**	**5.94**	**6.74**

V_1 = BARI Mung 3, V_2 = BARI Mung 4, V_3 = BARI Mung 5, V_4 = BARI Mung 6, V_5 = BARI Mung 7, V_6 = BARI Mung 8, V_7 = BINA Mung 5, V_8 = BINA Mung 6, V_9 = BINA Mung 8, V_{10} =$BMXK_1$-09015-6, V_{11} =BMX-08011-2, V_{12} = BMX-08011-8, V_{13} =$BMXK_1$ -09015-2, V_{14} =$BMXK_1$-09012-1, V_{15} =PM-5 and V_{16} =IPM-02-03

4.8 Water saturation deficit (WSD)

The amount of water vapor needs to be increased in the air to attain a saturation point without disturbing the environmental condition (temperature and pressure) is called water saturation deficit. It is opposite to relative water content. PEG concentration had highly significant influence on water saturation deficit among different mungbean genotypes (Table 8). The results revealed that, water saturation deficit ranged from 25.56, 32.90, 45.82, 58.16 and 74.45 in IPM-02-03, to 5.22, 8.29, 11.16, 15.80 and 19.06 in BINA Mung 6 at 0%, 5%, 10%, 15% and 20% PEG concentration, respectively were recorded. IPM-02-03, $BMXK_1$ -09015-2 and $BMXK_1$-09015-6 showed very much sensitivity to higher PEG concentration. Therefore, BINA Mung-6, BARI Mung 4 and BARI Mung 7 mungbean genotypes exerted better tolerance against drought stress condition in case of water saturation deficit. The sensitive

mungbean genotypes had less capability to uptake water under drought stress condition. So, the relative water content is lower and water saturation deficit is higher. But the comparatively tolerant genotypes able to uptake enough water necessary for running the physiological process smoothly under drought stress condition, thus there was a less water deficit occurred in tolerant genotypes than the sensitive ones.

Table 8. Water saturation deficit of mungbean genotypes under polyethylene glycol concentrations

Treatments	Water saturation deficit at different polyethylene glycol (PEG) concentrations				
	0%	5%	10%	15%	20%
V_1	18.32 de	22.48 cd	26.63 f-h	37.72 fg	48.12 gh
V_2	7.103 h	10.15 i	14.30 k	17.94 ij	21.30 j
V_3	13.76 f	18.00 fg	25.16 g-i	35.65 g	46.05 hi
V_4	20.48 bc	25.83 b	32.89 de	42.75 d-f	56.83 c-e
V_5	11.27 g	14.90 h	18.37 j	21.59 i	23.91 j
V_6	13.65 f	16.69 gh	24.23 hi	35.58 g	48.98 f-h
V_7	13.56 f	18.78 fg	28.28 fg	40.61 e-g	53.95 d-g
V_8	5.217 h	8.290 i	11.16 k	15.80 j	19.06 j
V_9	13.45 f	19.82 ef	29.22 ef	42.32 d-f	54.18 d-f
V_{10}	20.23 cd	26.88 b	39.00 b	48.78 bc	62.43 bc
V_{11}	21.16 bc	24.27 bc	33.33 cd	43.60 c-e	58.19 cd
V_{12}	22.51 b	25.32 b	36.90 bc	47.13 cd	62.24 bc
V_{13}	25.44 a	31.09 a	43.28 a	52.74 b	66.05 b
V_{14}	16.81 e	21.47 de	28.72 fg	37.54 fg	51.35 e-h
V_{15}	12.58 fg	15.24 h	21.75 ij	29.81 h	40.64 i
V_{16}	25.56 a	32.90 a	45.82 a	58.16 a	74.45 a
LSD (0.01)	**2.13**	**2.64**	**3.95**	**5.22**	**6.03**
CV (%)	**5.85**	**5.69**	**6.16**	**6.14**	**5.48**

V_1 = BARI Mung 3, V_2 = BARI Mung 4, V_3 = BARI Mung 5, V_4 = BARI Mung 6, V_5 = BARI Mung 7, V_6 = BARI Mung 8, V_7 = BINA Mung 5, V_8 = BINA Mung 6, V_9 = BINA Mung 8, V_{10} =$BMXK_1$-09015-6, V_{11} =BMX-08011-2, V_{12} = BMX-08011-8, V_{13} =$BMXK_1$ -09015-2, V_{14} =$BMXK_1$-09012-1, V_{15} =PM-5 and V_{16} =IPM-02-03

4.9 Water retention capacity (WRC)

The amount of water useful for crop hold by the crop plant is the water retention capacity. The turgid weight/dry weight (TW/DW) ratio illustrates the water retention capacity (WRC) of plants that are determined by the cell structures. Plants grown under a high moisture regime maintains a higher ratio and that might be due to the lower destruction of plant tissues by moisture deficit (Sangakkara *et al*., 1996). PEG concentrations significantly influenced water retention capacity of mungbean

genotypes (Table 9). Highest water retention capacity ranged from 24.98, 24.32, 23.38, 22.36 and 21.27 in BINA Mung 6 to 11.39, 10.37, 9.12, 6.85 and 3.08 in IPM-02-03 0%, 5%, 10%, 15% and 20% PEGS concentration, respectively were attained. IPM-02-03, $BMXK_1$ -09015-2 and $BMXK_1$-09015-6 showed very much sensitivity to higher PEG concentration. Therefore, BINA Mung-6, BARI Mung 4 and BARI Mung 7 mungbean genotypes performed better against drought stress condition in respect of water retention capacity. The tolerance cultivars have the capacity to uptake water under salt stress condition than the sensitive ones and gained the maximum turgid weight, in consequence they gained the maximum water retention capacity. The reduction in the leaf WRC in drought sensitive genotypes could be result of hemi-cellulose and cellulose accumulation in the cell wall. There is a negative relationship between WRC and drought resistance index (DRI) under water stress. A decrease in the leaf WRC indicated a decrease in cell size. A reduction in cell size is one of the most common anatomical changes observed in water stressed leaves. Sanagakkara *et al.* (1996) observed similar results in *Phaseolus vulgaris*. Chowdhury *et al*. (2017) concluded that, Shohag and BARI Soybean-6 showed the lowest reduction in WRC, and thus an indication of their tolerance to water stress.

Table 9. Water retention capacity of mungbean genotypes under polyethylene glycol concentrations

Treatments	Water retention capacity at different polyethylene glycol (PEG) concentrations				
	0%	5%	10%	15%	20%
V_1	11.78 fg	11.16 gh	10.04 g-i	7.96 h-j	5.46 h
V_2	23.81 a	23.04 a	21.90 a	20.55 b	17.87 b
V_3	11.96 fg	11.39 f-h	10.53 f-i	8.55 g-i	6.83 g
V_4	15.35 d	14.21 d	12.55 de	10.66 ef	8.40 ef
V_5	21.25 b	20.97 b	19.51 b	17.96 c	15.7 c
V_6	12.48 e-g	11.80 e-h	10.92 e-h	8.28 h-j	6.18 gh
V_7	13.62 d-f	13.24 d-f	11.86 d-f	9.45 e-h	7.23 fg
V_8	24.98 a	24.32 a	23.38 a	22.36 a	21.27 a
V_9	14.77 d	13.58 de	12.84 d	11.13 e	9.07 e
V_{10}	11.95 fg	11.06 gh	9.85 hi	7.63 ij	3.93 i
V_{11}	12.24 fg	11.77 e-h	10.53 f-i	8.26 h-j	6.46 gh
V_{12}	14.13 de	13.54 de	12.51 de	10.23 e-g	8.21 ef
V_{13}	11.42 g	10.89 gh	9.83 hi	7.27 ij	3.82 i
V_{14}	12.83 e-g	12.40 d-g	11.58 d-g	9.37 f-h	7.24 fg
V_{15}	17.81 c	17.19 c	16.13 c	14.40 d	12.34 d
V_{16}	11.39 g	10.37 h	9.12 i	6.85 j	3.08 i
LSD (0.01)	**1.88**	**2.01**	**1.68**	**1.69**	**1.29**
CV (%)	**5.55**	**6.22**	**5.63**	**6.70**	**6.43**

V_1 = BARI Mung 3, V_2 = BARI Mung 4, V_3 = BARI Mung 5, V_4 = BARI Mung 6, V_5 = BARI Mung 7, V_6 = BARI Mung 8, V_7 = BINA Mung 5, V_8 = BINA Mung 6, V_9 = BINA Mung 8, V_{10} =$BMXK_1$-09015-6, V_{11} =BMX-08011-2, V_{12} = BMX-08011-8, V_{13} =$BMXK_1$ -09015-2, V_{14} =$BMXK_1$-09012-1, V_{15} =PM-5 and V_{16} =IPM-02-03

4.10 Germination coefficient (CG)

Drought stress levels significantly influenced the coefficient of germination of mungbean genotypes (Table 10). Maximum mungbean genotypes perform better in respect of germination coefficient up to 5% PEG concentration but with the advancement of stress the sensitive ones performed poor compare to that of tolerant ones. The result of the investigation revealed that, maximum coefficient of germination ranged from 22.27, 22.10, 21.38, 20.74 and 19.58 in BINA Mung 6 to 17.84, 17.23, 15.13, 11.52 and 6.06 in IPM-02-03 mungbean genotype at 0%, 5%, 10%, 15% and 20% PEG concentration, respectively were recorded. Therefore, BINA Mung-6, BARI Mung 4 and BARI Mung 7 mungbean genotypes performed better against drought stress condition in respect of germination coefficient. The Germination coefficient is indicative of the speed of germination and quick

establishment in reduced water potentials. Kaur *et al.* (2017) revealed that, the higher the germination coefficient the quicker establishment capacity of the mungbean genotypes. The cumulative germination coefficient was quite high in SML-1141 mungbean genotype which was also characterized by a higher level of germination potentiality. Many reports indicated that germination coefficient can be utilized as screening criteria for stress tolerance crop (Ahmad *et al*., 2009). The high germination coefficient in mungbean genotype would indicate higher level of tolerance to drought (Dhopte and Livera, 1989) which emphasized the use of germination coefficient in screening drought tolerance in pulses. Mbarek *et al.* (2013) and Turner (1986) indicated that the germination coefficient is inversely proportional to the PEG8000 concentration in the media culture. Evolution of this parameter according to concentration shows the negative action of the PEG8000 on the germination coefficient. Similar results also obtained by Hanen and Ahmad (2016) who reported that, the germination coefficient of drought sensitive genotypes reduced with the increasing of drought stress induced by PEG.

Table 10. Coefficient of germination of mungbean genotypes under polyethylene glycol concentrations

Treatments	Coefficient of germination at different polyethylene glycol (PEG) concentrations				
	0%	5%	10%	15%	20%
V_1	20.55 a-c	19.50 b-f	17.69 c-g	15.53 c-f	12.67 d-g
V_2	22.11 a	21.99 a	21.00 ab	19.96 a	18.66 ab
V_3	21.36 a	20.21 a-d	18.03 c-f	15.50 c-f	12.33 e-g
V_4	20.15 a-d	19.41 b-f	17.57 d-g	15.15 d-f	12.29 fg
V_5	21.97 a	21.68 ab	19.87 a-c	18.89 ab	17.44 b
V_6	21.56 a	20.96 a-c	18.04 c-f	16.26 c-e	14.00 c-f
V_7	21.72 a	21.30 a-c	19.40 a-d	17.44 bc	14.26 cd
V_8	22.27 a	22.10 a	21.38 a	20.74 a	19.58 a
V_9	21.83 a	21.21 a-c	19.07 b-d	17.30 b-d	14.23 cd
V_{10}	18.54 b-d	17.74 ef	15.86 f-h	13.45 f-h	9.45 h
V_{11}	21.13 ab	20.60 a-d	18.46 c-e	16.09 c-e	14.08 c-e
V_{12}	20.62 a-c	18.35 d-f	17.36 d-g	15.17 d-f	12.77 d-g
V_{13}	18.35 cd	17.43 f	15.60 gh	12.36 gh	7.50 i
V_{14}	19.85 a-d	19.09 c-f	16.73 e-h	14.51 e-g	12.20 g
V_{15}	21.53 a	20.00 a-e	18.40 c-e	17.08 b-d	15.31 c
V_{16}	17.84 d	17.23 f	15.13 h	11.52 h	6.06 i
LSD (0.01)	**2.69**	**2.44**	**2.20**	**2.18**	**1.76**
CV (%)	**5.80**	**5.47**	**5.44**	**6.06**	**5.91**

V_1 = BARI Mung 3, V_2 = BARI Mung 4, V_3 = BARI Mung 5, V_4 = BARI Mung 6, V_5 = BARI Mung 7, V_6 = BARI Mung 8, V_7 = BINA Mung 5, V_8 = BINA Mung 6, V_9 = BINA Mung 8, V_{10} =$BMXK_1$-09015-6, V_{11} =BMX-08011-2, V_{12} = BMX-08011-8, V_{13} =$BMXK_1$ -09015-2, V_{14} =$BMXK_1$-09012-1, V_{15} =PM-5 and V_{16} =IPM-02-03

4.11 Vigour Index (VI)

Table 11 showed that, the vigor index was significantly differed by different mungbean genotypes. The magnitude of reduction of vigor index was slower up to 10% PEG concentration but with the increasing of PEG concentration from 10% to 20% there were drastic reduction occurred for maximum mungbean genotypes. But the tolerant genotypes maintained a standard reduction with the advancement of drought stress. The result of the study revealed that, maximum vigor index ranged from 233.90, 230.40, 220.50, 205.20 and 186.10 in BINA Mung 6 to 114.80, 99.37, 76.44, 41.02 and 13.45 in IPM-02-03 mungbean genotype at 0%, 5%, 10%, 15% and 20% PEG concentration, respectively were recorded. Therefore, BINA Mung-6, BARI Mung 4 and BARI Mung 7 mungbean genotypes performed better against drought stress condition in respect of vigor index. ROS are generated by the forced

transfer of excess electrons produced during either photochemistry in chloroplasts (Edreva, 2005) or respiration in the mitochondria (Navrot *et al.*, 2007). Jiang *et al.* (2013) reported that, O^{-2} and H_2O_2 significantly increased after increasing the PEG concentration. H_2O_2 can damage membranes and result in peroxidation of membrane lipids. MDA is one of the end products of lipid peroxidation, and its level reflects the degree of membrane lipid peroxidation. The content of MDA increased about 1.3-fold in grass pea, while it was up 2.1-fold in pea compared with their respective untreated controls, suggesting that more serious oxidative stress occurred in pea compared with grass pea. The increasing in membrane lipid per oxidation reduced the seedling vigority consequently the comparatively sensitive ones attained lower vigor index in our study. Rana *et al*. (2017) reported that, the results on vigor index showed that the speed of germination was reduced with the increment of water deficit stress but the degree of reduction was not similar for all wheat genotypes at moderate and higher water deficit stress compared to control. The mungbean genotypes varied significantly for vigour index in all the concentrations. Swathi *et al.* (2017) found that, in reduced water potential of -0.3 MPa the vigour index ranged between 476.40 (EC 396117) and 2088.70 (ML 267) with an average mean of 1214.32. Similarly, at -0.6MPa vigour index ranged between 324.00 (WGG 2) and 1393.60 (WGG 37) with an average mean of 804.67. At -0.9 MPa the vigour index ranged between 136.65 (TM 96-2) and 1425.45 (ML 267) with an average mean of 564.87. Based on the Non-parametric study, the genotype ML 267 showed better drought tolerance capability. Kaur *et al.* (2017) reported that, the mean vigour index of all genotypes 602.20 in the control significantly decreased to 192.76, 88.76 and 19.28 in -0.4MPa, -0.6MPa and -0.8MPa, respectively. The cumulative vigour index was maximum (294.5) in SML-1411mungbean genotype. These results were also in accordance with the findings of Dutta and Bera (2008) and Kuar *et al.* (2011) in mungbean.

Table 11. Vigor index of mungbean genotypes under polyethylene glycol concentrations

Treatments	Vigor index at different polyethylene glycol (PEG) concentrations				
	0%	5%	10%	15%	20%
V_1	141.2 hi	125.0 g-i	105.1 gh	80.45 g	53.37 gh
V_2	228.9 a	221.4 a	213.6 ab	190.8 ab	168.3 b
V_3	192.0 cd	178.6 bc	156.8 cd	122.1 cd	77.41 e
V_4	156.3 f-h	140.3 e-g	121.1 fg	84.99 fg	51.43 hi
V_5	221.3 ab	213.1 a	199.9 b	177.3 b	152.2 c
V_6	151.5 gh	136.8 fg	119.9 fg	88.27 fg	54.90 gh
V_7	174.9 d-f	158.7 c-e	140.8 de	108.1 de	63.14 fg
V_8	233.9 a	230.4 a	220.5 a	205.2 a	186.1 a
V_9	149.3 gh	132.4 gh	108.8 gh	76.57 g	41.58 i
V_{10}	133.7 h-j	116.7 h-j	94.76 hi	58.90 h	29.49 j
V_{11}	188.1 c-e	169.6 b-d	148.6 cde	111.8 de	72.78 ef
V_{12}	180.2 c-e	166.4 b-d	140.9 de	105.0 e	68.24 ef
V_{13}	120.8 ij	106.6 ij	83.22 i	49.15 hi	22.27 jk
V_{14}	167.3 e-g	153.2 d-f	132.3 ef	99.67 ef	62.71 fgh
V_{15}	199.6 bc	183.5 b	161.0 c	135.3 c	107.3 d
V_{16}	114.8 j	99.37 j	76.44 i	41.02 i	13.45 k
LSD (0.01)	**22.86**	**20.06**	**19.42**	**14.86**	**11.34**
CV (%)	**5.94**	**5.67**	**6.25**	**6.13**	**6.63**

V_1 = BARI Mung 3, V_2 = BARI Mung 4, V_3 = BARI Mung 5, V_4 = BARI Mung 6, V_5 = BARI Mung 7, V_6 = BARI Mung 8, V_7 = BINA Mung 5, V_8 = BINA Mung 6, V_9 = BINA Mung 8, V_{10} =$BMXK_1$-09015-6, V_{11} =BMX-08011-2, V_{12} = BMX-08011-8, V_{13} =$BMXK_1$ -09015-2, V_{14} =$BMXK_1$-09012-1, V_{15} =PM-5 and V_{16} =IPM-02-03

CHAPTER V

SUMMARY AND CONCLUSION

The experiment was conducted at Laboratory of Department of Agronomy, Sher-e-Bangla Agricultural University (SAU), Sher-e-Bangla Nagar, Dhaka-1207during the period from February to March, 2018 to screening of drought tolerance capability of mungbean genotypes under drought stress condition. The experiment was laid out in a Completely Randomized Design (CRD) with five replications.

Sixteen mungbean genotypes *viz*. - BARI Mung 3, BARI Mung 4, BARI Mung 5, BARI Mung 6, BARI Mung 7, BARI Mung 8, BINA Mung 5, BINA Mung 6, BINA Mung 8, $BMXK_1$-09015-6, BMX-08011-2, BMX-08011-8, $BMXK_1$ -09015-2, $BMXK_1$-09012-1, PM-5 and IPM-02-03 were used as test crop. Chemicals such as PEG and distilled water were utilized for experiment. Five levels of PEG solutions (0%, 5%, 10%, 15% and 20%) were used for inducing drought stress upon the tested mungbean genotypes.

Thirty seeds from each of the treatments were selected randomly and placed in 90 mm diameter Petri dishes on whatman No.1filter paper and filter paper was moistened with distilled water and PEG solutions as per treatment.

Germination was measured to have occurred when radicles were 2 mm long. Germination progress was examined and data were collected at every 24 h intervals and continued up to 8 days. The abnormal or dead seedlings were excluded during counting. The data regarding germination, seedling growth and water relation behavior of mungbean like germination percentage, shoot length, root length, shoot dry weight, root dry weight, root shoot ratio, relative water content, water saturation deficit, water retention capacity, coefficient of germination and vigor index. The collected data were analyzed statistically following CRD design by MSTAT-C computer package program and the treatments were compared by Least Significance Differences (LSD) test at 1% level of probability.

Sixteen mungbean genotypes *viz.* V_1 = BARI Mung 3, V_2 = BARI Mung 4, V_3 = BARI Mung 5, V_4 = BARI Mung 6, V_5 = BARI Mung 7, V_6 = BARI Mung 8, V_7 = BINA Mung 5, V_8 = BINA Mung 6, V_9 = BINA Mung 8, V_{10} =$BMXK_1$-09015-6, V_{11} =BMX-08011-2, V_{12} = BMX-08011-8, V_{13} =$BMXK_1$-09015-2, V_{14} =$BMXK_1$-09012-1, V_{15} =PM-5 and V_{16} =IPM-02-03 were tested under drought stress condition with a view to screening of drought tolerant capability of mungbean genotypes and for that context germination, seedling growth and water relation behavior of mungbean genotypes were evaluated under five levels (0%, 5%, 10%, 15% and 20%) of PEG solutions.

The findings of the investigation revealed that, BINA Mung-6 consistently scored highest values for all of parameters except water saturation deficit which was statistically similar with BARI Mung 4 and BINA Mung 5 for most of the cases where as IPM-02-03 advanced lines of mungbean consistently performed poor along with BMXK1 -09015-2 and BMXK1-09015-6 advance lines of mungbean. The maximum germination percentage (98.12%), shoot length (139.40 mm), root length (99.07 mm), shoot dry weight (22.32 mg), root dry weight (12.26 mg), root shoot ratio (0.55), relative water content (94.78), water retention capacity (24.98), coefficient of germination (22.27) and vigor index (233.90) were recorded from BINA Mung 6 at 0% PEG concentration. The minimum germination percentage (28.22%), shoot length (31.17 mm), root length (16.50 mm), shoot dry weight (2.21 mg), root dry weight (0.49 mg), relative water content (25.55), water retention capacity (3.08), coefficient of germination (6.06) and vigor index (13.45) were recorded from IPM-02-03 advanced lines of mungbean at 0% PEG concentration. The minimum root shoot ratio (0.20) at 20% PEG concentration from BARI Mung-5. The maximum water saturation deficit (74.45) was recorded from IPM-02-03 at 20% PEG concentration and the minimum one (5.22) from BINA Mung-6 at 0% PEG concentration.

Considering the above findings achieved from the present piece of work it may be concluded that among 16 mungbean genotypes BINA Mung-6, BARI Mung-4 and BINA Mung-5 performed best under drought stress condition which were attributed to higher germination percentage, root shoot ratio, relative water content, water retention capacity, coefficient of germination and vigor index and rest of the mungbean genotypes found to be moderately to strongly sensitive to drought stress.

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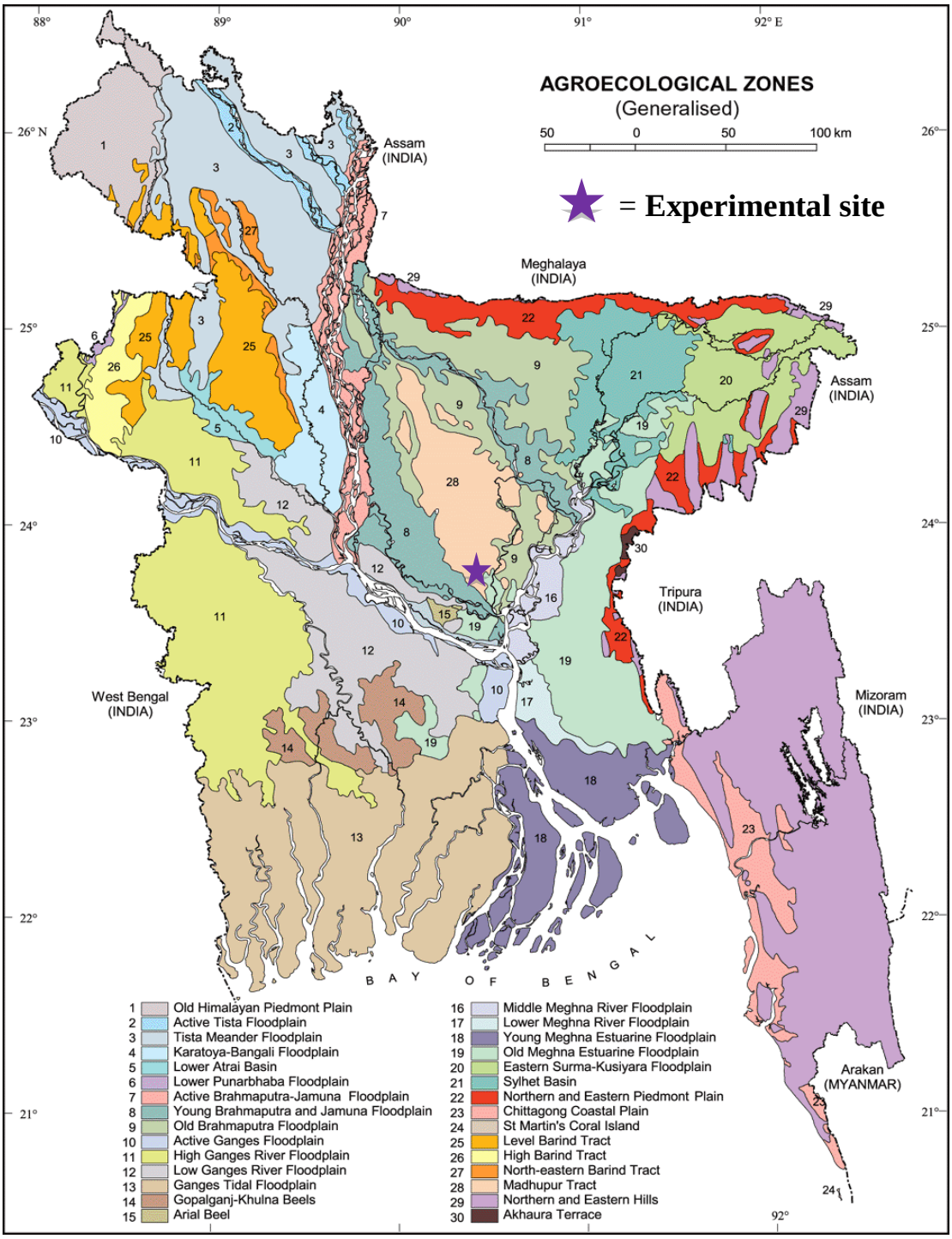
APPENDICES

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



Appendix II. Monthly records of Temperature and Relative humidity of the experiment lab during the period from February to March, 2018

Year	Month	Air Temperature (^{0}c)			Relative humidity (%)
		Maximum	Minimum	Mean	
2018	February	28.0	22.1	25.05	60.5
	March	30.4	25.6	28.00	68.6

Source: Bangladesh Meteorological Department (Climate division), Agargaon, Dhaka-1212.Source: Bangladesh Meteorological Department (Climate division), Agargaon, Dhaka-1212.

Appendix III. Analysis of variance of the data on germination percentage as influenced by different mungbean genotypes

Source of variation	df	Mean square of germination percentage at different PEG concentration				
		0%	5%	10%	15%	20%
Treatment	15	128.69**	177.47**	288.39**	521.00**	893.57**
Error	32	15.89	17.52	22.31	13.70	12.16

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

Appendix IV. Analysis of variance of the data on shoot length as influenced by different mungbean genotypes

Source of variation	df	Mean square of shoot length at different PEG concentration				
		0%	5%	10%	15%	20%
Treatment	15	833.87**	989.57**	1141.24**	1553.07**	2270.98**
Error	32	42.07	37.98	34.44	29.66	18.51

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

Appendix V. Analysis of variance of the data on root length as influenced by different mungbean genotypes

Source of variation	df	Mean square of root length at different PEG concentration				
		0%	5%	10%	15%	20%
Treatment	15	546.75**	631.15**	779.84**	1023.88**	1266.94**
Error	32	19.61	19.15	15.95	12.70	8.79

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

Appendix VI. Analysis of variance of the data on shoot dry weight as influenced by different mungbean genotypes

Source of variation	df	Mean square of shoot dry weight at different PEG concentration				
		0%	5%	10%	15%	20%
Treatment	15	49.20**	55.27**	59.33**	74.48**	95.09**
Error	32	0.79	0.79	0.59	0.46	0.22

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

Appendix VII. Analysis of variance of the data on root dry weight as influenced by different mungbean genotypes

Source of variation	df	Mean square of root dry weight at different PEG concentration				
		0%	5%	10%	15%	20%
Treatment	15	25.03**	22.65**	19.80**	21.41**	19.99**
Error	32	0.13	0.10	0.07	0.05	0.03

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

Appendix VIII. Analysis of variance of the data on root shoot ratio as influenced by different mungbean genotypes

Source of variation	df	Mean square of root shoot ratio at different PEG concentration				
		0%	5%	10%	15%	20%
Treatment	15	0.03**	0.02**	0.02**	0.01**	0.02**
Error	32	0.001	0.001	0.001	0.0001	0.001

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

Appendix IX. Analysis of variance of the data on relative water content as influenced by different mungbean genotypes

Source of variation	df	Mean square of relative water content at different PEG concentration				
		0%	5%	10%	15%	20%
Treatment	15	108.38**	143.13**	285.04**	429.09**	773.77**
Error	32	16.87	19.53	14.48	13.58	11.72

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

Appendix X. Analysis of variance of the data on water saturation deficit as influenced by different mungbean genotypes

Source of variation	df	Mean square of water saturation deficit at different PEG concentration				
		0%	5%	10%	15%	20%
Treatment	15	108.86**	143.13**	285.04**	429.09**	773.77**
Error	32	0.91	1.39	3.12	5.44	7.27

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

Appendix XI. Analysis of variance of the data on water retention capacity as influenced by different mungbean genotypes

Source of variation	df	Mean square of water retention capacity at different PEG concentration				
		0%	5%	10%	15%	20%
Treatment	15	60.05**	60.76**	60.32**	71.41**	82.34**
Error	32	0.70	0.81	0.56	0.57	0.33

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

Appendix XII. Analysis of variance of the data on coefficient of germination as influenced by different mungbean genotypes

Source of variation	df	Mean square of coefficient of germination at different PEG concentration				
		0%	5%	10%	15%	20%
Treatment	15	5.96**	7.83**	9.61**	19.05**	39.41**
Error	32	1.44	1.19	0.97	0.95	0.62

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

Appendix XIII. Analysis of variance of the data on vigor index as influenced by different mungbean genotypes

Source of variation	df	Mean square of vigor index at different PEG concentration				
		0%	5%	10%	15%	20%
Treatment	15	4100.98**	4779.95**	5717.27**	7033.02**	7905.84**
Error	32	104.49	80.46	75.40	44.19	25.71

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