INDUCTION OF SALT TOLERANCE CAPABILITY IN MUNGBEAN THROUGH POLYETHYLENE GLYCOL (PEG) AND HYDRO PRIMING

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CERTIFICATE

This is to certify that the thesis entitled, "**INDUCTION OF SALT TOLERANCE CAPABILITY IN MUNGBEAN THROUGH POLYETHYLENE GLYCOL (PEG) AND HYDRO PRIMING**" submitted to the Faculty of Agriculture, Sher-e-Bangla Agricultural University, Dhaka, in partial fulfillment of the requirements for the degree of **MASTER OF SCIENCE IN AGRONOMY**, embodies the result of a piece of *bona fide* research work carried out by **NURJAHAN SHITHI**, Registration No. **12-04919** under my supervision and guidance. No part of the thesis has been submitted for any other degree or diploma.

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

Dated:
Place: Dhaka, Bangladesh

……..…….....……
Prof. Dr. Md. Abdullahil Baque
Department of Agronomy
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Dedicated to
My
Beloved Parents

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LIST OF ACCRONYMS AND ABBREVIATIONS

Agric.	=	Agriculture
Agron.	=	Agronomy
BARI	=	Bangladesh Agricultural Research Institute
BBS	=	Bangladesh Bureau of Statistics
Biotechnol.	=	Biotechnology
^{o}C	=	Degree centigrade
CRD	=	Completely Randomized Design
CV	=	Coefficient of variation
ed.	=	Edition
EG	=	Energy of emergence
Environ.	=	Environmental
et al.	=	And others
GI	=	Germination Index
GP	=	Germination Percentage
hr	=	Hour
i.e.	=	that is
ISTA	=	International Seed Testing Association
LSD	=	Least significant difference
mg	=	Milligram
MGT	=	Mean Germination Time
Pathol.	=	Pathology
PEG	=	Polyethylene Glycol
Physiol.	=	Physiology
Res.	=	Research
ROS	=	Reactive Oxygen Species
RWC	=	Relative water content
Sci.	=	Science
VI	=	Vigour Index
viz.	=	Namely
WRC	=	Water retention capacity
WSD	=	Water saturation deficit

ABSTRACT

A series of experiment was conducted under the laboratory condition of the Department of Agronomy, Sher-e-Bangla Agricultural University, Dhaka from 25 May to 15 August, 2018 to evaluate the effect of pre-sowing seed treatment with Polyethylene Glycol (PEG) on germination behavior of Mungbean (BARI Mung 5 and BARI Mung 6) in relation to salt tolerance and to optimize priming time. In the 1st experiment, seeds of BARI Mung 5 and BARI Mung 6 were pre-soaked in water, 5, 10, 15 and 20% PEG solution and untreated seeds were served as control. Results revealed that seed priming enhanced germination percentage (GP), vigor index (VI) and germination coefficient (CG). The highest GP (94.59%), VI (234.2) and CG (22.87) were obtained from seeds of BARI Mung 6 , pre-treated with 10% PEG solution compared to BARI Mung 5 (85.50%, 182.7 and 20.66 of GP, VI and CG, respectively) and then decreased gradually with increasing PEG concentration. In the 2nd experiment, BARI Mung 6 was primed for 3,6,9,12,15 and 18 hours with 10% PEG solution and distilled water. The highest GP (95.31%), VI (237.8) and CG (22.35) were obtained from seeds pre-soaked for 9 hr with 10% PEG compared to hydro-priming (87.33%, 194.4 and 20.73 of GP, VI and CG, respectively) and then decreased gradually with increasing priming time. In the 3rd experiment osmo and hydro primed seeds were allowed to grow under 5 levels of salt concentration (0 dSm^{-1}, 5 dSm^{-1}, 10 dSm^{-1} , 15 dSm^{-1} and 20 dSm^{-1}). Seeds primed with 10% PEG for 9 hr enhanced germination behavior and seedling growth over distilled water primed seeds. The salt tolerant capability of hydro primed seeds decreased drastically as salt stress increased but osmo primed seeds showed considerable tolerance capability upto 10 dSm^{-1} NaCl concentration then significantly decreased with increasing salinity level. These results suggest that Mungbean seed primed with 10% PEG for 9 hours is considered as best priming treatment to induce salt tolerance capability.

CHAPTER I

INTRODUCTION

Pulse crop belongs to grain legume. Pulses constitute the main source of protein for the people, particularly the poor sections of Bangladesh. These are also the best source of protein for domestic animals. Besides, this crop has the capability to enrich soils through nitrogen fixation (Sharma and Behera, 2009). Pulse protein is rich in lysine that is deficient in rice. According to FAO (2013) recommendation, a minimum intake of pulse by a human should be 80 g/day, whereas it is 7.92 g in Bangladesh (BBS,2012). In Bangladesh, total production of pulses is only 0.65 million ton against 2.7 million tons requirement. This means the shortage is almost 80% of the total requirement (Rahman and Ali, 2007). This is mostly due to low yield (MoA, 2013). At present, the area under pulse crop is 0.406 million hectare with a production of 0.322 million tons (BBS, 2013), where mungbean is cultivated in the area of 0.108 million ha with production of 0.03 million tons (BBS, 2014).

Mungbean (*Vigna radiata*) is an important source of protein in South and South East Asia. This is cultivated both as Rabi as well as Kharif season crop (Naseem *et al.*, 1997). Its grain is characterized by good digestibility, flavor, high protein content and absence of any flatulence effects (Ahmed *et al*., 2008). It holds the 3rd in protein content and 4th in both acreage and production in Bangladesh (MoA, 2014). Mungbean grain contains 51% carbohydrate, 26% protein, 10% moisture, 4% mineral and 3% vitamins (Khan *et al*., 2005). On the nutritional point of view, mungbean is one of the best among pulses (Khan *et al*., 2005).

Soil salinity is a major abiotic stress in plant agriculture world wide. This has led to research into salt tolerance with the aim of improving crop plants (Zhu, 2001). High concentrations of salts cause ion imbalance and hyper osmotic stress in plants. As a consequence of these primary effects, secondary stresses often occur (Zhu, 2001). On the other hand, salt and water stresses are

responsible for both inhibition or delayed seed germination and seedling growth (Farooq *et al.*, 2006). Under these stress conditions there is a decrease in water uptake both during imbibition and seedling establishment, and in the case of salt stress, this can be followed by excessive uptake of ions (Uhvits,1946; West and Francois (1982). It has also been shown that the inhibition of radicle emergence is mainly because of a decrease in water potential gradient between the external environment and the seeds (Ene´asFilho *et al.*, 1995). Salinity inhibition of embryo-axis growth during seedling establishment is the result of both delayed reserve mobilization (Prisco and Vieira, 1976) and membrane disturbance caused by salinity and evidenced by increased leakage of materials from the embryo-axis (Prisco, 1987).The accumulation of soluble salts in soils leads to an increase in osmotic pressure of the soil solution, which may limit the absorption of water by the seeds or by the plant roots. Salt damage to plants is attributed to the reduction in water availability, toxicity or specific ions, and nutritional imbalance caused by such ions. Poor crop establishment is a constraint for mungbean production (Naseem *et al.*, 1997; Rahmianna *et al.*, 2000) and high yields can be associated with early vigour (Kumar *et al*., 2002).

There are different approaches to mitigate the salt hazards, which include the development of stress tolerant plants by selection of stress resistant varieties, *in vitro* selection, use of plant growth hormones (ABA, GA, cytokinin, SA), antioxidants (ascorbic acid) and osmo protectants as foliar application and seed treatment (Farooq *et al.*, 2009). Since tolerance to salt in plants is a complex trait, conventional breeding techniques have had limited success in improving this trait in crops (Flower *et al*, 1997). But these are not economically viable technology to facilitate crop production under stress conditions. Seed priming has proved to be an effective method in imparting stress tolerance to plants. Seed priming is the induction of a particular physiological state in plants by the treatment of natural and synthetic compounds to the seeds before sowing. The physiological state in which plants are able to faster or better activate defense responses is called the primed state of the plant (Beckers and Conrath , 2007).

It has been found a realizable technology to enhance rapid and uniform emergence, high vigor and better yields for vegetable and field crops (Janmohammadi *et al.*, 2009). In fact, this technique is a treatment that applied before germination in a specific environment that seeds are partially hydrated to a point where germination processes begin but radical emergence does not occur (Kaur *et al.*, 2002). Seed priming can be accomplished through different methods such as hydro priming (soaking in DW), osmo priming (soaking in osmotic solutions such as mannitol, PEG, potassium salts, e.g., KCl, K_2SO_4) and plant growth inducers (CCC, Ethephon, IAA) (Chiu *et al.*, 2002; Harris *et al.*, 1999).Therefore, seed priming is a technology that enhances rapid emergence (7-10 d) and early establishment of mungbaen. Moreover, it is also important to study more about the performance of on the germination, vigour and other attributes of mungbean. Therefore the present study on seed priming of mungbean was formulated with the following objectives,

i) To evaluate the effect of different concentrations of Polyethyline glycol on the germination behavior of mungbean.

ii) To optimize the priming time of PEG on germination behavior of mungbean.

iii) To evaluate the effect of pre-sowing seed treatment with Polyethyline glycol on germination behavior of mungbean occurring saline stress conditions.

CHAPTER II

REVIEW OF LITERATURE

Jisha *et al.* (2013) reported that plants are exposed to number of potentially adverse environmental conditions such as water deficit, high salinity, extreme temperature, submergence, etc. These abiotic stresses adversely affect the plant growth and productivity. Now-a-days various strategies are employed to generate plants that can withstand these stresses. In recent years, seed priming has been developed as an indispensable method to produce tolerant plants against various stresses.

2.1 Seed priming

Jisha *et al*. (2013) reported that Seed priming is the induction of a particular physiological state in plants by the treatment of natural and synthetic compounds to the seeds before germination. In plant defense, priming is defined as a physiological process by which a plant prepares to respond to imminent abiotic stress quickly by cellular defense. Priming for enhanced resistance to abiotic stress in different metabolic processes. The seedlings emerging from primed seeds showed early and uniform germination. Moreover, the overall growth of plants is enhanced due to the seed-priming treatments (Farooq *et al*., 2006). The main objective of this review is to provide an overview of various crops in which seed priming is practiced and about various seed-priming methods effects.

Seed priming techniques such as hydro priming, hardening, osmo conditioning, osmo hardening and hormonal priming have been used to accelerate emergence of roots and shoots, more vigorous plants, and better drought and salt tolerance in many field crops like wheat (Iqbal and Ashraf, 2007), chickpea (Kaur *et al*., 2002), sunflower (Kaya *et al*., 2006) and cotton (Casenave and Toselli, 2007). Incase of mungbean, 4 hours and 8 hours primed seeds showed significant difference in germination percentage and seed moisture percentage over non-primed seeds (Saha *et al*., 2006).

2.2 Priming effect on germination parameters

2.2.1 Germination percentage

Laghari *et al*. (2016) reported that seed priming is a controlled hydration method in which seeds are soaked in water or low osmotic potential solution for a point where germination related metabolic exercises start in the seeds, however radical development does not happen. During seed priming, it was found effective for legumes that is, yields of legume harvest were increased impressively by priming seeds before sowing. The maximum mean seed germination (86.78%) was recorded at Hydro-priming period 4 hours, whereas the lower seed germination (68.88%) no priming in mungbean.

According to Sun *et al., (*2010), PEG priming with moderate concentration resulted in higher tolerance to salt stress than hydro-priming, while higher concentrations of PEG had negative effects on seed germination.

According to Posmyk and Janas (2007), at low temperature hydro priming and hydro priming along with proline can be practiced as a harmless priming process for betterment of seed germination and growth of *Vigna radiata*. Stress injuries also repaired fast through hydro priming. More even germination and emergence were found in primed seeds on canola (*Brassica compestris*) (Zheng *et al.*, 1994), wheat (*Triticum aestivum*), (Nayyar *et al.*, 1995) and rice *(Oryza sativa)* (Lee and Kim, 2000; Basra *et al.*, 2003) who defined hydro priming for 24 hr betterment hardening, germination rate and percentage in seeds.

Kumar *et al.* (2017) reviewed that, osmo-priming treated seed showed significantly higher germination percentage in PEG at 20% followed by mannitol 4% in chick pea.

It was informed by Kaya *et al*. (2006) seed priming had an important result on increasing of germination percent; germination speed and seedling dry weight of sunflower. Priming also decreased abnormal seedling in drought stress. Therefore, the positive effects of priming may be more obvious under

unfavorable rather than favorable conditions (Parera and Cantliffe, 1994). In mungbean, 4 hr and 8 hr primed seeds presented substantial variation in germination and seed moisture percentage over un-primed seeds (Saha *et al*., 2006). Increasing germination rate by 10-15% was reported through hardening (150 gm seeds soaked in 500 mL water for 18 and 24 hr) of older rice seeds, as the process of increase total sugar content and α-amylase activity (Lee and Kim, 2000).

Seed priming hasten germination percentage, lessened emergence time and enhanced yields are reviewed in many crops (Afzal *et al.,* 2006). Judicious doses of PEG (Polyethylene Glycol) showed better tolerance at drought stress condition than hydro-priming, while more doses of PEG had negative effects on germination (Sun *et al*., 2010).

Natural priming has been shown to increase germination synchrony, rate and final percentage in many species (Gonz_alez-Zertuche *et al.*, 2001; Santini and Martorell, 2013). Seed priming treatments not only improved germination rate and time, but it also enhanced seedling vigour, as indicated by longer roots and shoots and higher seedling dry weight (Mahajan *et al*., 2011).

2.2.2 Mean germination time (days)

According to Basra *et al*., (2005) priming to reduce mean germination time over the un-primed one. The MGT is dependent on the duration of imbibitions and/or internal metabolic activities after imbibitions . Priming activates internal metabolism required for hastening the germination process.

Tavili *et al. (*2011) reported that speed of germination of Bromus increased with seed priming treatments than nonpriming (control). Similarly, Elkoca *et al*., (2007) determined that hydro priming treatment in chickpea induced faster and more synchronous germination compared to the nonprimed seeds. Furthermore, Korkmaz (2005) for sweet pepper and Korkmaz and Pill (2003) for lentil reported that priming treatments generally improved the germination synchrony. Besides, in terms of priming duration, priming treatment for 24 hr

generally reduced the germination synchrony compared with the treatment for 12 hr. This result indicated that longer priming duration may overcome effect of decreased water potential in osmo-priming treatments of lentil seeds.

At the time of different phases of seedling establishment priming plays very significant role to reduce the time between planting and emergence and safe the seeds from environmental stress. Uniform stands and improved yield could be maintained through earlier and synchronized emergence (Farooq *et al*., 2006; Afzal *et al*., 2006). Like germination percentage, prime seeds need lower mean emergence time (MET) than nonprimed seeds. Priming creates stimulatory effects on the early phases of emergence by occurrence of cell division in germinating seeds (Hassanpouraghdam *et al*., 2009; Sivritepe *et al*., 2003).

Improved seed priming methods were recognized to lessen emergence time, achieve even emergence and ensure good crop stand in several horticultural and field crops (Ashraf and Foolad, 2005). According to Finch-Savage *et al*. (2004) without changing germination percentage priming minimize the optimum and ceiling temperature for germination and also facilitated in progressing the germination time.

Rashid *et al*. (2006) reported that hydro priming was very efficient in speed germination, good crop establishment and increased yields for many crops in different environmental condition.

Yucel (2012) found that Priming treatment influenced the MGT compared with nonprime and control seeds at all of the germination temperatures. Ghassemi-Golezani *et al*. (2008a) reported that lentil seeds priming with PEG, KNO_3 and water showed the highest germination percentage. Like germination percentage, it has been reported that primed seeds had lower mean germination time. Seeds primed for 24 hr increased the germination percentage as well as decreased mean germination time compared with seeds primed for 12 hr. This result indicated that longer priming time may overcome adverse effects of

decreased water potential in osmo-priming treatments than control seeds (Sadeghi *et al*., 2011, Sağlam *et al*., 2010, and Dezfuli *et al*., 2008).

According to Basra *et al*. (1989) priming with PEG or potassium salt (K_2HPO_4 or KNO_3) resulted in faster emergence in corn seed. When salinity and drought stress cause the reduction of germination speed, hydro priming perform as a decent, simple and economical seed stimulating treatment for inbreed lines of maize (Janmohammadi *et al*., 2009).

Gray *et al.* (1990) reported that (-0.5 MPa) reduced the mean germination time of seeds of lettuce, onion and carrot.According to Goobkin (1989) and Ozbingol *et al*. (1999), priming of tomato seeds with PEG, germinate sooner than non-primed seeds and its might be due to quicker water uptake. The possible cause for early emergence of the primed seed could be for the completion of pre-germination metabolic activities making the seed ready for radicle protrusion and the primed seed germinated soon after planting compared with nonprimed dry seed (Arif, 2005).

According to Harris *et al*. (1999) early emergence and maturity in seed priming treatment could be due to advancement in metabolic state. Musa *et al.* (1999) also concluded that priming improve plant stand and provide benefits in term of maturity. Seed priming resulted in earlier emergence of seedlings by 1-3 days and significantly increased plant stand and initial growth vigour.

Singh *et al*., (2014) found that a steady decrease in number of days taken for seed emergence from control or non-treated seeds to 16 hr of hydro priming. i.e., average number of days taken by control or non-treated seeds for 50% seed emergence was 5.00 which was highest, and average number of days taken by 12 hr of hydro priming was 2.78 days which was lowest. Hydro priming of seeds could have achieved earlier and more uniform germination than the unprimed seeds. These positive effects are probably due to stimulatory effects of priming on early stage of germination process by mediation of cell division in germinating seeds (Ghassemi-Golezani *et al*., 2010c) .

2.2.3 Seed germination index

Hydro priming could have advances the germination rate, speed (germination index) and uniformity even under less optimum field condition (Kant *et al*., 2006). Hosseein *et al*. (2011) reported that seed priming resulted in anti-oxidant increment as glutathione and ascorbate in seed. These enzymes led to higher germination speed via reduction of lipid peroxidation activity.

A critical analysis done by Singh *et al*. (2017) on different hydro priming and osmo-priming treatments on pea have significant effect on germination index, 20% Polyethylene glycol (PEG) for 24 hr (9.11) shows significant effect on rest of the treatments except 20% Polyethylene glycol (PEG) for 12 hr (8.92).

Elangbam *et al*. (2017) experimented that, the speed of germination in chickpea as influenced by various seed treatment: Lemon juice, Panchgavya, GA_3 and IAA. The maximum speed of germination with GA_3 might be due to its influence in early germination and increased percent germination. The results are in conformity with findings of Rajamanickam and Anbu (2001).

Rashid *et al*. (2006) stated that priming boosted speed of germination, better crop stand and amplified yields in varied situations for a lot of crops (Khan *et al*., 2008). Arif (2005) who described that seed priming hasten germination index as it attributed to repair processes, a buildup of germination metabolites or osmotic regulations during priming.

Kaur *et al.* (2003) conducted study to determine the effect of seed priming with mannitol (4%), water and potassium nitrate on chickpea. The response of chickpea seedlings to salt stress was also studied. In general priming with water and mannitol resulted in early germination under salt stress. Priming with 4% mannitol was also as effective as mannitol and water in the enhancement of root and shoot growth. Osmo-priming methods, Mannitol primed seed gave higher germination index than that of NaCl primed.

2.2.4 Coefficient of velocity

Coefficient of velocity (CV) is a measure of vigour (Scott *et al*., 1984). Generally, CV increases as more seeds germinate and with shorter germination time. The CV gives an indication of the speed and uniformity of seedling growth (*i.e.,* a higher CV means higher vigour).

2.2.5 Energy of emergence (%)

Seed priming boosts rapidity and uniformity of germination (Khalil *et al*., 2010; Khan *et al*., 2008; Heydecker *et al*., 1975) through inducing several chemical alterations in the seed. That alterations are obligatory to begin the germination, such as breaking of dormancy, hydrolysis or mobilization of inhibitors, imbibition and enzyme activation. Some or all of these ways that lead the germination are faster by priming and continue following the re-desiccation of the seeds (Asgedom and Becker, 2001).

Primed seed can quickly uptake and revive the seed metabolism, subsequently advanced germination rate and lessening the internal physiological heterogeneity in germination (Rowse, 1995). The consequential improved crop stand can apparently enhance the drought tolerance, decrease pest damage and hasten crop yield in cereals and legumes (Harris *et al*., 1999; Musa *et al*., 1999; Harris *et al*., 2000; Khan *et al*., 2005).

Many metabolic processes are related in the early stages of germination and those are stimulated by priming. It is well-known that seedlings from primed seeds germinate faster, grow more rapidly and perform better in negative conditions (Cramer, 2002). Roy and Srivastava (1999) found that soaking wheat kernels in water improved their germination rate under saline conditions. It also had pronounced effect on field emergence its rate and early seedling growth of maize crop and it improved the field stand and plant growth both at vegetative and maturity of maize (Nagar *et al*., 1998). Similar to other priming techniques, hydro-priming generally enhance seed germination and seedling emergence under saline and non-saline conditions and have beneficial effect on

rapid germination. In sorghum seeds soaked in $CaCl_2$ solution increased the activity of total amylase and proteases in germinating seeds under salt stress (Kadiri and Hussaini, 1999). In pigeon pea seed treatment with $CaCl_2$ or KNO_3 generally exhibited improvement in proteins, free amino acid and soluble sugars during germinating under salt stress (Jyotsna and Srivastava, 1998).

Zheng *et al*. (1994) reported that rice (*Oryza sativa*) showed earlier and uniform emergence when seeds are osmo-primed with KCl and $CaCl_2$ and mixed salts under flooded conditions. Confounding results, different research workers reported no beneficial results from priming (Mwale *et al*., 2003; Giri and Schillinger, 2003).

2.3 Effect on growth parameters

2.3.1 Shoot length (mm)

Increased shoot and root length may be due to early emergence induced by priming treatment as compared to unprimed seeds (Stofella *et al*., 1992). Kumar *et al*. (2017) experimented on chickpea that shoot length has recorded high in case of osmo-primed seeds than that of unprimed seeds. Among different osmo-priming treatments 20% PEG showed the highest shoot length followed by 4% mannitol and control showed the lowest shoot length. Sarvjeet *et al.* (2017) found that hydro priming of seeds influenced the seedling length of chickpea and maximum seedling length 17.47 cm and 18.07 cm were associated with 16 hours hydro priming.

It was stated by Kaur *et al*. (2002, 2005) that osmo and hydro priming of chickpea seeds with mannitol and water lightened the negative effects of water deficiency and salt stress on seedling development. The treatment of seeds with water, 2% and 4 % mannitol improved the length and biomass of roots and shoots of chickpea seedlings as compared to non-primed controls under salt stressed conditions.

Hydro priming and osmo-priming treatments on shoot length provide significant variation. 20% Polyethylene glycol (PEG) for 24 hr (13.14cm) shows better effect on rest of the treatments except at 100 ml distilled water for 12 hr (12.11 cm) and 20% Polyethylene glycol (PEG) for 12 hr (12.77 cm) on pea (*Pisum sativum*) experimented by Singh *et al*. (2017).

A field experiment was conducted by Gupta and Singh (2012) in inceptisols to find out the effects of seed priming on chickpea. The treatments consisted of seed priming (seed soaking in water for 8 hr). The results revealed that the growth parameters of chickpea were significantly affected by seed priming. Soaking chickpea seeds in water for about 8 hr significantly influenced plant height and nodule dry weight in comparison to un-soaked seeds.

Shehzad *et al*. (2012) conducted an experiment to study the influence of priming techniques on emergence and seedling growth of forage sorghum. Therefore, this study was designed with different seed priming techniques, un-soaked seed (control), Hydro-priming (soaked with distill water), Halo-priming with KNO_3 and $CaCl_2$ (1% solution). All the priming treatments significantly affected the fresh weight, shoot length, number of roots, root length, vigour index, and time to start emergence, time to 50% emergence and energy of emergence of forage sorghum. Seed priming increase cell division and seedling roots which cause an increase in plant height. It is concluded that seed priming may serve as an appropriate treatment for accelerating the emergence of sorghum genotypes studied.

Singh *et al*. (2014) conducted an experiment to study the effect of osmo-priming duration on germination, emergence and early growth of cowpea in Nigeria. Treatment consisted three osmo-priming duration (soaking in 1 % KNO_3 salt for 6, 8 and 10hr) and one hydro-primed control (10 hr). The results showed that osmo-priming with KNO_3 for different durations were superior to unprimed treatment in term of seed germination, emergence, plant height and dry matter accumulation in cowpea. Primed seeds (both osmo-priming and hydro-priming) increased performance of cowpea. However, osmo-priming

with KNO_3 salt (soaked in 1 % KNO_3 salt solution and dried before sowing) for 6 hr could result in greater seed germination and seedling height than hydro-priming.

2.3.2 Root length (mm)

Priming may hasten germination by quickening imbibition, which in turn would simplify the emergence stage and enhancing the division of radicle cells (Kaya *et al*., 2006).

Kulkarni and Eshanna (1988) said that seed treated prior at sowing with 10 ppm IAA increased root length, germination percentage and seedling vigour. Kathiresan *et al*. (1984) also reported similar results, highest root and shoot growth, seedling height and field emergence was attributed with $CaCl_2$ primed seeds in sunflower.

Kumar *et al*. (2017) conducted an experiment on effect of osmo-priming on seed germination behavior and vigour of chickpea (*Cicer arietinum)* and found that 20% PEG and 4% mannitol produce maximum (22.07 cm) root length. Smallest root length was recorded by (15.38cm) with control. Laghari *et al*. (2016) also stated that maximum mean root length cm (5.324) was recorded at Hydro-priming period 4 hours whereas the lower (3.093) found at no priming or check in case of mungbean.

Singh *et al*. (2017) reported that a significant effect of different hydro priming and osmo-priming on root length of pea. 20% polyethylene glycol for 24 hr (14.11 cm) priming showed better performance over untreated (11.98 cm), 3% mannitol for 12 hr (12.37 cm), 3% mannitol for 24 hr (12.67 cm) and 5% glycerol for 12 hr (12.99 cm) priming. Ashraf and Rauf (2001) found that GA_3 treatment enhanced the vegetative growth of two wheat cultivars. It enhanced the deposition of Na^+ and Cl^- in both root and shoots of wheat plant. It also caused a significant increase in photosynthetic at the vegetative stage of the crops.

Sarika *et al*. (2013) conducted a lab experiment to study various physiological and biochemical changes by priming in French bean at Bangalore. They reported that chemo priming with GA_3 and Ethrel improved the seed quality and showed improved seedling length, seedling dry weight which in turn improved higher seedling vigour index, germination speed and mean germination time. Significant increase in initial (6.02 cm) and final (11.5 cm) root length, initial and final shoot length, seedling vigour index and dry seedling weight with GA_3 is observed in the crop.

Chitosan treatment of wheat seeds induced resistance to certain disease and improved seed quality (Reddy *et al*.,1999). Seed soaked with chitosan increased the energy of germination, germination percentage, lipase activity, and gibberellic acid (GA_3) and indole acetic acid (IAA) levels in peanut (Zhou *et al*., 2002). The results showed that the chitosan priming increased the chilling tolerance of maize seedlings demonstrated by improving germination speed and shoot and root growth and maintaining membrane integrity and higher activities of anti-oxidative enzymes. The 0.50% chitosan seems to be a suitable concentration for seed priming it significantly increased seedling growth, root dry weight and root length as compared to control.

2.3.3 Seedling dry weight (mg)

Osmo-priming boosted plumule dry weight was reported by Harris *et al*., (2004).They reported that higher plant dry weight and seed yield following seed priming. The increase in the dry weight and grain yield of mungbean was due to better emergence and better performance per plant by seed priming (Parera and Cantliffe, 1994).

Kumar *et al*. (2017) experimented on chickpea and found that in case of seedling dry weight it was higher (1.02 mg to 1.59mg) in PEG 20% seeds followed by mannitol 4% when compared with control.

Laghari *et al*. (2016) found that, shoot dry weight (mg) has affected by temperature regimes, hydro-priming periods showed highly significant where

as their Interaction was significant for shoot dry weight (mg). The maximum mean shoots dry weight mg (54.74) was recorded at hydro-priming period 4 hours whereas the lower shoot dry weight mg (38.56) found at no priming or check. The maximum mean root dry weight mg (7.898) was observed at hydro-priming period 4 hours whereas the lower root dry weight mg (5.496) found at no priming or check.

At Varanasi, Srivastava and Bose (2012) conducted an experiment on seed priming of rice varieties with or without nitrate salts Mg $(NO_3)_2$ and (KNO_3). Results showed the beneficial effect of priming treatments which was clearly exhibited in plant height, leaf area and number of leaf and yield attribute characteristics i.e. fertile tillers, panicle and grain quality, with nitrate treated varieties. Seed priming treatment resulted in increased crop growth rate in treated sets which encouraged deposition of more photo-assimilates in key plant parts, greatly affecting the dry weight and final yield.

Singh *et al*. (2016) cited on different hydro priming and osmo-priming treatments on dry weight. Polyethylene glycol (PEG) @ 20% for 24 hr (0.54) shows significant effect on Untreated (0.40), Menitol @ 3% for 12 hr (0.44), Menitol @ 3% for 24 hr (0.43), Glycerol @ 5% for 12 hr (0.46) and Glycerol @ 5% for 24 hr (0.48) on dry weight parameters.

Sarika *et al*. (2013) conducted a lab experiment to study various physiological and biochemical changes by priming in French bean at Bangalore. They reported that chemo priming with GA_3 and Ethrel improved the seed quality and showed improved seedling length, seedling dry weight which in turn improved higher seedling vigour index, germination speed and mean germination time. Significant increase in initial (6.02 cm) and final (11.5 cm) root length, initial and final shoot length, seedling vigour index and dry seedling weight with GA_3 is observed in the crop.

2.3.4 Vigour index

During priming, the embryo expands and compresses the endosperm (Liptay and Zariffa, 1993). The compression force of the embryo and hydrolytic activities on the endosperm cell walls may deform the tissues that have lost their flexibility upon dehydration (Lin *et al*., 1993), producing free space and facilitating root protrusion after rehydration and enhance vigour of seed.The probable reason for the highest vigour index might be due to photosynthetic capacity treated with bio fertilizers increases due to increased supply of nutrition (Farnia and Shafie, 2015).

Priming improved seedling vigour experimented by Safiatou (2012). Seedling vigour increased by using seed priming methods in sorghum and Bambara groundnut. Also, highest seedling vigour was achieved by osmo-priming (Mannitol priming) in Bambara groundnut and by hydro-priming in sorghum.

The adverse effects of Reactive Oxygen Species (ROS) lessened and improve the antioxidant enzymes activity through seed priming (Del Ryo *et al*., 2002). It might be meaningfully enhanced the germination rate and vigor of the mungbean seedlings (Umair *et al*., 2010). Harris *et al*. (1999) confirmed that on-farm seeds soaking overnight in water noticeably improved establishment and early vigor of upland rice, maize and chickpea, resulting in faster development, earlier flowering and maturity and higher yields. Also, vigorous growth is often connected with higher yields (Okonwo and Vanderlip, 1985; Austin, 1989; Carter *et al.*, 1992). Seed-treating technology has twofold benefits: greater, speedy and even emergence, with high vigour and better yields in vegetables and floriculture (Bruggink *et al.*, 1999) and some field crops (Basra *et al*., 2005; Kaur *et al.*, 2005).

2.4 Relative water content (%), water saturation deficit (%) and water retention capacity

Relative water content is influenced by seed quality and seed priming technique. Significantly higher relative water content was recorded in leaves

obtained from plots sown with higher quality seeds as compared to those obtained from plots sown with lower quality seeds. The leaves obtained from plots having seed primed with $CaCl_2.2H_2O$ (0.5%) showed significantly highest relative water content which was on par with the leaves from plots having seed primed with KH_2PO_4 (50 ppm) followed by leaves obtained from plots having seed primed with GA_3 (20ppm) (84.57%) while the lowest relative water content (79.02 %) was recorded in leaves obtained from plots having seed primed with KCl (100ppm). The Interaction effect had also a significant effect with the highest relative water content recorded in leaves obtained from plots sown with the higher quality seeds treated by $CaCl_2.2H_2O$ (0.5) (Assefa, 2008).

Baque *et al*.(2002) observed that higher doses of potassium in drought affected wheat generally showed the maximum relative water content, higher water retention capacity and exudation rate. Higher levels of K significantly reduced the water saturation deficit. Fertilizer potassium however, made leaf water potential more negative. The beneficial effect of fertilizer potassium on water stress tolerance in wheat plants were more pronounced under water stressed conditions than under control conditions.

Sangakkara *et al.* (1996) observed that when *Phasiolous vulgaris L*. plants were subjected to moisture stress, the WRC increase with the increasing potassium concentrations.

2.5 Effect of osmo priming against salt stress

Abdoli (2014) set an experiment to evaluate the effects of seed priming on certain important seedling characteristic and seed vigor of fennel (*Foeniculum vulgare* L.) at Department of Agronomy and Plant Breading Faculty of Agriculture, Maragheh University in Maragheh state, Iran. Treatment included untreated seeds (control) and those primed in water (H_2O), sodium chloride (NaCl, 100 mM) and polyethylene glycol 6000 ($PEG\text{-}_{6000}$,water potential-1.6MPa), in darkness for 18 hrs . Among them unsoaked seed (control) and hydro priming treatments had the lowest plumule, radicle and seedling length,

seedling dry weight and seedling vigor index. PEG and NaCl in all of traits were better than the water priming treatments, respectively. PEG-$_{6000}$ (1.6 Mpa) is the best treatment for breaking of fennel seed dormancy.

Aymen *et al.* (2014) conducted an experiment to evaluate the effects of NaCl priming on growth traits and some biochemical attributes of safflower (*Carthamus tinctorius L. cv Safola*) in salinity conditions. Seeds of safflower were primed with NaCl (5 g L^{-1}) for 12 h in 23°C. Primed (P) and non-primed (NP) seeds were directly sown in the field. Experiments were conducted using various water concentrations induced by NaCl (0, 3, 6, 9 and 12 g L^{-1}) in salinity experiment. They found that growth (plant height, fresh and dry weight) and biochemical (chlorophyll, proline and proteins content) of plants derived from primed seeds were greater of about 15 to 30% than that of plants derived from non-primed seeds.

Ajirlo *et al*. (2013) reported that Germination and early growth under prevailing environmental conditions improves by seed priming technique. Their result showed that all the priming treatments significantly affect the fresh weight, shoot length, number of roots, root length, vigor index, time to start emergence, time to 50% emergence and energy of emergence of forage maize. The interactive effect of varieties and priming techniques were not significant for mean emergence time and coefficient of uniformity of emergence.

Seed priming, a controlled hydration process followed by re-drying is pragmatic approach to counteract the salinity effects in many crops because of its simplicity, low cost and effectiveness (Wahid *et al*., 2007; Afzal *et al*., 2011). It improved the germination percentage and uniformity of growth following reduced emergence time and increased yields are reported in many field crops including rice (Farooq *et al*., 2006b; Afzal *et al*., 2006; Afzal *et al*., 2011). But such enhancements are often found under non-saline conditions (Farooq *et al*., 2006a; 2006b) and few studies are available for alleviation of adverse salinity effects in rice during germination and early seedling growth by seed priming (Xu *et al*., 2011). Patade *et al*. (2009) suggest that salt priming is

an effective pre-germination practice for overcoming salinity and drought induced negative effects in sugar-cane.

Farhoudi and Sharifzadeh (2006) while working with canola reported salt priming induced improvement in seed germination, seedling emergence and growth under saline conditions.

The higher germination percentage in seeds primed with $CaCl_2$ is according to Ashraf and Rauf (2001) for wheat who reported an increase in germination percentage of plants raised from seeds primed with calcium salt under salinity stress. Short term seed priming with a low NaCl concentration also increases germination rate, field emergence and acquired stress tolerance (Nakaune *et al*., 2012).

Sun *et al*. (2010) also concluded that PEG priming with moderate concentration resulted in higher tolerance to drought stress than hydro-priming, while higher concentrations of PEG had negative effects on seed germination. It was reported seed priming had significant effect on increment of germination percent; germination speed and seedling dry weight of sunflower in drought condition (Demir *et al*., 2006).

Osmo priming with PEG was described as a good technique for improving seed germination of Bromus seeds under salt and drought stress (Tavili *et al*., 2011) and for increasing the germination percentage and seedling vigor of bersim (*Trifolium alexandrinum*) seeds (Rouhi *et al*., 2010).

Osmo priming with PEG results in strengthening the antioxidant system and increasing the seed germination potential, finally resulting in an increased stress tolerance in germinating seeds of spinach (Chen and Arora, 2011). Osmo conditioning of Italian ryegrass (*Lolium multiflorum*) and sorghum (*Sorghum bicolor*) seeds with 20% PEG-8000 for 2 d at 10°C increased germination percentage, germination rate, seedling establishment and dry matter production under water stress, water logging, cold stress and saline conditions (Hur, 1991).

According to Posmyk and Janas (2007), hydro priming and hydro priming along with proline can be used as a safe priming method for improving seed germination and growth of *Vigna radiata* seedlings at low temperature and also allowing fast repair of injuries caused by stress.

Osmotic seed priming of maize caryopses resulted in more homogenous and faster seed germination as compared to the control was reported by Fotia, *et al*., (2008). Priming with KNO_3 can be used to increase watermelon germination (Demir and Mavi, 2004) and in tomato, seed priming with KNO_3 increased germination percentage, germination index, root length, shoot length and seedling fresh weight (Nawaz *et al*., 2011). It was reported that osmo and hydro priming of chickpea seeds with mannitol and water alleviated the adverse effects of water deficiency and salt stress on seedling growth. The treatment of seeds with water, 2 and 4 % mannitol increased the length and biomass of roots and shoots of chickpea seedlings as compared to non-primed controls under salt stressed conditions (Kaur *et al*., 2005).

Priming of chickpea seeds with mannitol and water improved seedling growth under salt stressed conditions (Kaur *et al*., 2003). Previous studies on tomato (Cuartero *et al*., 2006) and melon (Sivritepe *et al*., 2003), showed that seed priming improves seed germination, seedling emergence and growth under saline conditions. Farhoudi and Sharifzadeh (2006) and Sarwar *et al*. (2006) while working with canola and chickpea, respectively, reported salt priming-induced improvement in seed germination, seedling emergence and growth under saline conditions.

Paul and Choudhury (1991) also observed that seed soaking with 0.5 to 1% solution of KCl or potassium sulfate (K_2SO_4) significantly increased plant height, yield attributes, and grain yield in wheat. The beneficial effects of gibberellic acid (GA_3) on germination are well known (Khan *et al*., 2002). ABA-primed seeds of *Brassica napus* exhibited earlier (2-7 days) germination and higher final percent radicle protrusion than non-primed control seeds,

under salt (100 mM NaCl) or water stress (20 % PEG 8000) and at a low temperature (8 LC) (Gao *et al*., 2002).

Ajouri *et al*. (2004) reported a stimulation of P and Zn uptake, as well as an improved germination and seedling growth in barley after soaking seeds in water and in solutions containing 5-500 mMP. PEG is frequently used to simulate drought stress (Chen *et al*., 2010; Farahani *et al*., 2010) as an inert osmoticum in germination tests (Dodd and Donovan, 1999) and is a non-penetrating solute (Almansouri *et al*., 2001), which results in osmotic stress that inhibits seed germination through the prevention of water uptake. However, it has been reported that the inhibitory effect of PEG on germination may not be solely related to water imbibition (Almansouri *et al*., 2001). Wang *et al*. (2009a) have observed that the fresh weight and the length of the roots and shoots of two alfalfa cultivars (Xinmu No.1 and Northstar) were significantly inhibited by 35% PEG treatment. For a potential medicinal plant, Matricaria chamomilla, both the seed germination rate and seedling growth have been found to be reduced with the PEG- mediated increasing osmotic potential of the growth medium (Afzali *et al*., 2006).

Rouhi *et al*. (2011) also suggested that different priming techniques (hydro and osmo priming) had a varying effects on germination on each of the four grass species (Bromus inermis, Festuca arundinacea, Agropyron e/ongatum and Festuca ovina) and the result showed that, for most evaluated germination parameters, osmo priming treatment (with PEG) was more useful technique to reduce abiotic stress than hydro priming treatment.

Patade *et al*. (2009) suggest that salt priming is an effective pre-germination practice for overcoming salinity and drought induced negative effects in sugar-cane.

Farhoudi and Sharifzadeh (2006) while working with canola reported salt priming induced improvement in seed germination, seedling emergence and growth under saline conditions.

Priming of chickpea seeds with mannitol and water improved seedling growth under salt stressed conditions (Kaur *et al*., 2003). Seed treatment with water and mannitol is also useful under water deficit stress and primed chickpea seeds gave high yield as compared to non-primed seeds (Kaur *et al*., 2002).

Musa *et al*. (1999) reported that overnight priming of chickpea seeds gave better crop production in Bangladesh. Priming with H_2O_2 failed to improve emergence and seedling growth in rice cultivars which is inconsistent with Wahid *et al*. (2007) who reported improved salt tolerance in wheat by alleviation of salt stress and oxidative damage by H_2O_2 pre-treatment.

In basil (*Ocimum basilicum L.*) under saline conditions, the seedling vigor, germination percentage and seedling dry weight was found to increase due to hydropriming (Farahani and Maroufi, 2011). Sivritepe *et al*. (2002) evaluate the effect of salt priming on salt tolerance of melon seedling and reported that total emergence and dry weight were higher in melon seedlings derived from primed seeds and they emerged earlier than non-primed seeds. They also observed that total sugar and proline accumulation and prevented toxic and nutrient deficiency effects of salinity because less Na but more K and especially Ca was accumulated in melon in melon seedlings.

Afzal *et al*. (2005) also found that the priming-induced salt tolerance was associated with improved seedling vigor, metabolism of reserves as well as enhanced K^+ and Ca^{2+} and decreased Na^+ accumulation in wheat plants. Primed crops grew more vigorously, flowered earlier and yielded higher.

Chiu *et al*. (2006) reported that KNO_3 effectively improved germination, seedling growth and seedling vigour index of the seeds of sunflower varieties. Salt priming with KNO_3, is an effective way to improve seed and seedling vigour of sunflower and cucumber (Ghassemi-Golezani and Esmaeilpour, 2008).

Razaji *et al*. (2014) observed that priming is one of the seed enhancement methods that might be resulted to increase seed performance (germination and

emergence) under stress conditions such as salinity, temperature and drought stress. The objective of this study was to evaluate the effects of priming with ascorbic acid on improvement of morphological and biochemical characteristics of rapeseed (*Brassica napus L.*) under simulated drought stress. Results indicated that with increasing in drought stress germination percentage, seedling fresh weight, seedling dry weight, shoot length, root length, and vigor index significantly decreased whereas catalase activity (CAT), peroxidase activity (POX) and Proline content increased as compared to control. However it is concluded that priming resulted improvement in germination components, seedling growth and enzymes activity of rapeseed on drought stress condition and boost the resistance of rapeseed to drought stress condition.

Zamirifar and Bakhtiari (2014) reported that in order to investigate the effects of seed priming, germination percentage and rate, radicel and hypocotyl length and dry weight, root and shoot length, root and shoot and leaves dry weight, leaf number and leaf area per plant was measured. Results showed that *Nigella sativa* germination was sensitive to drought and higher drought intensity resulted in lower germination percentage and rate. Other seedling traits injured by drought too. Seed priming diminished negative effects of drought and higher germination percentage and rate observed in primed seeds. Drought resulted in lower green area in each plant by reducing leaf number and leaf area, thus photosynthesis decreased. Total dry matter aggregation decreased due to low photosynthesis capacity in each plant.

Ajirlo *et al*. (2013) conducted an experiment where seeds were primed for 20, 40 and 60 hours in seven priming media (PEG 5%, PEG 10%, KNO3 1%, KNO_3 2%, KCl 2%, KCl 4% and distilled water as control). Results show that when seed primed by KNO3 2% maximum seed germination percentage was observed. The most seedling length and radical length were obtained for seeds with KCl 2% for 60 h and KCl 4% respectively. Rate of germination was improved when the seed soaked KNO_3 2% compared with PEG, KCL and water. Increasing of seed soaking duration improved some parameters such as

seedlings length, radicle length, stem dry weight and rate of germination. There was Interaction between seed priming media × priming duration showed the beneficial effects on seedling length and number of germination.

Ghiyasi and Tajbakhsh (2013) mentioned that germination and seedling establishment are critical stages in the life cycle of plants especially under stress conditions. The objective of this study was to evaluate the effect of osmo priming on germination and seedling growth of Soybean (*Glycine max L.*) seeds under drought stress. Seeds were primed in aerated solutions of PEG 6000, KNO_3 and KH_2PO_4 have -1.2 MPa osmotic potential. Final germination percentage, time to get 50% germination (T50), seedling vigor index (SVI), germination index (GI), reduction percentage of germination (RPG), seedling dry weight and length were measured. The results indicated that inhibition of germination and seedling growth due to drought stress should be overcome by using osmo priming treatments in soybean. Among the materials used for osmo priming PEG 6000 has the greatest impact on mitigating the effects of drought stress on germination and early growth stages.

Moghanibashi *et al*. (2012) reported that as drought and/or salinity levels increased, germination percentage, germination rate, germination index, root and shoot length and weight and vigour reduced, while mean germination time, coefficient of uniformity of emergence and free proline content increased. The decrease and increase was higher in NaCl than in PEG at the same water potential as the seed was not able to germinate at 0.9 and 1.2 MPa PEG. It was concluded that inhibition of germination at the same water potential of NaCl and PEG resulted from an osmotic effect rather than salt toxicity. However, both the priming treatments clearly improved all of the parameters under drought and salinity conditions. It can be suggested from the results that hydro- and osmo priming enhanced the germination and seedling growth of sunflower under stress conditions. Therefore, these treatments can be used to improve seed performance of sunflower under normal and stress conditions.

A series of experiment were conducted under the laboratory condition of the Department of Agronomy, Sher-e-Bangla Agricultural University, Dhaka from 29 August 2013 to 13 February 2014 to evaluate the effect of pre-sowing seed treatment with Polyethylene Glycol (PEG) on germination behavior of wheat (BARI Gom 27 and BARI Gom 28) in relation to drought tolerance and to optimize priming time of PEG.Nahar (2014) ,showed that wheat seeds primed with 10% PEG and distilled water enhanced germination behavior and seedling growth over nonprimed seeds. The drought tolerant capability of nonprimed and hydro primed seeds decreased drastically as drought stress increased but osmo primed seeds showed considerable tolerance capability upto stress level induced by 10% PEG then significantly decreased with increasing drought stress. Seeds pre-soaked with 10% PEG and distilled water showed better performance in terms of germination behavior and seedling growth compared to untreated control under drought stress. These results suggest that wheat seed primed with 10% PEG for 12 hours is considered as best priming concentration and time to induce drought tolerance capability of wheat for enhancing germination behavior and seedling growth under a certain level of water stress conditions.

2.6 Effect of hydro priming against salt stress

Janmohammadi *et al*. (2009) reported that poor seed germination and crop stand are major problems in saline areas. However, seed vigour enhancement treatments might be able to alleviate the negative effects of salinity. Germination and early growth were affected by both stresses, while at the same osmotic potentials the depressive effect of PEG was more severe than NaCl. Hydro priming significantly improved germination and seedling growth presented as final germination percentage, germination index, seedling vigour index and length of seedling under both stress and non-stress conditions. Hydro priming could alleviate the effects of salinity and drought stress on germination and seedling early growth. This study indicated that hydro priming could be

suitable seed invigoration treatment under saline and drought-prone environments.

Various works have shown that hydro priming of seeds have many advantages as compared to non-primed seeds. Hydro priming has resulted in 3 to 4-fold increases in root and shoot length in comparison with seedlings obtained from non-primed seeds in drought condition (Kaur *et al*., 2002). This phenomenon was explained to be due to faster emergence of roots and shoots, more vigorous plants, better drought tolerance under adverse conditions (Lee-suskoon *et al*., 1998).

Harris *et al*. (1999) also found that hydro priming enhanced seedling establishment and early vigour of upland rice, maize and chickpea, resulting in faster development, earlier flowering and maturity and higher yields.

Hydro priming improved the early and vigorous crop establishment in maize (Nagar *et al*., 1998) .However, other studies resulted in poor emergence from hydro primed Kentucky bluegrass seeds under field conditions (Pill and Necker, 2001).

Moghanibashi *et al*. (2012) reported that the effect of hydro priming for 24 h increased germination percentage, germination rate, germination index, root and shoot length, root and shoot weight of seed sunflower as compared with the control. However, as salinity and/or drought level increased, all of these parameters reduced under both conditions. Primed seeds produced higher germination rate and percentage, D_{50} and GI under all salinity and drought levels as compared with non-primed seeds.

Ghassemi-Golezani *et al*. (2008a) conducted Laboratory tests to evaluate the effects of hydro and osmo priming (PEG: Polyethylene glycol $_{6000}$ at -0.8MPa) on seed germination and field emergence of lentil. Analysis of variance for laboratory data showed that hydro priming significantly improved germination rate and root weights, compared to other seed treatments. However germination percentage for seeds primed with water and PEG were statistically similar, but

higher than those for unprimed seeds. Over all, hydro priming treatment was comparatively superior in laboratory tests. Invigoration of lentil seeds by hydro priming resulted in higher seedling emergence in the field, compared to control and seed priming with PEG. Seedling emergence rate was also enhanced by priming seed with water. Thus, hydro priming could be used as a simple method for improving seed germination and seedling emergence of lentil in the field. Ghassemi-Golezani *et al*. (2008b) mentioned that early emergence and stand establishment of lentil *(Lens culinaris Medik*) are considered to be the most important yield contributing factors in rainfed areas. Analysis of variance of laboratory data showed that hydro priming significantly improved imbibition rate, germination rate, seed vigor index, shoot, root and seedling dry weights and reduced electrical conductivity of seed leachates, compared to other seed treatments. However, germination percentage for seeds primed with KNO_3, water and PEG were statistically similar, but higher than those for unprimed and NaCl priming. Overall, hydro priming treatment was comparatively superior in the laboratory tests. Invigoration of lentil seeds by hydro priming and NaCl priming resulted in higher seedling emergence and establishment in the field, compared to control and seed priming with KNO_3 and PEG. Seedling emergence rate was also enhanced by priming seeds with water, NaCl and KNO_3. It was, therefore, concluded that hydro priming is a simple, low cost and environmentally friendly technique for improving seed and seedling vigor of lentil.

Yağmur and Kaydan (2008) conducted an experiment to find out the effects of seed priming treatments with 0.5% KH_2PO_4 (w/v) solution and water on germination and seedling characters of hexaploid triticale in different osmotic potential of NaCl and PEG solutions. Germination percentage and seedling growth and also relative water content (RWC) decreased with the decrease in osmotic potential of PEG 6000 and NaCl. But root-to-shoot length ratios increased with the effects of osmotic stress of PEG_{6000} and NaCl. Despite the negative effects of two stress conditions, the two priming treatments were effective in improving germination percentage and seedling growth in Presto.

But seed primed treatment was effective at the lowest osmotic potentials; therefore, seedling growth survived at the highest concentrations. Consequently, the effect of hydro priming is very pronounced particularly in improving germination and seedling growth in low stress. Lee *et al*. (1998) mentioned that germination and emergence rates, plumule height, and radicle length of primed seeds were higher than those of untreated seeds at any soil moisture and temperature examined. The time from planting to 50% germination of primed seeds was less than that of untreated seeds by 0.9～3.7 days. Germination rate, emergence rate, plumule height, and radicle length were highest at the soil moisture of 80% field capacity among the soil moistures. Priming effects of rice seeds on germination and emergence rates were more prominent under the unfavorable soil moistures (60, 100, 120, and 140% field capacity) than those under the optimum soil moisture condition (80% field capacity). However, priming effects on seedling growth were greater at near optimum soil moisture compared with too lower or higher soil moistures. Therefore, these findings suggest that priming of rice seeds may be a useful way for better seedling establishment under the adverse soil condition.

Meena *et al.* (2013) conducted an experiment for two consecutive years 2010-11 and 2011-12 to evaluate the influence of hydro priming on the water use efficiency and grain yield of wheat (*Triticum aestivum L.*) under moisture stress. The hydro primed and pre-germinated seeds established earlier than dry seeds leading to better crop establishment under optimum, sub optimum soil moisture as well as dry soil conditions leading to higher tillering and grain yield.

CHAPTER III

MATERIALS AND METHODS

To observe the effect of Polyehylene glycol seed priming for inducing salt tolerance capability in mungbean (*Vigna radiata*) under salt stress, a series of experiment were carried out from 25 May to 15 August, 2018. A illustration of the experimental site, temperature and humidity of the laboratory room, experimental materials, treatments and design, methods of the study, data calculation procedure and data analysis are discussed at this section. The detailed materials and methods that were used to conduct the study are presented below under the following headings:

3.1 Descriptionof the experimental site

3.1.1 Location of the experimental site

The experiment was carried out at the Agronomy Laboratory of Wazed Mia Research Centre , Sher-e-Bangla Agricultural University (SAU), Sher-e-Bangla Nagar, Dhaka-1207. It was located in 24.09^0 N latitude and 90.26^0 E longitudes.

3.1.2 Conditions of laboratory room

The experiment was not conducted at controlled environment. Thats why the temperature and relative humidity of the laboratory room were recorded every day with a digital thermo hygrometer (TERMO, TFA, Germany). The laboratory was at room temperature (average temperature was 25 to 35 C, average minimum and maximum relative humidity was 50% and 80%, respectively).

3.2 Test crops

Two diseased free, healthy mungbean varieties: BARI Mung 6 and BARI Mung 5 were collected from Bangladesh Agricultural Research Institute (BARI) which were used in this experiment.

3.3 Materials for the experiment

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

3.4 Chemicals

Polyethylene glycol and distilled water were utilized as osmo and hydro priming agent. Seed treating chemical was 75% alcohol and salt (NaCl) was used as salt inducing agent.

3.5 Experimental treatments and design

The experiment comprises of

(a) Six levels of priming agent concentration viz. distilled water, 0, 5%, 10%,15% and 20% Polyethylene Glycol (PEG).

(b) Six levels of priming time viz. 3, 6, 9, 12, 15, and 18 hours .

(c) Five levels of salinity stress viz. 0 dS/m, 5 dS/m, 10 dS/m, 15dS/m and 20 dS/m NaCl.

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

3.6 Experimental details

The whole experiment was conducted under three different experiments.

3.6.1. First Experiment

Study on the effect of different concentrations of Polyethylene Glycol on the germination behavior of mungbean.

3.6.1.1Weight of seeds

200 g seeds were weighted from the total seed from each of two mungbean variety BARI Mung 6 and BARI Mung 5 to reduce the unnecessary loss of seeds.

3.6.1.2 Surface treatment

Seeds were treated with 75% alcohol for 5 minutes then were rinsed 2 minutes with distilled water for 3 times to reduce the effect of alcohol from the seed surface. At last, seeds were dried in room temperature to regain the normal condition.

3.6.1.3 Treatments

The experiment was comprised with two mungbean variety and six types of priming solutions.

Factor A : Mungbean variety (02):

1. V_1: BARI Mung 6

2. V_2: BARI Mung 5

Factor B : Six types of treatments:

1. T_1 =Seeds without priming (control)

2. T_2= Seeds primed with distilled water

3. T_3=Seeds primed with 5% Polyethylene glycol solution

4. T_4=Seeds primed with 10% Polyethylene glycol solution

5. T_5=Seeds primed with 15% l Polyethylene glycol solution

6. T_6=Seeds primed with 20% Polyethylene glycol solution

3.6.1.4 Priming agents

5%, 10%, 15%, and 20% of Polyethylene glycol solution and distilled water were used as priming agents.

3.6.1.5 Preparation of priming solutions

a) Polyethylene Glycol (PEG) solutions (5%, 10%, 15% and 20%)

5% PEG solution was prepared by mixing 12.5g of PEG at 250 mL distilled water. Similarly, 25g, 37.5g, 50g PEG was mixed with 250 mL of distilled water to prepare 10%, 15% and 20% solution of PEG (6000) respectively.

b) Distilled water

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

3.6.1.6 Priming technique

Two priming techniques viz., osmo priming and hydro priming were applied on both the mungbean varieties. The surface sterilized seeds were sub-sampled into three parts. One of the sub-samples was considered as control (unprimed) and the other two sub-samples were primed with priming agents. For hydro priming seeds of a sub-sample were soaked in distilled water and for osmo priming seeds of another sub-sample were divided into another four sub-sample and pretreated with Polyethylene Glycol (PEG) at a four levels of concentration of 5%, 10%, 15%, and 20% for 9 hours. Priming was done in different plastic containers covered with lids to prevent evaporation loss. All seeds were removed from the priming solution at the same time. The primed seeds were rinsed thoroughly with distilled water for three times and dried lightly using blotting paper and finally air dried near to original weight (Umair *et al*., 2011) in room temperature for 24 hours back to the original moisture level.

3.6.1.7 Germination of seeds

Thirty seeds from each of the treatments were selected randomly and placed in 90 mm diameter Petri dishes on whatman No.1 filter paper moist with 8 ml of distilled water. Here, whatman No.1 filter paper were used as growth media for germination. Experimental units (60 Petri dishes) were arranged factorialy in a completely randomized design with five replications. During the test filter papers in the Petri dishes were kept saturated with water. Seeds were kept at room temperature 25±1°C under normal light to facilitate germination for 8 days. Germination was considered to have occurred when radicles were 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.These types of abnormal or dead seedlings were excluded during counting. At the end of germination test five seedlings were chosen randomly and seedling growth was measured by dry and fresh weights of different parts of the seedling, by the height of the seedling on the eight day after emergence. 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. Then dry weight was taken.

3.6.1.8 Relative water content (%), water saturation deficit (%) and water retention capacity of shoot

At 8th day of germination test, five seedlings were selected randomly from each treatment and fresh weight was measured immediate after removing roots. Thereafter, the shoots were submerged at distilled water at room temperature in the dark for 24 hr. Shoots turgid weight was measured after removing the excess water by gently wiping with tissue paper. Then shoots were packed in brown paper and oven dried at 75°C for 72 hours for measuring dry weight. The fresh, turgid and dry weights of shoots were utilized to calculate relative water content (%), water saturation deficit (%) and water retention capacity (Baque *et al*., 2002).

3.6.2 Second Experiment

Optimization of pre-sowing priming time on the germination behavior of mungbean.

3.6.2.1 Weight of seeds

Seeds were weighted 200g from the total seed of BARI Mung 6 for this experiment to reduce the unnecessary loss of seeds. Remaining seeds are taken in poly bag and preserved in refrigerator.

3.6.2.2 Surface treatment

Seeds were treated with 75% alcohol for 5 minutes then were rinsed 2 minutes with distilled water for 3 times to reduce the effect of alcohol from the seed surface. Seeds were dried in room temperature to regain the normal condition.

3.6.2.3 Treatments and design

Mungbean variety: BARI Mung 6

Factor A: Priming of seeds

P_1: Osmo priming (10% PEG)

P_2: Hydro priming (Distill water)

Factor B: Six types of priming times

T_1= Seeds primed for 3 hours,

T_2= Seeds primed for 6 hours,

T_3=Seeds primed for 9 hours,

T_4= Seeds primed for 12 hours

T_5= Seeds primed for 15 hours and

T_6= Seeds primed for 18 hours

3.6.2.4 Priming solutions

10% Polyethylene Glycol (PEG) is used as osmo priming and distilled water was used as hydro priming solution.

3.6.2.5 Preparation of priming solutions

a) Polyethylene Glycol (PEG) solutions (10%)

25 g of Polyethylene Glycol (PEG) was dissolved in 250 ml of water to prepare 10% solution of Polyethylene Glycol (PEG).

b) Distilled water

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

3.6.2.6 Priming technique

The surface sterilized seeds were sub-sampled into two parts. Seeds of a sub-sample were divided into six sub-sample soaked in distilled water for six different priming times such as 3,6,9,12,15 and 18hours for hydro priming. For osmo priming the remaining sample of seeds were divided into more six sub-sample and pretreated with Polyethylene Glycol (PEG) for 3, 6,9, 12,15 and 18 hours. Priming is done in different plastic containers covered with lids to prevent evaporation loss. Seeds were removed from the priming solution at the required time. The primed seeds were rinsed thoroughly with distilled water for three times and dried lightly using blotting paper and finally air dried near to original weight (Umair *et al*., 2011) in room temperature for 24 hours back to the original moisture level.

3.6.2.7 Germination of seeds

Thirty seeds from each of the treatments were selected randomly and placed in 90 mm diameter petri dishes on whatman No.1 and filter paper was moistened with 8 ml of distilled water. Here, whatman No.1 filter paper were used as growth media for germination. Experimental units (60 Petri dishes) were

arranged factorialy in a completely randomized design with five replications. During the test filter papers in the Petri dishes were kept saturated condition with water. Seeds were kept at room temperature 25±1°C under normal light to facilitate germination for 8 days. Germination was considered to have occurred when radicles were 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.

3.6.2.8 Relative water content (%), water saturation deficit (%) and water retention capacity of shoot

At 8th day of germination test, five seedlings were selected randomly from each treatment and fresh weight was measured immediate after removing roots. Thereafter, the shoots were submerged at distilled water at room temperature in the dark for 24 hr. Shoots turgid weight was measured after removing the excess water by gently wiping with tissue paper. Then shoots were packed in brown paper and oven dried at 75°C for 72 hours for measuring dry weight. The fresh, turgid and dry weights of shoots were utilized to calculate relative water content (%), water saturation deficit (%) and water retention capacity (Baque *et al*., 2002).

3.6.3 Third experiment

Germination behavior of primed seed (Mungbean) under salt (NaCl) stress condition.

3.6.3.1 Weight of seeds

Seeds were weighted 200g from the total seed of BARI Mung 6 for this experiment to reduce the unnecessary loss of seeds.

3.6.3.2 Surface treatment

Seeds were treated with 75% alcohol for 5 minutes then were rinsed 2 minutes with distilled water for 3 times to reduce the effect of alcohol from the seed surface. At last, seeds were dried in room temperature to regain the normal condition.

3.6.3.3 Treatments

Factor A: Priming

P_1: Osmopriming for 9 hours with 10% PEG

P_2: Hydropriming for 9 hours

Factor B: Salt stress level:

1. T_0= Primed seeds placed without salt 0 dS/m, (control),
2. T_1= Primed seeds placed with 5 dS/m, NaCl,
3. T_2= Primed seeds placed with 10 dS/m, NaCl,
4. T_3= Primed seeds placed with 15 dS/m, NaCl and
5. T_4= Primed seeds placed with 20 dS/m, NaCl

3.6.3.4 Priming solutions and time

10% Polyethylene Glycol (PEG) solution and distilled water were used as priming solutions and 9 hours as priming time.

3.6.3.5 Preparation of priming solutions

a) Polyethylene Glycol (PEG) solutions (10%)

25 g of Polyethylene Glycol (PEG) was dissolved in 250 ml of water to prepare 10% solution of Polyethylene Glycol (PEG) .

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

3.6.3.6 Preparation of salt solutions

Salt (NaCl) solutions (5 dS/m, 10 dS/m, 15dS/m, and 20 dS/m,)

0.731 g of sodium chloride (NaCl) was dissolved in 250 ml of water to prepare 5 dS/m, solution of salt (NaCl). Similarly, 1.436 g, 2.18 g, 2.925 g sodium chloride (NaCl) was dissolved in 250 ml of water to prepare 10 dS/m, 15 dS/m and 200dS/m solution of sodium chloride (NaCl), respectively.

3.6.3.7 Priming technique

Two priming techniques viz. osmo priming and hydro priming were applied on BARI Mung 6. The surface sterilized seeds were sub-sampled into two parts. One of the sub-samples was soaked in distilled water for hydro priming and seeds of another sub-sample were pretreated with PEG for osmo priming at a concentration of 10% for 9hours, respectively. Priming is done in different plastic containers covered with lids to prevent evaporation loss. All seeds were removed from the priming solution at the same time. The primed seeds were rinsed thoroughly with distilled water for three times and dried lightly using blotting paper and finally air dried near to original weight (Umair *et al.*, 2011) in room temperature for 24 hours back to the original moisture level.

3.6.3.8 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 primed and control seeds were irrigated with solutions of 8 ml salt stress levels. Here whatman No.1 filter paper were used as growth media for germination .Experimental units (75 Petri dishes) were arranged factorialy in a completely randomized design with five replications. During the test filter papers in the Petri dishes were kept water saturated state. Seeds were kept at room temperature 25±1°C under normal light to facilitate germination for 8 days. Germination was considered to have occurred when radicles were 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.

3.6.3.9 Relative water content (%), water saturation deficit (%) and water retention capacity of shoot

At 8th day of germination test, five seedlings were selected randomly from each treatment and fresh weight was measured immediate after removing roots. Thereafter, the shoots were submerged at distilled water at room temperature in the dark for 24 hr. Shoots turgid weight was measured after removing the excess water by gently wiping with tissue paper. Then shoots were packed in brown paper and oven dried at 75°C for 72 hours for measuring dry weight. The fresh, turgid and dry weights of shoots were utilized to calculate relative water content (%), water saturation deficit (%) and water retention capacity (Baque *et al*., 2002).

3.7 Data recording

Parameters that are measured as follows:

3.7.1 Total germination (TG%)

Total germination (TG) was calculated as the number of seeds which was germinated within 8 days as a proportion of number of seeds shown in each treatment, expressed as a percentage (Othman *et al*., 2006).

$$TG\ (\%) = \frac{\text{number of germinated seeds}}{\text{total number of seeds set for germination}} \times 100$$

Where, n = number of seeds germinated on day D, and D = number of days counted from the beginning of germination.

3.7.2 Coefficient of velocity (CV)

Coefficient of velocity (CV) = (number of germinated seeds per day) is measured according to Kader and Jutzi (2004) formula.

$CV = (\Sigma Ni / 100) \times (\Sigma Ti\ Ni)$

Where,

Ti= number of days after sowing and

Ni = number of seeds germinated on ith day.

3.7.3 Shoot (mm), root (mm) and seedling length (mm)

Randomly selected 5 seedlings from each treatment were collected and cotyledons were removed from them. Shoot, root and seedling length was measured with a ruler and accuracy of measurement was 1 mm.

3.7.4 Shoot dry weight (mg), root dry weight (mg)

By using an electrical balance, the dried shoots and roots were weighted to the nearest milligram (mg).

3.7.5 Vigour Index (VI)

Vigour Index (VI) was calculated from total germination and seedlings length by using the formula of Abdul- Baki and Anderson (1970).

$$VI = \frac{TG\ (\%) \times \text{seedlings length (mm)}}{100}$$

Here, TG = total germination.

3.7.6 Relative Water Content (RWC %)

Relative water content was calculated from the fresh, turgid and dry weights of shoots by using the following formula used by Baque *et al*. (2002). Relative water content expressed in percentage.

$$\text{Relative Water Content (RWC)} = \frac{\text{Fresh wt.} - \text{Dry wt.}}{\text{Turgid wt.} - \text{Dry wt.}} \times 100$$

3.7.7 Water Saturation Deficit (%)

Water saturation deficit was calculated from RWC by using the following formula used by Baque *et al.* (2002).

Water saturation deficit (WSD) = 100- Relative water content

3.7.8 Germination coefficient (GC) %

Germination coefficient (GC) was calculated using the following formula Copeland, L., 1976.

$$\text{Germination coefficient (\%)} = \frac{A1 + A2 + \ldots\ldots + Ax}{A1T1 + A2T2 + \ldots\ldots AxTx} \times 100$$

Where,
A= Number of seeds germinated
T= Time corresponding to A
x= Number of days to final count.

3.7.9 Water retention capacity (WRC)

Water retention capacity (WRC) was measured following formula of Baque, karim and Hamid (2002)

$$\text{Water retention capacity (WTC)} = \frac{\text{Turgid weight}}{\text{Dry weight}}$$

3.8 Statistical analysis

The data obtained for different parameters were statistically analyzed to observe the significant difference among the treatments. The mean value of all the parameters was calculated and analysis of variance was performed. The significance of difference among the treatments means was estimated by the least significant difference (LSD) test at 1% level of significance. A computer software MSTAT-C was used to carry out the statistical analysis.

CHAPTER IV

RESULTS AND DISCUSSION

The experiment was carried out to investigate the effect of seed priming and priming time on germination behavior of Mungbean in relation to salinity tolerance.

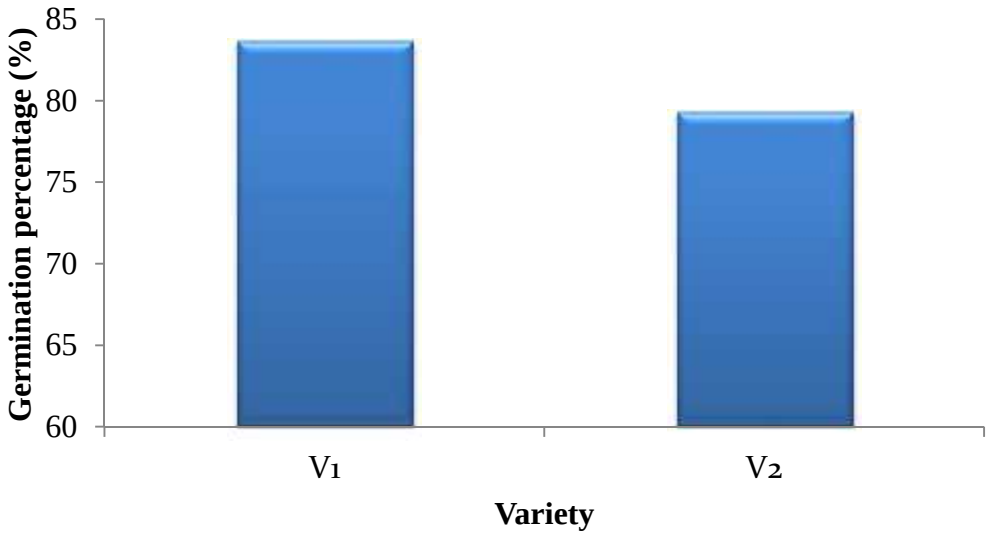
4.1 Experiment 1. Study on the germination behavior of Mungbean at different concentrations of priming agents (PEG) and distilled water

Results obtained from the present study regarding the germination behavior of Mung at different concentrations of priming agents have been demonstrated and discussed. The analytical results have been displayed in Figures 1 to 10 and Tables 1 to 3.

4.1.1 Germination percentage:

4.1.1.1 Effect of variety

Mungbean varieties exhibited significant difference in respect of the germination percentage (Fig. 1). Among the varieties, BARI Mung 6 (V_1) showed the maximum germination percentage (83.57%) and BARI Mung 5 (V_2) showed minimum germination percentage (79.21%). This result is corroborates with previous research work of Ajirlo *et al*. (2013). Results indicated that the, variety V_1 showed the highest germination rate at all concentration of PEG ,water and control treatment where variety V_2 (BARI Mung 5) showed the lowest germination rate at all PEG concentration, water and control treatment.The highest germination percentage (95.33%) of BARI Mung 6 was observed in 4% mannitol primed seed (Abdullah, 2017) **.** Ajirlo *et al*. (2013) reported that germination is improved by seed priming technique. He also reported that seed priming treatments reduced the mean emergence time and promoted germination.



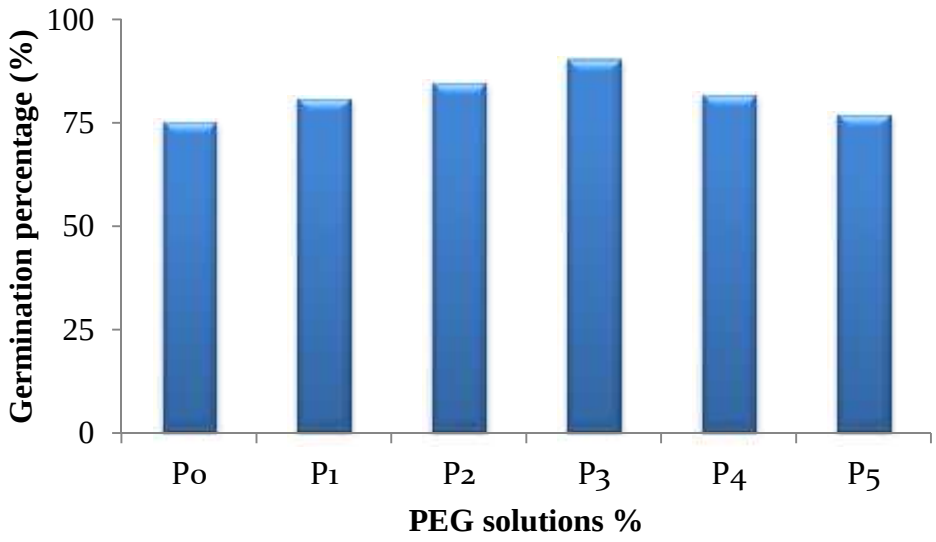
V_1= BARI Mung 6, V_2= BARI Mung 5

Figure 1. Effect of variety on the germination percentage of Mungbean ($LSD_{0.01}$=3.17)

4.1.1.2 Effect of priming solutions:

Germination percentage of Mungbean was influenced by various Polyethylenene Glycol (PEG) concentrations (Fig. 2) and there was completely significant difference between control (nonprime seeds) and primed seeds. Germinatio percentage (GP%) increases with increasing PEG concentration up to 10% then decreases gradually with increasing PEG concentration. P_3 (primed with 10% PEG concentration) showed the maximum germination percentage (90.05%) and there after decrease due to increasing concentration of PEG. The lowest germination percentage 74.91 was found in P_5 which is statistically similar with P_0. The result of the present study corroborates with the study of previous researchers (Ajouri *et al*., 2004; Farooq *et al*.,2006; Sun *et al*., 2010). According to Rahman 2014, Priming with PEG can be used as increasing germination up to certain level. According to Ajouri *et al*. (2004) priming induces a range of biochemical changes in the seed that required initiating the germination process i.e., breaking of dormancy, hydrolysis or metabolism of inhibitors, imbibitions and enzymes activation. According to, Nahar *et al.,* (2017), Germination percentage (GP%) is increases with

increasing PEG concentration up to 10% then decreases gradually with increasing PEG concentratn.



P_0 = Seeds without priming (control); P_1 = Seeds primed with 0% PEG for 9 hours; P_2 = Seeds primed with 5% PEG for 9 hours, P_3 = Seeds primed with 10% PEG for 9 hours; P_4 = Seeds primed with 15% PEG for 9 hours, P_5 = Seeds primed with 20% PEG for 9 hours

Figure 2. Effect of priming solutions on the germination percentage of mungbean (LSD $_{0.01}$=5.48)

4.1.1.3 Interaction effect of variety and priming solutions

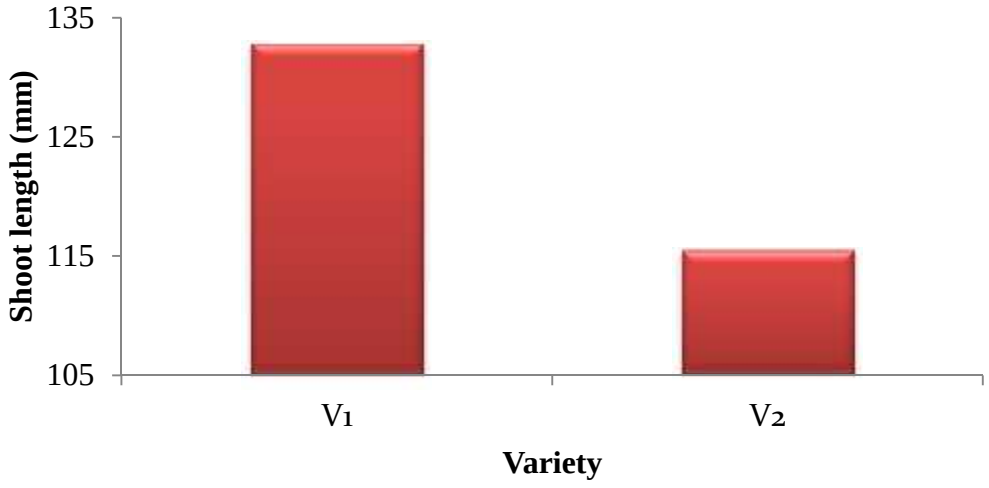
Germination percentage of Mungbean genotypes significantly is influenced by different PEG concentrations (Table.1).Germination percentage (GP%) increases with the increasing of PEG concentration up to 10% then there was a gradual decrease observed with increasing the PEG concentration. The result of the experiment revealed that the maximum germination percentage (94.59%) was recorded from Mungbean genotype BARI Mung 6 with 10% PEG solution (V_1P_3) and the minimum germination percentage (73.78%) was recorded from Mungbean genotype BARI Mung 5 with control (V_2P_0) which is statistically similar with V_1P_5,V_2P_1, V_1P_0 and V_2P_5 .The result of the experiment collaborated with the results of Baque *et al.* (2002), Kaya *et al.* (2006), Afzal *et al. (2006)*, Basra *et al.* (2003), and Demir *et al*. (2006) . Baque *et al.* (2016) concluded that 10% PEG was best for improing germination behavior of wheat.

The report of the Sun *et al.* (2010) revealed the compatible PEG concentration was 20% for Gangyou 527 (*indica* hybrid rice) and 10%-15% for Nongken 57 (conventional *japonica* rice). At the time of germination, PEG concentration above the optimum level had adverse effects. Singh *et al.* (2015) showed that over-priming (KNO_3) prolonged soaking in primed solution (KNO_3) that might have injured the cellular organile.

4.1. 2 Shoot length (mm)

4. 1.2.1 Effect of variety

Mungbean variety exhibited significant difference in respect of the shoot length (Fig. 3). Among the varieties, BARI Mungbean 6 (V_1) showed the greater shoot length (132.71 mm) and BARI Mungbean 5 (V_2) showed smaller shoot length (115.40 mm). Significant difference was recorded on shoot length with different concentrations of PEG and water for BARI Mung 6 and Binamoog 5, Shoot length was increased up to 10% for BARI Mung 6 compared to Binamoog 5 (Abdullah, 2017). According to, Nahar (2014), the highest plumule length of BARI Gom 27 was obtained from seeds pre-treated with 10% PEG solution over BARI Gom 28.

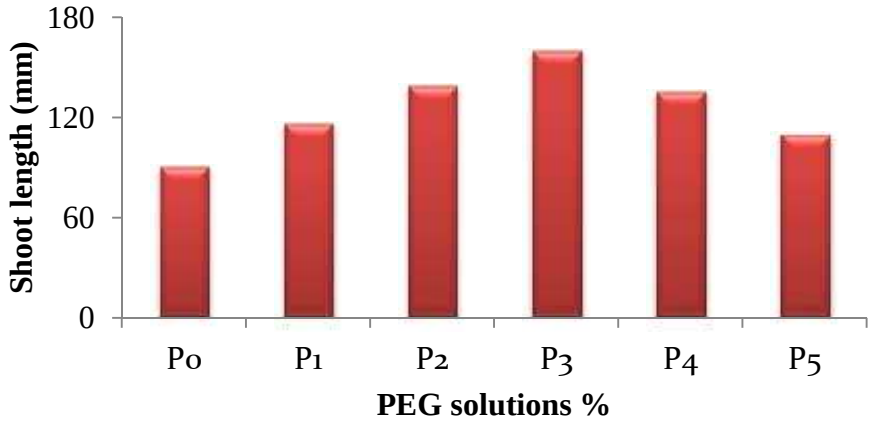


V_1: BARI Mung 6 V_2: BARI Mung 5

Figure 3. Effect of variety on the shoot length of mungbean ($LSD_{0.01}$=4.69)

4.1.2.2 Effect of priming solutions

Priming with different PEG concentrations and water showed higher amount of variation in shoot length over nonprime seed (Fig. 4). Shoot length was affected by hydro priming and osmo priming (PEG). Shoot length increases with increasing PEG concentration up to 10% then decreases gradually with increasing PEG concentration. P_3 (primed with 10% PEG concentration) showed the greater shoot length (72.14 mm) and there after decrease due to increasing concentration of PEG. The lowest shoot length was found in P_0 (89.35 mm). According to Kumar *et al*. (2017), chickpea shows that shoot length has recorded high in case of osmo-primed seeds than that of unprimed seeds. According to Nahar *et al.* (2014), plumule length (mm) of wheat was influenced by various Polyethylene Glycol (PEG) concentrations and variance analysis results showed that there was significant difference between control (non primed seeds) and primed seeds. Kaur *et al.* (2002), reported that shoot length and shoot biomass of water and mannitol primed plants were greater compared to these from non-primed plants.



P_0 = Seeds without priming (control); P_1 = Seeds primed with 0% PEG for 9 hours; P_2 = Seeds primed with 5% PEG for 9 hours, P_3 = Seeds primed with 10% PEG for 9 hours; P_4 = Seeds primed with 15% PEG for 9 hours, P_5 = Seeds primed with 20% PEG for 9 hours

Figure 4. Effect of different priming solutions on the shoot length of mungbean ($LSD_{0.01}$=8.11)

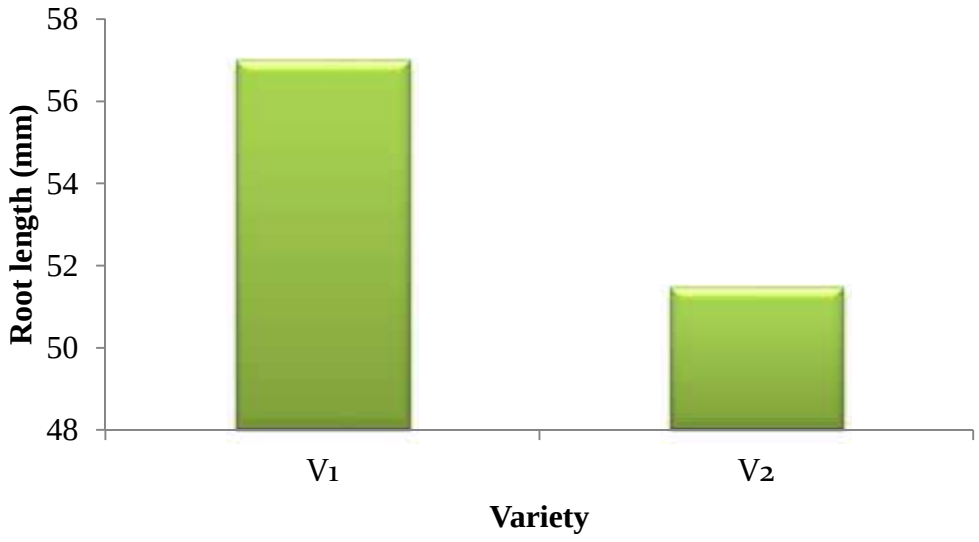
4.1.2.3 Interaction effect of variety and priming solutions

Shoot length of Mungbean varieties significantly affected by different PEG solutions (Table.1). Shoot length of Mungbean genotypes increased with the increasing of PEG concentration up to 10% and there after a gradual decreased observed with increasing the PEG concentration. The result of the experiment revealed that the maximum shoot length (171.6 mm) was recorded from Mungbean genotype BARI Mung 6 (V_1) with 10% PEG concentration (V_1P_3) whereas the minimum shoot length (85.22 mm) was recorded from BARI Mung 5 (V_2) with control treatment (V_2P_0) which is statistically similar with V_1P_0 . This result is in agreement with the findings of several workers Baque *et al*. (2016). Rennick and Tiernan (1978) reported that there was a rapid and more extended elongation of coleoptile occurred in treated seeds than non-treated and over primed seeds. Lee and Kim (2000), revealed that, priming increased the metabolic activities of seed ultimately gained the substantial shoot length than nonprimed seed.

4.1.3. Root length (mm)

4.1.3.1 Effect of variety

Mungbean varieties exhibited significant difference in respect of the root length (Fig. 5). Among the varieties, BARI Mung 6 (V_1) showed the maximum root length (56.96 mm) and BARI Mung 5 (V_2) showed smaller root length (51.45 mm).According to Abdullah, (2017), root length was improved up to 6% and 4% of mannitol concentration for BARI Mung 6 than Binamoog 5, respectively and therefore decreased gradually with increasing mannitol concentration for both varieties.

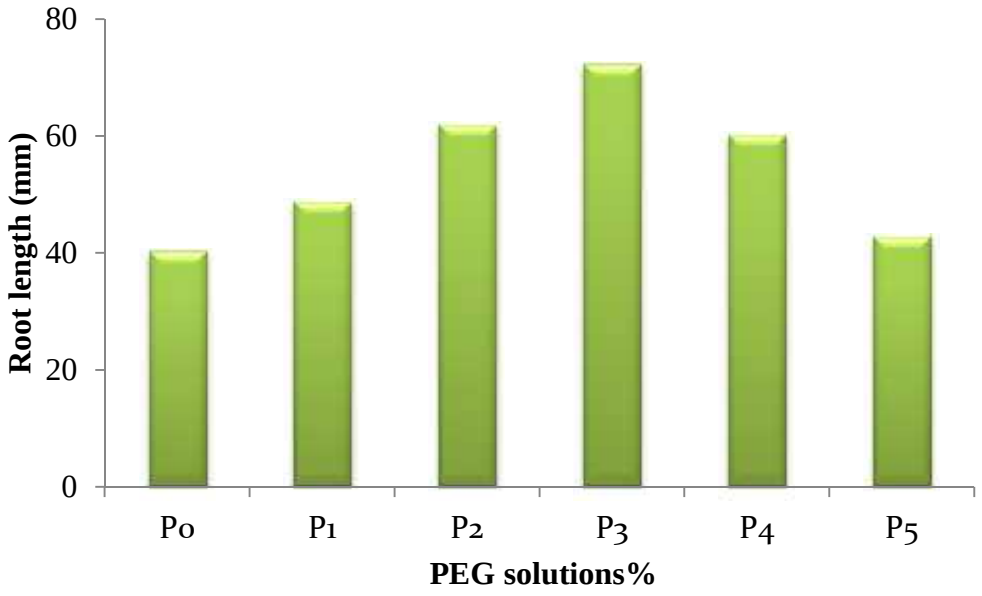


V_1: BARI Mung 6 V_2: BARI Mung 5

Figure 5. Effect of variety on the root length of mungbean ($LSD_{0.01}$=2.16)

4.1.3.2 Effect of priming solutions

Osmo priming with different concentrations of PEG and water showed significant variation in respect of root length (Fig. 6) .Root length increases with increasing PEG concentration up to 10% then decreases gradually with increasing PEG concentration. P_3 (primed with 10% PEG concentration) showed the highest root length (72.14mm) and thereafter decrease due to increasing concentration of PEG. The lowest shoot length was found in P_0 (40.08 mm), which is statistically similar with P_5. Sarwar *et al*. (2006) who reported that root length were better when treated with water and PEG over hydro primed and control. Baque *et al*. (2016), showed that priming seed with PEG can increase germination capacity, root length, length of plumule, seedling dry weight and plant height than the dry seed.



P_0 = Seeds without priming (control); P_1 = Seeds primed with 0% PEG for 9 hours; P_2 = Seeds primed with 5% PEG for 9 hours, P_3 = Seeds primed with 10% PEG for 9 hours; P_4 = Seeds primed with 15% PEG for 9 hours, P_5 = Seeds primed with 20% PEG for 9 hours

Figure 6. Effect of different priming solutions on the root length of Mungbean ($LSD_{0.01}$=3.73)

4.1.3.3 Interaction effect of variety and priming solutions

For root length, highest root length was recorded from treatment combination V_1P_3 (76.09 mm) and lowest root length was showed in treatment combination V_2P_0 (37.80 mm) which is statistically similar with treatment combination V_1P_0,V_1P_5 and V_2P_5.Nahar (2014),found hight root length at 10% PEG and lowest from nonprime seed.Root length increased for up to 10 % PEG treatment combination incase of both variety but decreases for further increased.

Table 1. Interaction effect of variety and different priming concentrations on the germination and growth behaviors of mungbean

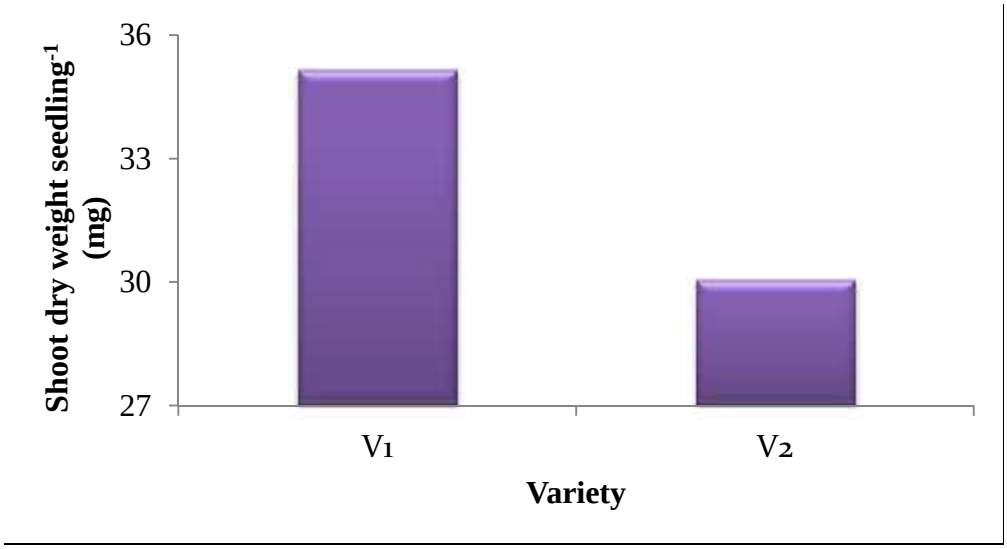
Treatment combinations	Germination percentage (%)	Shoot length (mm)	Root length (mm)	Shoot dry weight (mg)	Shoot dry weight (mg)
V_1P_0	76.43 d-f	93.49 fg	42.37 fg	25.25 g	12.10 hi
V_1P_1	82.38 b-e	125.4 c	51.90 e	29.84 d-f	15.45 de
V_1P_2	85.99 b	148.1 b	63.60 bc	41.15 b	17.87 b
V_1P_3	94.59 a	171.6 a	76.09 a	49.13 a	19.86 a
V_1P_4	84.02 b-d	144.4 b	62.70 cd	37.53 c	16.81 b-d
V_1P_5	78.00 c-f	113.2 de	45.11 f	27.78 e-g	13.36 gh
V_2P_0	73.38 f	85.22 g	37.80 g	22.03 h	10.86 i
V_2P_1	78.63 b-f	105.1 e	45.21 f	26.81 fg	13.76 fg
V_2P_2	82.90 b-e	127.6 c	59.93 cd	31.76 d	15.80 c-e
V_2P_3	85.50 bc	145.7 b	68.19 b	43.78 b	17.14 bc
V_2P_4	79.27 b-f	123.9 cd	57.49 d	30.84 de	15.02 ef
V_2P_5	75.55 ef	104.9 ef	40.07 fg	24.92 gh	11.85 i
$LSD_{(0.01)}$	7.74	11.47	5.28	3.20	1.38
CV (%)	5.61	5.45	5.74	5.79	5.42

V_1P_0: Nonprimed seeds of BARI Mung 6 , V_1P_1: BARI Mung 6 seeds primed with Distilled water , V_1P_2: BARI Mung 6 seeds primed with 5% PEG, V_1P_3: BARI Mung 6 seeds primed with 10 % PEG, V_1P_4: BARI Mung 6 seeds primed with 15 % PEG , V_1P_5: BARI Mung 6 seeds primed with 20 % PEG , V_2P_0: Nonprimed seeds of BARI Mung 5 , V_2P_1: BARI Mung 5 seeds primed with Distilled water, V_1P_2:BARI Mung 5 seeds primed with 5% PEG, V_2P_3: BARI Mung 5 seeds primed with 10 % PEG ,V_2P_4: BARI Mung 5 seeds primed with 15 % PEG, V_2P_5: BARI Mung 5seeds primed with 20 % PEG

4.1.4 Shoot dry (mg)

4.1.4.1 Effect of variety

Mungbean variety exhibited significant difference in respect of the shoot dry weight seedling $^{-1}$ (Fig. 7). Among the varieties, BARI Mung 6 (V_1) showed the maximum shoot dry weight (35.11 mg) and BARI Mungbean 5 (V_2) showed lower shoot dry weight seedling^{1} (30.02 mg).

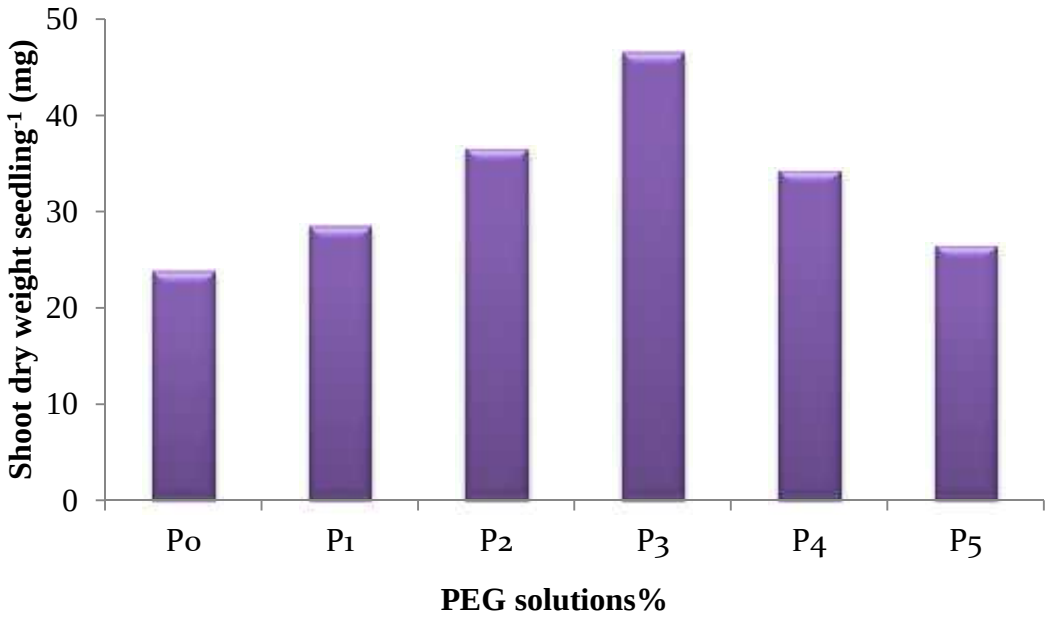


V_1: BARI Mung 6 V_2: BARI Mung 5

Figure 7. Effect of variety on the shoot dry weight seedling-1 of Mungbean ($LSD_{0.01}$=1.31)

4.1.4.2 Effect of different priming solutions

Priming with different concentrations of PEG and water showed significant variation in respect of shoot dry weight seedling $^{-1}$ (Fig. 8). Shoot dry weight is affected by hydro priming and different PEG solutions. Shoot dry weight increases with increasing PEG concentration up to 10% and decreases due to increasing concentration. P_3 (primed with 10% PEG concentration) showed the highest (46.45mg) and thereafter decrease due to increasing concentration of PEG. The lowest shoot dry weight seedling $^{-1}$ was found in P_0 (23.64 mg). According to Harris *et al*. (2004) higher plumule dry weight due to osmo priming was reported over nonprimed. Sarika *et al*. (2013) conducted a lab experiment to study various physiological and biochemical changes by priming in French bean at Bangalore. They reported that chemo priming with GA_3 and Ethrel improved the seed quality and showed improved seedling length, seedling dry weight which in turn improved higher seedling vigour index, germination speed and mean germination time.



P_0 = Seeds without priming (control); P_1 = Seeds primed with 0% PEG for 9 hours; P_2 = Seeds primed with 5% PEG for 9 hours, P_3 = Seeds primed with 10% PEG for 9 hours; P_4 = Seeds primed with 15% PEG for 9 hours, P_5 = Seeds primed with 20% PEG for 9 hours

Figure 8. Effect of different priming solutions on the shoot dry weight seedling-1 of Mungbean (LSD 0.01=2.26)

4.1.4.3 Interaction effect of variety and priming solutions

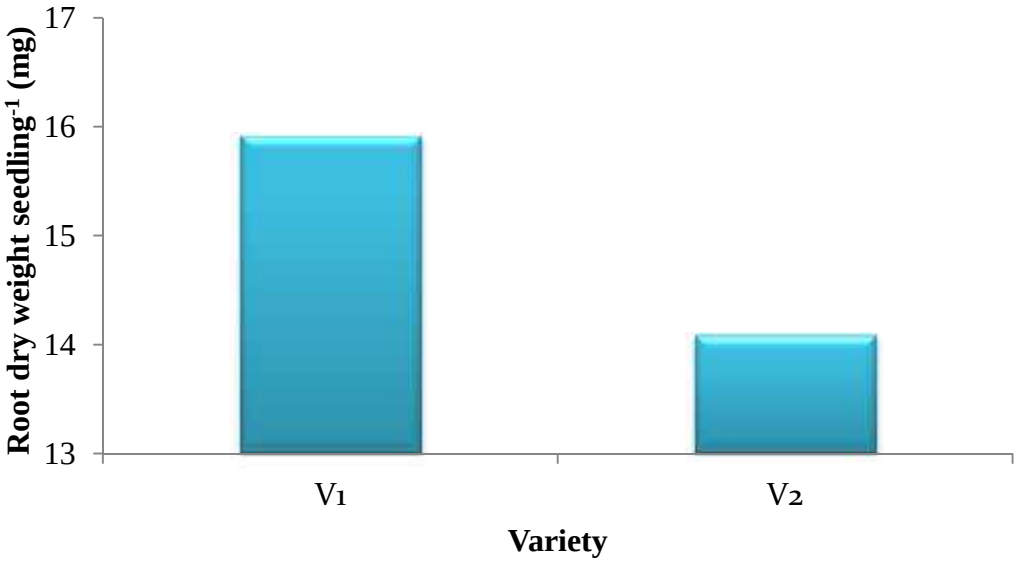
The Interaction between variety and different priming agents (Distilled water and different PEG solutions) revealed that the highest shoot dry weight (49.13 mg) was scored by BARI Mungbean 6 with 10% PEG concentration (V_1P_3) whereas the minimum shoot dry weight (22.03 mg) was recorded from BARI Mungbean 6 with non-primed seed (V_1P_0) (Table.1) which is statistically similar with V_2P_5. The result of the present study was also supported by the result of previous researchers Baque *et al*. (2016), Khalil *et al*. (2010), Ghassemi-Golezani *et al*.(2008a) and Sarwar *et al*.(2006). Priming presumably permitted some repairs of damaged to membrane caused by deterioration and exerted better germination pattern and higher vigor level than non- primed Ruan *et al*. (2002). Basra *et al.* (2005) showed that the refinement in germination and vigor of standard/low-vigor seed might be due to preserve transportation of food material, trigger and re-synthesis of some enzymes,

DNA and RNA synthesis start during osmotic priming. Removing of obstacle speed up the germination of seed ultimately produced the vigorous shoot and increased shoot dry weight of wheat genotypes. Baque *et al*. (2016) found that maximum shoot dry weight was recorded when the seed primed with 10% PEG solution compared to that of osmo and hydro primed seed.

4.1.5 Root dry weight (mm)

4.1.5.1 Effect of variety

Mungbean variety exhibited significant difference in respect of the root dry weight seedling $^{-1}$ (Fig.9). Among the varieties, BARI Mungbean 6 (V_1) showed the maximum root dry weight (15.91 mg) and BARI Mungbean 5 (V_2) showed minimum root dry weight seedling $^{-1}$ (14.07 mg).

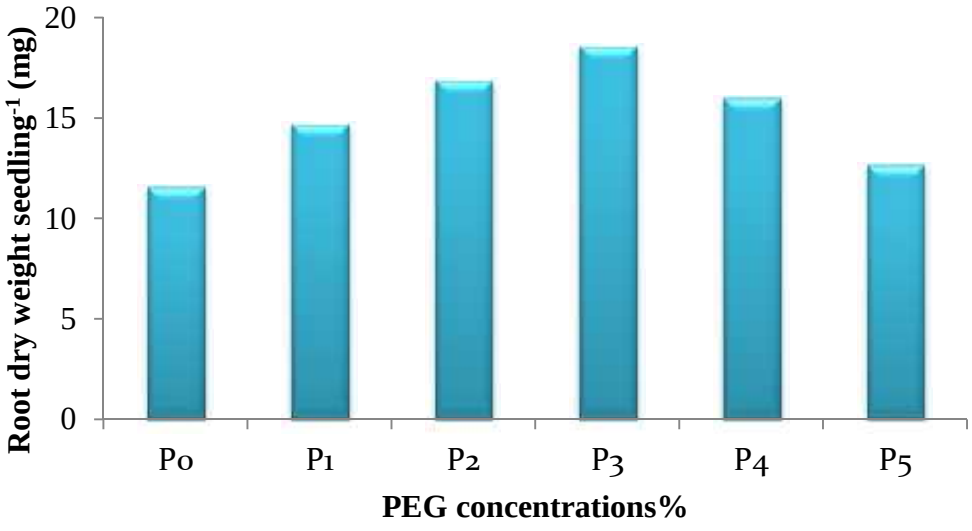


V_1: BARI Mung 6 V_2: BARI Mung 5

Figure 9. Effect of variety on the root dry weight seedling^{-1} of mungbean ($LSD_{0.01}$=0.56)

4.1.5.2 Effect of priming solutions

Priming with PEG solutions and water showed significant variation in respect of shoot dry weight seedling $^{-1}$ (Fig. 8).P_3 (primed with 10% PEG concentration) showed the highest (18.50mg) and there after decrease due to increasing concentration of PEG. The lowest shoot dry weight seedling $^{-1}$ was found in P_0 (11.48 mg). Moradi *et al*. (2012) reported that the highest radical length of wheat grass was obtained from seeds pre-treated with 10% PEG solution over control and osmo primed seeds, respectively. Sarwar *et al*., (2006) reported that root length were better when treated with water and mannitol over control.



P_0 = Seeds without priming (control); P_1 = Seeds primed with 0% PEG for 9 hours; P_2 = Seeds primed with 5% PEG for 9 hours, P_3 = Seeds primed with 10% PEG for 9 hours; P_4 = Seeds primed with 15% PEG for 9 hours, P_5 = Seeds primed with 20% PEG for 9 hours

Figure 10. Effect of different priming solutions on the root dry weight seedling-1 of mungbean ($LSD_{0.01=0.97}$)

4.1.5.3 Interaction effect of variety and priming solutions

Interaction between variety and priming agents showed significant difference on root dry weight seedling^{1}, highest root dry weight was recorded from V_1P_3 (19.86 mg) and lowest data was recorded from V_2P_0 (10.86 mg) which is

statistically similar with V_2P_5 and V_1P_0. Baque *et al*. (2016) reported that 10% PEG was best for improved root dry weight of wheat. According to result it is said that Interaction of variety and different concentration of PEG shows significant variation in case of germination percentage, shoot length, root length, shoot dry weight and root dry weight. BARI 6 with 10% PEG showed better performance than any other treatment combination. PEG concentration up to 10% with BARI Mungbean 6 were gradually increase in case of above parameters and were gradually decrease more than 10% PEG concentration.

4.1.6 Relative water content%

4.1.6.1. Effect of variety

RWC could be the perfect indicator of plant hydrologic condition as it denotes the physiological consequences of cellular water deficit. Mungbean variety exhibited significant difference in respect of relative water content (Table 2). Among the varieties, BARI Mung 6 (V_1) indicated the higher relative water content (76.25%) than BARI Mungbean 5 (V_2) which showed relative water content 68.71%.Abdullah (2017), reported that relative water content of BARI Mung 6 was higher than Binamoog 5.

4.1.6.2 Effect of priming solutions

Priming with different concentrations of PEG and water showed significant variation in respect of relative water content (Table 2). Relative water content was increased up to 10% PEG and decreased when this concentration increased. P_3 (primed with 10% PEG concentration) showed the highest (93.14%) relative water content. The lowest relative water content was found in P_0 (52.90%). A similar finding was reported by Sairam *et al*. (2002). Nahar *et al*. (2017) revealed that the maximum relative water content (90.05%) was found from 10% PEG concentration and the minimum relative water content (61.63%) was found from 0% PEG concentration in wheat genotype. Under stress condition, osmo and hydro primed seedling can thrive and provide better water use efficiency (Flower *et al.*,1998).

4.1.6.3 Interaction effect of variety and priming solutions

Interaction between variety and priming agents showed significant difference on relative water content seedling[1].Maximum relative water content was obtained from treatment combination V_1P_3 (94.88%) which is statistically similar with treatment combination V_2P_3. Lowest data recorded from treatment combination V_2P_0 (47.24%) which is statistically similar with V_2P_5. Sairam *et al.*(2002), revealed that the maximum relative water content was found maximum under 10% PEG concentration and the minimum relative water content was found from 0% PEG concentration.

4.1.7 Water saturation deficit %

4.1.7.1 Effect of variety

Water saturation deficit of Mungbean genotypes influenced significantly by different priming solutions (Table 2). Among the varieties, BARI Mungbean 5 (V_2) showed the higher water saturation deficit (31.29%) than BARI Mungbean 6 (V_1) which showed lower water saturation deficit 23.72%.Abdullah (2017) revealed that the maximum water saturation deficit was found maximum from primed seed BARI Mung 5 and the minimum water saturation deficit was found from BARI Mung 6 .

4.1.7.2 Effect of priming solutions

Priming with different concentrations of PEG and water showed significant variation in respect of water saturation deficit (Table 2). It followed the opposite trend compared to the previously described parameter, *i.e.,* the water saturation deficit was maximum at 0% PEG concentration and gradually decreased up to 10% PEG concentration and then steadily increased. P_0 (control) showed the highest (47.10%) and the lowest water saturation deficit was found in P_3 (6.865%). The water saturation deficit was maximum at 0% PEG concentration and gradually decreased up to 10% PEG concentration and then steadily increased. Baque *et al*. (2002),due to lack of defense mechanism,

the non-primed seedling failed to uptake enough water necessary for running the physiological process smoothly than the primed seedling. Thus there was a massive water deficit occurred in case of non-primed genotypes than the primed genotypes.

4.1.7.3 Interaction effect of variety and priming solutions

Combined effect of variety and priming concentration showed significant variation in case of water saturation deficit (Table.3).When water saturation deficit of two Mungbean varieties was analyzed, highest water saturation deficit was found from treatment combination V_2P_0 (52.76%) which was statistically different from others and lowest was found from treatment combination V_1P_3 (5.120%). Due to lack of defense mechanism, the non-primed seedling failed to uptake enough water necessary for running the physiological process smoothly than the primed seedling. Thus there was a massive water deficit occurred in case of non-primed genotypes than the primed genotypes.

4.1.8. Water retention capacity

4.1.8.1 Effect of variety

Muingbean varieties exhibited significant difference in respect of water retention capacity (Table 2). Among the varieties, BARI Mungbean 6 (V_1) showed the higher water retention capacity (9.30) than BARI Mungbean 5(V_2) which showed lower water retention capacity (8.75).

4.1.8.2 Effect of priming solutions

Priming with different concentrations of PEG and water showed significant variation in respect of water retention capacity (Table 2). P_3 (primed with 10% PEG concentration) showed the highest (10.08) water retention capacity. The lowest water retention capacity was found in P_0 (7.75) which is statistically identical with P_5 (8.21). Maximum water retention capacity (17.50) was scored with 10% PEG solution while the minimum value (10.62) scored with 0% PEG

solutions (Baque *et al*., 2002). Generally higher doses of potassium resulted the maximum relative water content, higher water retention capacity and exudation rate in drought affected wheat. Water saturation deficit highly reduced with higher level of K. Potassium fertilizer also made the leaf water potential more negative. Under drought stressed conditions the beneficial effect of potassium on drought stress was more noticeable than under control conditions (Baque *et al.,* 2002).

4.1.8.3 Interaction effect of variety and priming solutions

Interaction between variety and priming agents showed significant difference on water retention capacity seedling[1]. Maximum water retention capacity was obtained from treatment combination V_1P_3 (10.55%) which is statistically similar with treatment combination V_1P_2. Lowest data recorded from treatment combination V_2P_0 (7.64%) which is statistically similar with V_2P_5, V_1P_0. Nahar *et al*. 2017 also reported that priming helps to activate the metabolic enzymes responsible for germination of seed before germination takes place, so the hydro and osmo-primed seedlings can uptake more water than the non-primed ones and obtained the maximum turgid weight, in consequence, they obtained the maximum water retention capacity. Generally higher doses of potassium resulted the maximum relative water content, higher water retention capacity and exudation rate in drought affected wheat.

4.1.9. Coeffiecient of germination

4.1.9.1 Effect of variety

Mungbean variety exhibited significant difference in respect of coefficient of germination (Table 2). Among the varieties, BARI Mung 6 (V_1) showed the higher coefficient of germination (20.0%) than BARI Mung 5 (V_2) which showed lower coefficient of germination (17.68%).

Table 2. Effect of variety and different priming concentrations on the growth and water relation behaviors of Mungbean

Treatments	Relative water content (%)	Water saturation deficit%	Water retention capacity	Coefficient of germination%	Vigor index
Effect of variety					
V_1	76.25 a	23.72 b	9.30 a	20.00 a	160.63 a
V_2	68.71 b	31.29 a	8.75 b	17.68 b	133.29 b
LSD $_{(0.01)}$	2.71	1.24	0.35	0.67	5.08
CV (%)	5.39	6.49	5.57	5.11	4.99
Effect of different priming solutions					
P_0	52.90 f	47.10 a	7.75 d	16.97 c	97.20 f
P_1	70.11 d	29.79 c	8.92 c	17.60 c	132.1 d
P_2	82.86 b	17.14 e	9.76 ab	19.40 b	168.7 b
P_3	93.14 a	6.865 f	10.08 a	21.76 a	208.4 a
P_4	77.82 c	22.18 d	9.44 bc	19.28 b	158.9 c
P_5	58.06 e	41.94 b	8.21 d	18.00 c	116.4 e
LSD $_{(0.01)}$	4.69	2.14	0.60	1.16	8.79
CV (%)	5.39	6.49	5.57	5.11	4.99

V_1:BARI Muung 6 V_2:BARI Mung 5

P_0: Control; P_1:0% PEG; P_2: 5% PEG ;P_3:10% PEG; P_4: 15%PEG; P_5: 20% PEG

4.1. 9.2 Effect of priming solutions

Priming with different concentrations of PEG and water showed significant variation in respect of coefficient of germination (Table 2). Coefficient of germination is increased up to 10% of PEG then gradually decreased. P_3 (primed with 10% PEG concentration) showed the highest (21.76%) coefficient of germination. The lowest coefficient of germination was found in P_0 (16.97%) which is statistically similar with P_1 and P_5. Sung (1993), reported that priming increases the antioxidant increment like glutathione and ascorbate which help to increase germination speed by reducing the lipid peroxidation

activity, thus it leads to higher germination coefficient in osmo primed and hydro primed than non-primed.

4.1.9.3 Interaction effect of variety and priming solutions

Different Polyethylene Glycol (PEG) solutions significantly influenced the germination coefficient of Mungbean genotypes (Table 3).The highest coefficient of germination value was recorded from treatment combination V_1P_3 (22.87%) and lowest was found in treatment combination V_2P_0 (15.86%) which is statistically similar with V_2P_1 and V_2P_5. Coefficient of germination in 10% solution incase of wheat cultivar (Nahar, 2014).

4.1.10 Vigor index

4.1.10.1 Effect of variety

Mungbean variety exhibited significant difference in respect of vigor index (Table 2). Among the varieties, BARI Mungbean 6 (V_1) showed the higher vigor index (160.63) than BARI Mungbean 5 (V_2) which showed lower vigor index (133.29).

4.1.10.2 Effect of priming solutions

Priming with different concentrations of PEG and water showed significant variation in respect of vigor index (Table 2). This parameter increases up **to** 10% then degrease due to higher concentrations. P_3 (primed with 10% PEG concentration) showed the highest (208.4) vigor index. The lowest vigor index was found in P_0 (97.20). According to Sadeghi *et al*. (2011) Germination and vigor advancement of Mungbean plant was occurred due to the reserve mobilization of food material, activation and re-synthesis of some enzymes, DNA and RNA synthesis started during osmotic priming.

4.1.10.3 Interaction effect of variety and priming solutions

Combined of Mungbean genotype and priming solutions showed significant variation incase of vigor index (Table 3 and Appendix VI). Maximum VI was

recorded from treatment combination V_1P_3 (234.2) and minimum was found in treatment combination V_2P_0 (90.40) .

Table 3. Interaction effect of variety and different priming concentrations on the growth and water relation behaviors of Mungbean

Treatment	Relative water content (%)	Water saturation deficit%	Water retention capacity	Coefficient of germination%	Vigor index
V_1P_0	58.56 gh	41.44 c	7.85 gh	18.09 de	104.0 f
V_1P_1	74.03 e	25.77 e	9.10 c-e	18.96 cd	146.1 c
V_1P_2	85.46 bc	14.54 g	10.15 ab	20.54 bc	181.9 b
V_1P_3	94.88 a	5.120 i	10.55 a	22.87 a	234.2 a
V_1P_4	81.07 cd	18.93 f	9.63 bc	20.26 bc	174.3 b
V_1P_5	63.50 fg	36.50 d	8.50 e-g	19.26 b-d	123.4 d
V_2P_0	47.24 i	52.76 a	7.64 h	15.86 f	90.40 g
V_2P_1	66.19 f	33.81 d	8.74 d-f	16.25 f	118.1 de
V_2P_2	80.26 c-e	19.74 f	9.37 b-d	18.26 de	155.5 c
V_2P_3	91.39 ab	8.610 h	9.60 bc	20.66 b	182.7 b
V_2P_4	74.57 de	25.43 e	9.25 c-e	18.30 de	143.6 c
V_2P_5	52.62 hi	47.38 b	7.92 f-h	16.75 ef	109.5 ef
$LSD_{(0.01)}$	6.63	3.03	0.85	1.63	12.43
CV (%)	5.39	6.49	5.57	5.11	4.99

V_1P_0: Nonprimed seeds of BARI Mung 6 , V_1P_1: BARI Mung 6 seeds primed with Distilled water , V_1P_2: BARI Mung 6 seeds primed with 5% PEG, V_1P_3: BARI Mung 6 seeds primed with 10 % PEG , V_1P_4: BARI Mung 6 seeds primed with 15 % PEG , V_1P_5: BARI Mung 6 seeds primed with 20 % PEG , V_2P_0: Nonprimed seeds of BARI Mung 5 , V_2P_1: BARI Mung 5 seeds primed with Distilled water, V_1P_2:BARI Mung 5 seeds primed with 5% PEG, V_2P_3: BARI Mung 5 seeds primed with 10 % PEG ,V_2P_4: BARI Mung 5 seeds primed with 15 % PEG, V_2P_5: BARI Mung 5seeds primed with 20 % PEG

4.2 Experiment 2: Optimization of pre-sowing priming time on the germination behavior of Mungbean

Results obtained from the present study regarding the effects of different priming time of PEG and water on the germination and water relation behavior of Mungbean (BARI Mung 6) have been presented and discussed .

4.2.1 Germination percentage (%)

4.2.1.1 Effect of priming agents

Osmo (PEG) and hydro priming showed significant difference on germination percentage of BARI Mung 6 (Table 4). Osmo priming done with 10% PEG (P_1) showed higher germination percentage (79.53%) than hydro priming (priming with distilled water). Hydro priming (P_2) was showed germination percentage (73.30%) which is lower than osmo priming (PEG) .Khan *et al.* (2005) reported that Seed priming had increased germination of several plants .Priming has been shown to be effective in improving stand establishment and crop vigor in a wide range of crops (Harris *et al*., 2001). Osmo conditioned seeds may have improved germination and uniformity, especially under adverse seed bed condition such as low temperature (Stoffella *et al.*, 1992), Khalil *et al*. (2001) reported improvement in germination of soybean.

4.2.1.2 Effect of priming times

Result exposed that the germination percentage was gradually increased with increasing the priming time up to 9 h and then decreased with increasing priming time .Different priming times (3 hr,6 hr,9 hr,12 hr ,15hr and 18 hr) showed significant variance on germination percentage of BARI Mung 6 (Table 4). T_3 (9hr) was found highest germination percentage (91.32%). Lowest germination percentage was found in T_6 (52.83%). This result is agreement with Khan *et al*. (2008) .They reported that 9hr priming time with different concentration like -1.1 MPa and -0.5 MPa showed better germination percentage in case of soybean and chickpea respectively. Abnavi and Ghobadi

(2012); Lemrasky and Hosseini (2012); Giri and Schillinger (2003) also observed that wheat seed priming with water for 12 hr achieved better than non-primed seeds.

4.2.1.3 Interaction effect of priming agents and times

Interaction of osmo (PEG) and hydro priming at different priming times revealed significant variation in respect of germination percentage (Table.5, Appendix III). Among the treatment combination the maximum germination percentage (95.31%) was obtained from treatment combination (P_1T_3), seeds primed with PEG for 9 hr which was statistically similar with P_2T_3. The lowest germination rate (47.94%) was obtained from treatment combination (P_2T_6), seeds primed with distilled water for 18 hr. Similar findings also observed by Nahar *et al*. (2017) in case of wheat. Hamidreza *et al.* (2013); Yari *et al.* (2010) concluded that, hydro priming duration from 6 to 12 h expressed better result in germination percentage of wheat over hydro priming duration from 18 to 24 hr.

4.2.2 Shoot length (mm)

4.2.2.1 Effect of priming agents

Hydro and Osmo priming showed significant difference on shoot length of BARI Mung 6 (Table 4). Osmo priming done with 10% PEG (P_1) showed higher shoot length (142.63 mm) than hydro priming (priming with distilled water). Hydro priming (P_2) was showed shoot length (126.83mm). Baque *et al*., (2016) reported that the highest shoot length of wheat was secured when the seed primed with osmo priming (10% PEG solution). Lee and Kim (2000) revealed that, priming increased the metabolic activities of seed ultimately gained the substantial shoot length.

4.2.2.2 Effect of priming times

Different priming times (3hr,6hr,9hr,12 hr, 15hr and 18 hr) showed significant variance on shoot length of BARI Mung 6 (Table 4). T_3 (9 hr) was found highest shoot length (169.1 mm) which was statistically identical with T_4.

Lowest shoot length was found in T_6 (100.1 mm). Nahar *et al*., (2017) reported that priming time may beef up the enzymatic activities which results in vigorous plant growth ultimately longer shoot length otherwise over time may leads to aging of seeds for which seeds lose their potentiality to germinate and growth. Yari *et al*. (2010) revealed that maximum shoot length of sunflower is achieved when it was osmo primed for 12 hr.

4.2.2.3 Interaction effect of priming agents and times

Interaction of osmo (PEG) and hydro priming at different priming times revealed significant variation in respect of shoot length (Table.5, Appendix III).In case of shoot length, highest shoot length was found in P_1T_3 treatment combination (177.7 mm) which is statistically similar with treatment combination P_1T_4 and P_2T_3, lowest shoot length was recorded from treatment combination P_2T_6 (92.80mm) which is statistiacally similar with treatment combination P_1T_6. Nahar *et al.* (2017) reported that priming with PEG for 9 hr give hiher shoot length incase of wheat than nonprime seeds.According to them priming time might help to augmented enzymatic activities of seed which trigger the vigorous plant growth and in significantly increased the shoot length of wheat; on the other hand, higher priming time could facilitate the ageing of seed that can be resulted lowering the potentiality for better germination, growth and development of seedling. Kumar *et al*. (2002) reported that 8 hours osmo priming of finger millet seeds in water resulted in an increased mean plant height by 9 cm.

Table 4. Effect of osmo and hydro priming and different priming times on the germination and growth behavior of mungbean

	Germination percentage (%)	Shoot length (mm)	Root length (mm)	Shoot dry weight (mg)	Root dry weight (mg)
Effect of osmo and hydro priming					
P_1	79.53 a	142.63 a	56.26 a	38.39 a	14.76 a
P_2	73.30 b	126.83 b	46.37 b	31.63 b	13.30 b
LSD $_{(0.01)}$	3.45	6.42	2.64	1.96	0.71
CV (%)	4.85	5.11	5.53	6.00	5.45
Effect of different priming times					
T_1	77.31 bc	118.8 d	41.19 d	27.79 d	11.87 d
T_2	81.70 b	130.8 c	55.49 b	34.47 c	14.09 c
T_3	91.32 a	169.1 a	66.94 a	46.76 a	18.21 a
T_4	82.56 b	153.4 b	59.64 b	41.77 b	16.08 b
T_5	72.75 c	136.1 c	50.35 c	34.99 c	13.49 c
T_6	52.83 d	100.1 e	34.27 e	24.27 e	10.46 e
LSD $_{(0.01)}$	5.98	11.12	4.58	3.39	1.24
CV (%)	4.85	5.11	5.53	6.00	5.45

P_1: Osmo priming P_2: Hydro priming

T_1: 3 hr; T_2: 6 hr; T_3: 9 hr ; T_4: 12 hr ; T_5: 15 hr; T_6: 18 hr

4.2.3 Root length (mm)

4.2.3.1 Effect of priming agents

Hydro and Osmo priming showed significant difference on root length of BARI Mung 6 (Table 4).Osmo priming done with 10% PEG (P_1) showed higher root length (56.26 mm) than hydro priming (priming with distilled water). Hydro priming (P_2) was showed root length (46.37 mm). Carceller and Soriano (1972) reported that root length of osmo and hydro primed seed

exerted the highest length than non-primed seed. Increased root length by osmo priming with PEG was also earlier reported in wheat.

4.2.3.2 Effect of priming times

Result exposed that the root length was gradually increased with increasing the priming time up to 9 h and then decreased with increasing priming time (Table 4). T_3 (9 hr) was observed highest root length (66.94 mm). Lowest root length was found in T_6 (34.27 mm). Ajirloo *et al. (* 2013) revealed that increasing time had greater effect on root length in case of wheat Result exposed that the germination percentage was gradually increased with increasing the priming time up to 9 h and then decreased with increasing priming time. Nahar *et al*., (2017) reported that maximum root length was obtained when wheat seed primed with 10% PEG for 9 hr. Murray (1989), who concluded that over priming may cause oxygen deficiency and the build-up of inhibitors. The findings of this study suggested that priming duration of 12 h was generally safer for pea.

4.2.3.3 Interaction effect of priming agents and times

Interaction of osmo (PEG) and hydro priming at different priming times revealed significant variation in respect of root length (Table.5, Appendix III).For root length, highest root length was recorded from treatment combination P_1T_3 (71.88 mm) and lowest root length was showed in treatment combination P_2T_6 (28.19 mm). According to Varshini *et al*., (2018) seed priming with -0.5 MPa PEG for 9 hours recorded highest root length (17.05 cm and 16.19 cm in between paper and sand methods, respectively) and 0 MPa for 24 hours recorded lowest root length. Arif *et al*. (2008) conducted a field experiment in Pakistan and they reported that priming (osmo) improved the seed establishment in soybean which might be due to the completion of pre-germination metabolic activities earlier which makes the seed ready for radical protrusion thus increase root length.

4.2.4 Shoot dry weight (mg)

4.2.4.1 Effect of priming agents

Osmo and hydro priming showed significant difference on shoot dry weight of Mungbean genotypes (Table 4). Osmo priming done with 10% PEG (P_1) showed higher shoot dry weight (38.39 mg) than hydro priming (priming with water). Hydro priming (P_2) showed shoot dry weight (31.63 mg). Nahar *et al.* (2014) concluded that osmo priming with 10% PEG had highest shoot dry weight than hydro priming. Sarika *et al.* (2013) conducted a lab that chemo priming with GA_3 and Ethrel improved the seed quality and showed improved seedling length, seedling dry weight.

4.2.4.2 Effect of priming times

Different priming times (3hr,6hr,9hr,12 hr, 15hr and 18 hr) showed significant variation on shoot dry weight of BARI Mung 6 (Table 4). T_3 (9 hr) was observed highest shoot dry weight (46.76mg). Lowest shoot dry weight was found in T_6 (27.27 mg) which is statistically identical with T_1. When seeds are Osmo primed for 12 hrs, it is significantly affected shoot dry weight and had highest shoot dry weight in wheat (Hamidreza *et al*., 2014). But Moghanibashi *et al*. (2012) showed that, when sunflower seed is hydro primed for 24 hr shoot dry weight was increased than non-primed seed.

4.2.4.3 Interaction effect of priming agents and times

Interaction of osmo (PEG) and hydro priming at different priming times revealed significant variation in respect of shoot dry weight (Table.5, Appendix III). Highest shoot dry was recorded from treatment combination P_1T_3 (49.26 mg) . Lowest shoot dry weight was showed in treatment combination P_2T_6 (20.67 mg) which is statistically similar with, P_1T_6 and P_2T_1. Hamidreza *et al.* (2013), reported that, shoot dry weight significantly affected by osmo priming times as 6 hours seed treatment had highest shoot dry weight of wheat. But Moghanibashi *et al*. (2012) revealed that, the effect of hydro priming for 24 h

increased shoot dry weight of sunflower as compared to non-primed seed. Lemrasky and Hosseini (2012) also concluded that, wheat seed priming with PEG 10% for 45 h increased shoot dry weight.

4.2.5 Root dry weight (mg)

4.2.5.1 Effect of priming agents

Osmo and hydro priming showed significant difference on root dry weight of BARI Mung 6 (Table 4). Osmo priming done with 10% PEG (P_1) showed higher root dry weight (14.76 mg) than hydro priming (priming with distilled water). Hydro priming (P_2) was showed root dry weight (13.30 mg). Baque *et al*. (2016) found that maximum root dry weight was recorded when the seed osmo primed with 10% PEG solution than hydro primed seed.

4.2.5.2 Effect of priming times

Soot dry weight of Mungbean genotypes significantly varied by different priming times. An increasing trend of root dry weight was observed with increasing priming time up to 9 hr (Table 4). T_3 (9 hr) was observed highest root dry weight (18.21 mg). Lowest root dry weight was found in T_6 (10.46 mg) .Hamidreza *et al*.(2013) revealed that osmo priming for 9 hr had highest root dry weight in case of wheat.

4.2.5.3 Interaction effect of priming agents and times

Combined effect of priming agents and time showed significant variation in case of root dry weight (Table.5 and Appendix III).Highest root dry weight was recorded from treatment combination P_1T_3 (18.59 mg) and lowest data was recorded from treatment combination P_2T_6 (9.39 mg) . Mubshar *et al*. (2006) concludrd that maximum root dry weight was found in BARI Gom 27 when seed primed with PEG for 12 hr and decreases for further increases of priming time. Nahar *et al*. (2017) showed that seed primed with PEG for 9 hr gives maximum root dry weight and over priming may cause oxygen deficiency and the build-up of inhibitors as a result decrease in root dry weight.

Table 5. Interaction effect of osmo and hydro priming and different priming times on the germination and growth behavior of mungbean

Treatment combinations	Germination percentage (%)	Shoot length (mm)	Root length (mm)	Shoot dry weight (mg)	Root dry weight (mg)
P_1T_1	81.04 b-d	121.8 e-g	45.89 fg	32.07 e	12.42 f
P_1T_2	85.44 bc	137.1 de	59.13 b-d	37.19 d	15.20 cd
P_1T_3	95.31 a	177.7 a	71.88 a	49.96 a	18.59 a
P_1T_4	84.04 bc	164.6 ab	64.55 b	44.41 b	16.38 bc
P_1T_5	73.62 de	147.1 cd	55.74 c-e	38.81 cd	14.45 de
P_1T_6	57.72 f	107.5 gh	40.35 gh	27.88 ef	11.54 f
P_2T_1	73.59 de	115.8 fg	36.49 h	23.52 fg	11.32 f
P_2T_2	77.96 c-e	124.6 ef	51.84 ef	31.75 e	12.97 ef
P_2T_3	87.33 ab	160.4 bc	62.00 bc	43.57 bc	17.83 ab
P_2T_4	81.07 b-d	142.3 d	54.72 de	39.13 cd	15.78 cd
P_2T_5	71.89 e	125.0 ef	44.97 g	31.16 e	12.52 f
P_2T_6	47.94 g	92.80 h	28.19 i	20.67 g	9.39 g
LSD $_{(0.01)}$	8.46	15.73	6.48	4.80	1.75
CV (%)	4.85	5.11	5.53	6.00	5.45

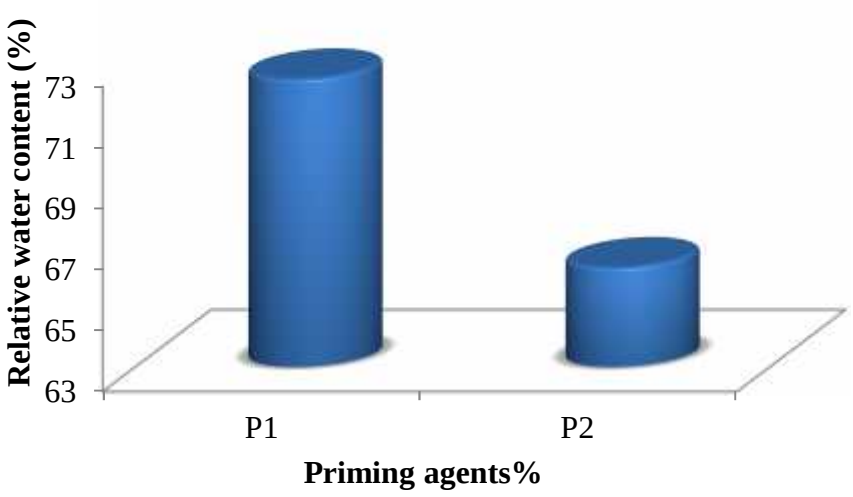
P_1T_1= PEG primed at 3 hr, P_1T_2= PEG primed at 6 hr, P_1T_3= PEG primed at 9 hr, P_1T_4= PEG primed at 12 hr, P_1T_5= PEG primed at 15 hr , P_1T_6= PEG primed at 18 hr ,P_2T_1= Hydro primed at 3 hr, P_2T_2= Hydro primed at 6 hr , P_2T_3= Hydro primed at 9 hr, P_2T_4= Hydro primed at 12 hr, P_2T_5= Hydro primed at 15 hr, P_2T_6= Hydro primed at 18 hr

4.2.6 Relative water content%

4.2.6.1 Effect of priming agents

Relative water content is influenced by seed quality and seed priming technique.Different priming agents showed significant variation in relative water content of BARI Mung 6 when it was analyzed (Fig. 11). It was observed that Osmo priming (P_1) showed higher performance (72.45%) than

hydro priming P_2 (66.26%). Significantly higher relative water content was recorded in leaves obtained from plots sown with higher quality seeds as compared to those obtained from plots sown with lower quality seeds. The leaves obtained from plots having seed primed with $CaCl_2.2H_2O$ (0.5%) showed significantly highest relative water content which was on par with the leaves from plots having seed primed with KH_2PO_4 (50 ppm) followed by leaves obtained from plots having seed primed with GA3 (20ppm) (84.57%) (Assefa, 2008).



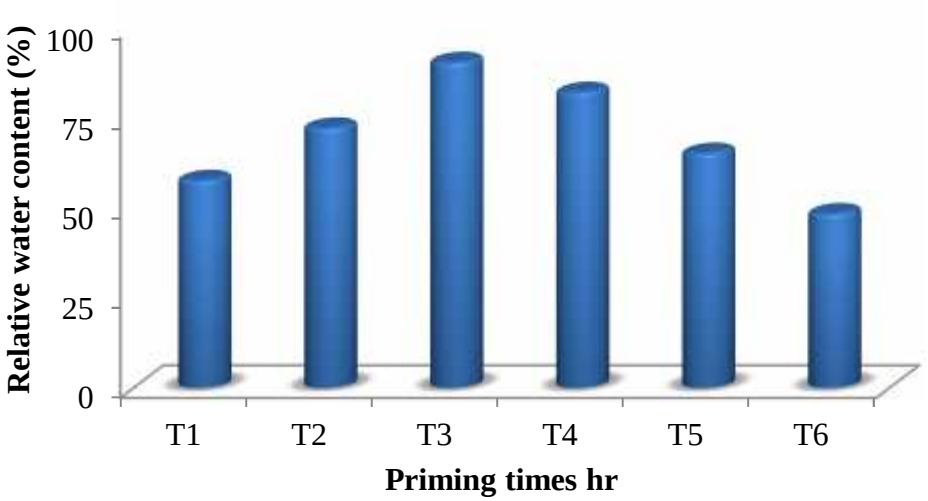
P_1: Osmo priming P_2: Hydro priming

Figure 11. Effect of osmo and hydro priming on the relative water content of mungbean (LSD $_{0.01}$=3.53)

4.2.6.2 Effect of priming times

Different priming times (3 hr,6 hr,9 hr,12 hr,15 hr and 18 hr) showed significant variance on relative water content of BARI Mung 6 (Fig. 12). T_3 (9 hr) showed highest relative water content 90.45%. T_6 (18 hr) showed lowest relative water content 48.44%. According to Nahar *et al.* (2017) Priming time helps to accelerated enzymatic activities of seed to facilitate the growth of healthy and vigorous seedling, and so this seedling may have higher relative water content. Over priming time beef up the ageing process of primed seed,

produced weak and lean seedling and the ultimate result is lower relative water content.



T_1= Priming for 3 hr; T_2= Priming for 6 hr; T_3= Priming for 9 hr; T_4= Priming for 12 hr; T_5= Priming for 15 hr; T_6= Priming for 18 h

Figure 12. Effect of different priming times on the relative water content of mungbean (LSD $_{0.01}$=6.12)

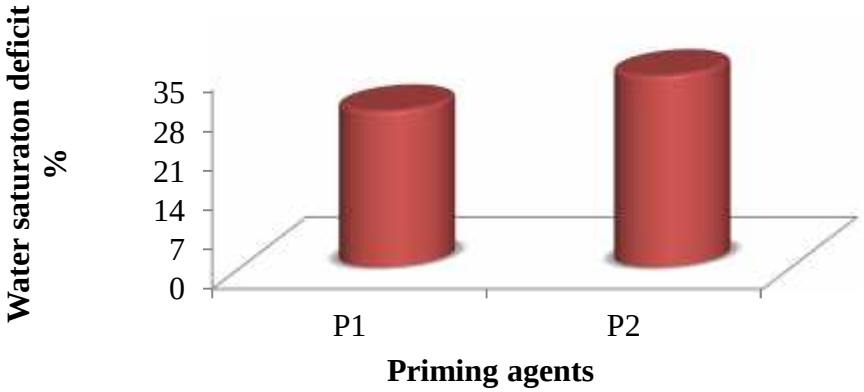
4.2.6.3 Interaction effect of priming agents and times

Combined effect of priming agents and time showed significant variation in case of relative water content (Table.6 and Appendix IV).According to analyzed data in the case of relative water content , highest relative water content was showed in treatment combination P_2T_3 (92.96%) which is statistically similar with treatment combination P_1T_4 and P_2T_3. Lowest relative water content treatment combination from P_2T_6 (45.56%) which is statistically similar with treatment combination P_1T_6, P_2T_5 and P_2T_1. Priming time helps to enhanced enzymatic activities of seed which facilitated the growth of healthy and vigorous seedling, which might have the capacity to provide higher relative water content. Over priming time trigger the ageing process of primed seed, produced weak and lean seedling ultimately exhibited lower relative water content .

4.2.7 Water saturation deficit %

4.2.7.1 Effect of priming agents

It followed the opposite trend compared to the previously described parameter. Different priming agents showed significant variation in water saturation deficit of BARI Mung 6 when it was analyzed (Fig. 13). It was observed that hydro priming (P_1) showed higher performance (33.75%) than osmo priming P_2 (27.55%). This result also in agreement with the findings of previous researchers in field crops like mungbean (Asaduzzaman, 2014; Rahman, 2014).

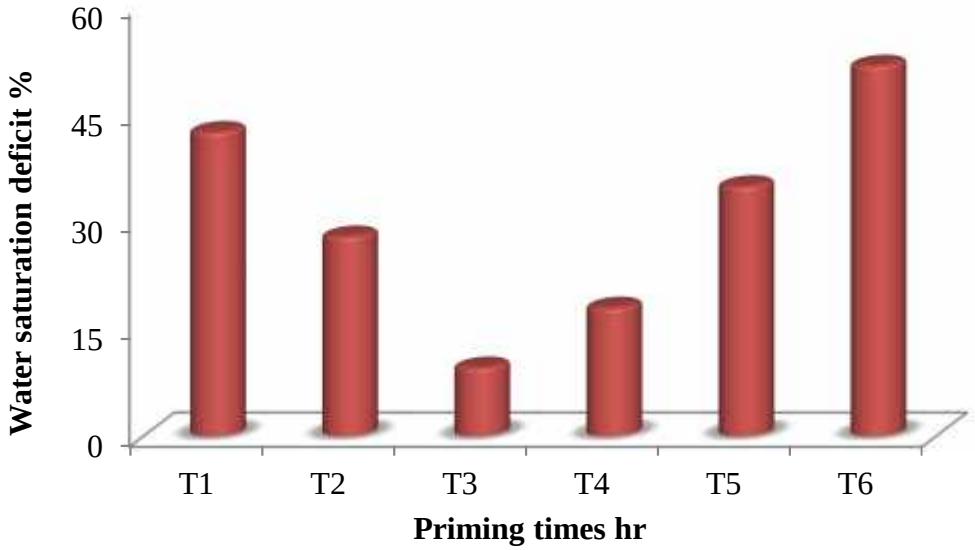


P_1: Osmo priming P_2: Hydro priming

Figure 13. Effect of osmo and hydro priming on the water saturation deficit of mungbean (LSD $_{0.01}$=1.61)

4.2.7.2 Effect of priming times

Different priming times (3 hr,6hr, 9hr,12 hr,15hr and 18hr) showed significant variance on water saturation deficit of BARI Mung 6 (Fig. 14). Water saturation deficit is reverse than other parameter.T_6 (18 hr) showed highest water saturation deficit 51.56. T_3 (9 hr) showed lowest water saturation deficit 9.552. According to Nahar *et al*. (2016) when priming time is low, enzymatic activities are also low, as a result weak and lean seed are produced. Again when over priming time result is same and they are unable to uptake water needed water and so water saturation deficit value is increased.



T_1= Priming for 3 hr; T_2= Priming for 6 hr; T_3= Priming for 9 hr; T_4= Priming for 12 hr; T_5= Priming for 15 hr; T_6= Priming for 18 hr

Figure 14. Effect of different priming times on the water saturation deficit of mungbean (LSD 0.01=2.79)

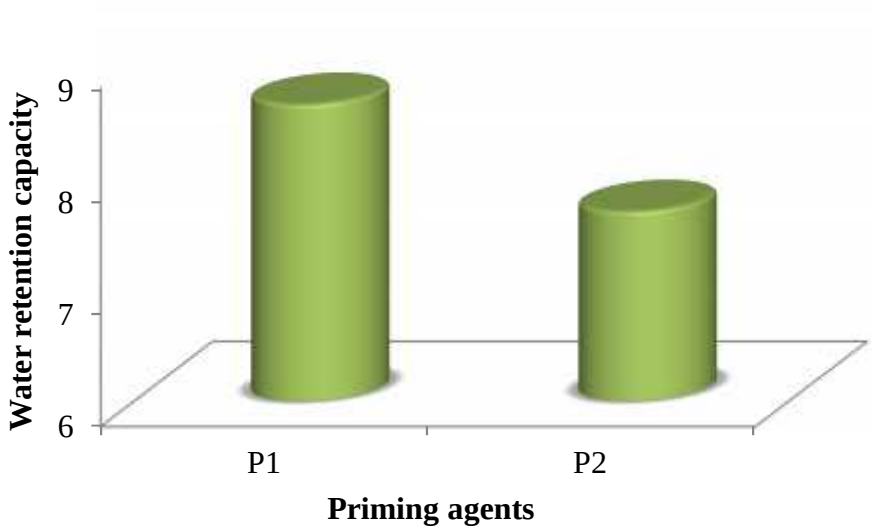
4.2.7.3 Interaction effect of priming agents and times

Combined effect of priming agents and time showed significant variation in case of water saturation deficit (Table.6 and Appendix IV). Maximum water saturation deficit found in treatment combination P_2T_6 (54.44) which was statistically different from others and lowest was found in treatment combination P_1T_3 (7.04). They both are statistically different from others. Nahar *et al*.(2017), revealed that, maximum water saturation deficit (70.00) was scored by wheat genotype when the seeds were primed with 10% PEG solution for 3 h and the minimum water saturation deficit (8.55) was scored when the seeds were primed with 10% PEG solution for 9 h.

4.2.8 Water retention capacity %

4.2.8.1 Effect of priming agents

Different priming agents showed significant variation in water retention capacity of BARI Mung 6 when it was analyzed (Fig. 15). It was observed that Osmo priming (P_1) showed higher performance (8.63%) than hydro priming P_2 (7.68%). Judicious doses of PEG (Polyethylene Glycol) showed better tolerance at salt stress condition than hydro-priming, while more doses of PEG had negative effects on water retention capacity (Sun *et al*., 2010).



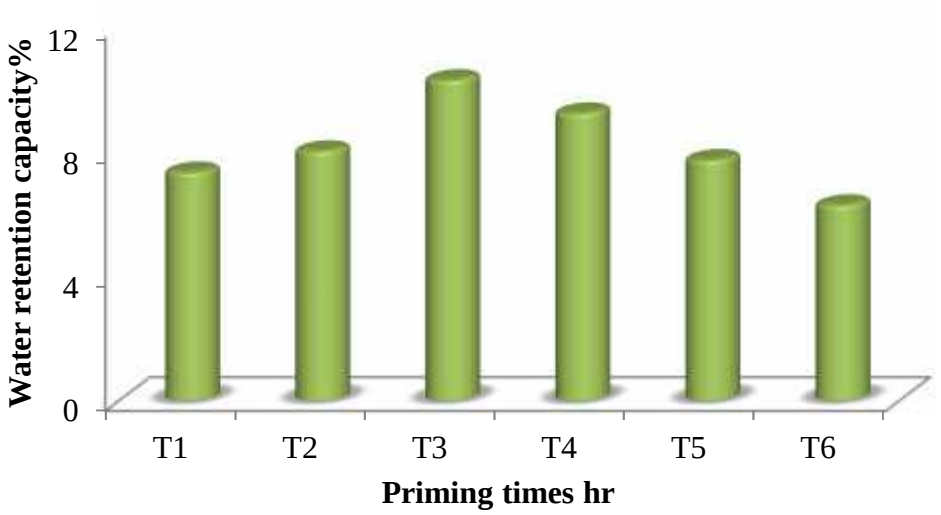
P_1: Osmo priming P_2: Hydro priming

Figure 15. Effect of osmo and hydro priming on the water retention capacity of mungbean (LSD $_{0.01}$=0.47)

4.2.8.2 Effect of priming times

Different priming times showed significant variation on water retention capacity on Mungbean genotype (Fig. 16). Water retention capacity increased with increasing priming time up to 9 h and then gradually decreased with increasing priming time. T_3 (9 hr) showed highest water retention capacity 10.31. T_6 (18 hr) showed lowest water retention capacity 6.287. Lower and

over priming time both have deleterious effect on growth and development of seedling. Vigorous seedling have higher water retention capacity than weak and lean seedlings (Nahar *et al*., 2014).



T_1= Priming for 3 hr; T_2= Priming for 6 hr; T_3= Priming for 9 hr; T_4= Priming for 12 hr; T_5= Priming for 15 hr; T_6= Priming for 18 hr

Figure16. Effect of different priming times on the water retention capacity of mungbean (LSD $_{0.01}$=0.82)

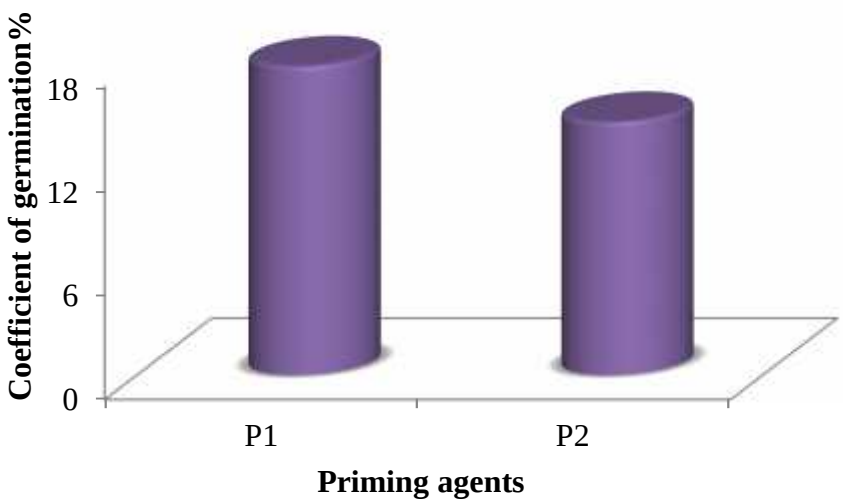
4.2.8.3 Interaction effect of priming agents and times

Combined effect of priming agents and time showed significant variation in case of water retention capacity (Table 6 and Appendix IV). Highest water retention capacity, 10.83% was found in treatment combination P_1T_3 (seeds primed with PEG for 9 hr) which is statistically similar with treatment combination P_2T_3, P_1T_4 and P_2T_4. The lowest, 5.71% was recorded from treatment combination P_2T_6 (seeds primed with distilled water for 18 hr) which is statistically similar with treatment combination P_2T_1 and P_1T_6.

4.2.9 Coefficient of germination%

4.2.9.1 Effect of priming agents

Different priming agents showed significant variation in coefficient of germination of Mungbean genotype (Fig. 17). It was observed that osmo priming (P_1) showed higher performance (17.76%) than hydro priming (P_2) (14.56%). Osmo priming of Italian ryegrass (*Lolium multiflorum*) and sorghum seeds with 20% PEG_{8000} for 2 d at 10°C enhanced germination coefficient of wheat genotypes under water stress, water logging, cold stress and saline conditions (Hur., 1991)



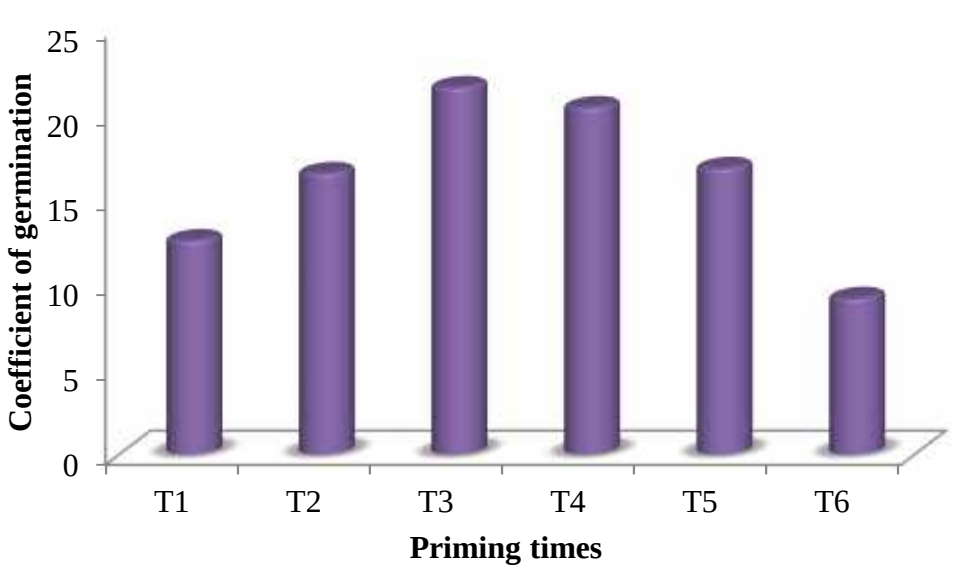
P_1: Osmo priming P_2: Hydro priming

Figure 17. Effect of osmo and hydro priming on the coefficient of germination of mungbean (LSD $_{0.01}$=1.08)

4.2.9.2 Effect of priming times

Different priming times (3 hr,6 hr,9 hr,12 hr,15 hr and 18hr) showed significant variance on coefficient of germination capacity of BARI Mung 6 (Fig. 18). T_3 (9 hr) showed highest coefficient of germination 21.54 % which is statistically similar with T_4. T_6 (18 hr) showed lowest coefficient of germination 9.173%.

Sadeghi *et al*., (2011) reported that, the highest germination coefficient was attained from -1.2 osmotic potential and 9 hr seed priming duration treatments (21.15 and 20.15, respectively). Hydro priming for 24 h enhanced germination coefficient of sunflower seed as compared with the control (Moghanbashi *et al*., 2012).



T_1= Priming for 3 hr; T_2= Priming for 6 hr; T_3= Priming for 9 hr; T_4= Priming for 12 hr; T_5=Priming for 15 hr; T_6= Priming for 18 hr

Figure 18. Effect of different priming times on the coefficient of germination of mungbean (LSD $_{0.01}$=1.87)

4.2.9.3 Interaction effect of priming agents and times

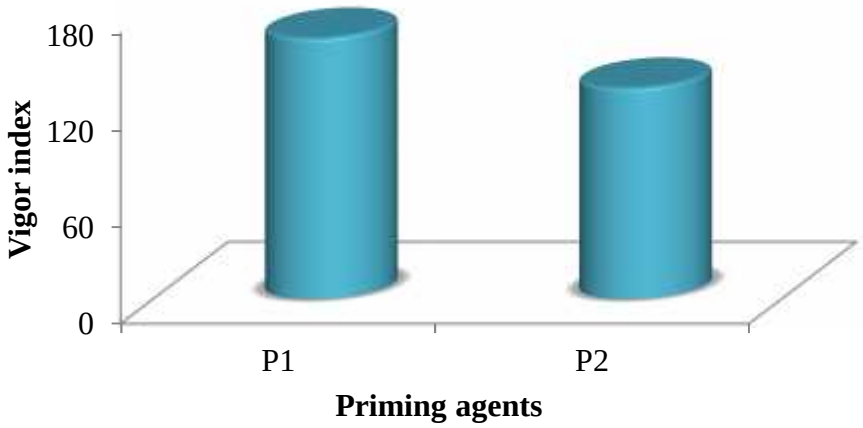
Combined effect of priming agents and time showed significant variation in case of coefficient of germination (Table 6 and Appendix IV). The maximum coefficient of germination value, 22.35% was recorded from treatment combination P_1T_3 (seeds primed with PEG for 9 hr) which is statistically similar with treatment combination P_1T_4 and P_2T_3. Lowest value 7.01% was found in treatment combination P_2T_6 (seeds primed with distilled water for 18 hr). The highest germination coefficient (17.93%) was observed in wheat genotype ESWYT-5 when the seeds primed with 10% PEG solution for 9 hr

(Nahar *et al*., 2017). Moghanibashi *et al*. (2012), reported that the impact of hydro priming for 24 h enhanced germination coefficient of sunflower seed as compared with the control. Sadeghi *et al*. (2011), also reported that, the highest germination coefficient was attained from -1.2 osmotic potential and 12 h seed priming duration treatments (21.15 and 20.15, respectively). Germination coefficient reduced by the reduction of osmotic potential and increment of seed hydro priming time. Seed priming resulted anti-oxidant increment as glutathione and ascorbate in seed. These enzymes make more germination speed via reduction of lipid proxidation activity.

4.2.10 Vigor index

4.2.10.1 Effect of priming agents

Different priming agents showed significant variation in coefficient of germination of BARI Mung 6 when it was analyzed (Fig. 19). It was observed that Osmo priming (P_1) showed higher performance (161.51) than hydro priming (130.59). It has been reported that primed seeds showed better germination pattern and highe r vigor level than non-primed (Ruan *et al*.,2002).

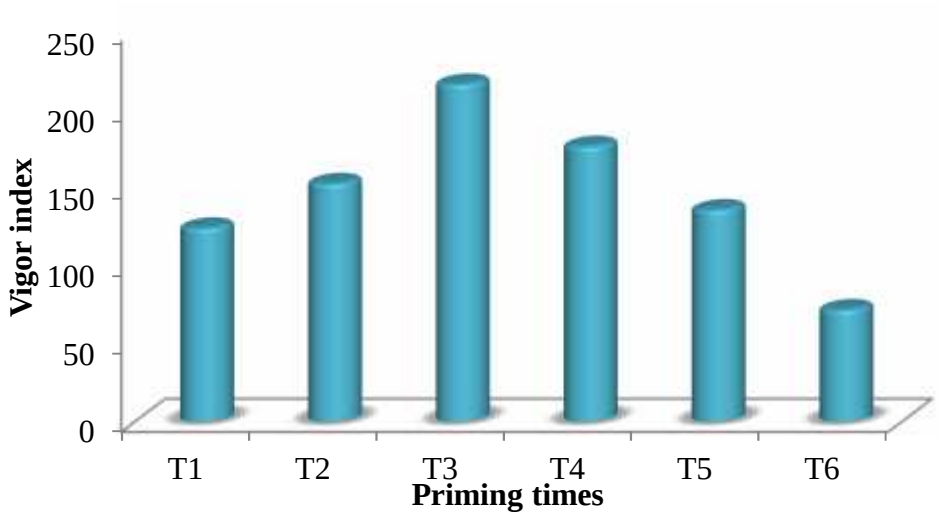


P_1: Osmo priming P_2: Hydro priming

Figure 19. Effect of osmo and hydro priming on the vigor index of mungbean ($LSD_{0.01}$=8.13)

4.2.10.2 Effect of priming times

Different priming times (3 hr,6 hr,9 hr,12 hr,15 hr and 18 hr) showed significant variance on vigour index of Mungbean genotype (Fig. 20). T_3 (9 hr) showed highest vigor index 216.1. T_6 (18 hr) showed lowest vigor index 71.70. The highest vigor index of BARI Gom 27 was recorded when the seed primed with 10% PEG solution for 12 h (Nahar *et al*., 2016 and Sadeghi *et al*., 2011).



T_1= Priming for 3 hr; T_2= Priming for 6 hr; T_3= Priming for 9 hr; T_4= Priming for 12 hr; T_5= Priming for 15 hr; T_6= Priming for 18 hr

Figure 20. Effect of different priming times on the vigor index of mungbean (LSD $_{0.01}$=14.08)

4.2.10.3 Interaction effect of priming agents and times

Interaction between priming agent (PEG and Distiiled water) and time showed statistical variation (Table 6 and Appendix IV) in case of vigor index of BARI Mung 6. Treatment combination P_1T_3 (seeds primed with PEG for 9 hr) showed highest vigor index (237.8) and lowest 57.98 was found in treatment combination P_2T_6 (seeds primed with distilled water for 18 hr) . Sadeghi *et al*., 2011), concluded that priming time significantly influenced vigor index of wheat. The highest vigor index of BARI Gom 27 was recorded when the seed

primed with 10% PEG solution for 12 h. Sadeghi *et al*. (2011), found that priming has the capability to repair some damages that have been produced from seed erosion and improve seed quality.

Table 6. Interaction effect of osmo and hydro priming and different priming times on the water relation behavior of mungbean

Treatment combinations	Relative water content (%)	Water saturation deficit%	Water retention capacity	Coefficient of germination%	Vigor index
P_1T_1	61.59 ef	38.41 c	7.83 d-g	14.40 f	136.0 de
P_1T_2	75.40 cd	24.60 e	8.51 c-e	19.28 b-d	167.7 c
P_1T_3	92.96 a	7.04 h	10.83 a	22.35 a	237.8 a
P_1T_4	84.39 ab	15.61 g	9.50 bc	21.79 ab	192.7 b
P_1T_5	69.05 de	30.95 d	8.23 d-f	17.39 de	149.4 cd
P_1T_6	51.33 g	48.67 b	6.86 gh	11.33 gh	85.42 g
P_2T_1	53.76 fg	46.24 b	6.85 gh	10.73 h	112.0 f
P_2T_2	69.05 de	30.98 d	7.53 e-g	13.73 fg	137.3 de
P_2T_3	87.94 ab	12.06 g	9.78 ab	20.73 a-c	194.4 b
P_2T_4	79.86 bc	20.14 f	8.98 b-d	18.94 cd	159.8 c
P_2T_5	61.38 ef	38.62 c	7.25 fg	16.21 ef	122.0 ef
P_2T_6	45.56 g	54.44 a	5.71 h	7.01 i	57.98 h
$LSD_{(0.01)}$	8.65	3.94	1.16	2.64	19.91
CV (%)	5.46	5.64	6.20	7.15	5.97

P_1T_1= PEG primed at 3 hr, P_1T_2= PEG primed at 6 hr, P_1T_3= PEG primed at 9 hr, P_1T_4= PEG primed at 12 hr, P_1T_5= PEG primed at 15 hr , P_1T_6= PEG primed at 18 hr ,P_2T_1= Hydro primed at 3 hr, P_2T_2= Hydro primed at 6 hr , P_2T_3= Hydro primed at 9 hr, P_2T_4= Hydro primed at 12 hr, P_2T_5= Hydro primed at 15 hr, P_2T_6= Hydro primed at 18 hr

4.3 Experiment 3: Effect of different level of drought stress on germination and water relation behavior of Mungbean

Pre-sowing seed treated with Polyethylene Glycol (PEG) believed to be a potential priming agent to increase the germination, seedling growth and water relation behavior of crop plants under salt stress condition. With this view, a lab investigation was carried out to find out the effect of PEG on the germination, seedling growth and water relation behavior of wheat under different salt levels.Results obtained from the present study regarding the germination behavior and water relation behavior of primed seed under salt (Sodium Chloride) stress condition.These are presented and discussed below:

4.3.1 Germination percentage (%)

4.3.1.1 Effect of priming agents

Germination percentage of PEG primed and hydro primed Mungbean seeds was influenced by different levels of salt stress (Table 7).Osmo priming done with 10% PEG and hydro priming done with Distilled water showed significant variation in respect of germination percentage. Results obtained from table .7, it can be concluded that osmo priming showed higher germination percentage P_1 (74.92%) than hydro priming P_2 (67.36%). Osmo priming with PEG results in improving the antioxidant enzymes and increasing the seed germination potential, finally resulting in an increased stress tolerance in germinating seeds of spinach (Chen and Arora, 2011). Good germination ensures better establishment of crop and it is a big sign for maximum crop yield. For that sometime seed treatment are necessary before sowing. Farooq *et al.* (2006) proposed that physiological changes exerted by osmo hardening such as metabolic activities proceed to repair and build up of nucleic acids, increase synthesis of proteins as well as repair membranes and enhance the activities of anti-oxidative enzymes increased the starch hydrolysis and made more sugars available for embryo growth as a result, the germination capability and tolerance under stress condition (especially salt stress) of seeds can be

promoted. Seed priming hasten germination percentage, lessened emergence time and enhanced yields are reviewed in many crops (Farooq *et al*., 2006b; Afzal *et al*., 2006; Afzal *et al*., 2011a). Judicious doses of PEG (Polyethylene Glycol) showed better tolerance at drought stress condition than hydro-priming, while more doses of PEG had negative effects on germination (Sun *et al*., 2010).

4.3.1.2 Effect of salinity levels

Different salinity levels revealed significant variation in respect of germination percentage (Table 7). Result exposed that the germination from PEG primed seeds decreased significantly with increasing salinity level. It was observed that the highest germination rate (87.39%) was under primed seeds (PEG) placed without salt (T_0) and after that the second highest germination rate (82.67%) was with Primed seeds placed with 5 dsm^{-1} (T_1) where the lowest germination rate (49.88%) was obtained from (T_4) with Primed seeds placed with 20 dsm^{-1} NaCl. Edalat-Pisheh *et al*. (2010) declared that total germination percentage in wheat seeds decreased when salinity of both primed and unprimed (control group) treatments increased. Kaya *et al*. (2006) and Khajeh-Hosseini *et al*. (2003) also find that reduction in total germination was significantly lower for non- primed seeds, compared to primed seeds and this may be due to the toxic effects of Na^+ and Cl^- in the process of germination.

4.3.1.3 Intaraction effect of priming agents and salinity levels

Interaction between priming (osmo and hydro priming) and different salinity levels showed significant variation in case of germination percentage (Table. 8 and Appendix V). Under different salinity levels osmo primed (PEG) seeds showed maximum tolerance capability and the magnitude of reduction was slow compare to hydro primed seeds those showed maximum susceptibility and the magnitude of reduction was rapid compare to that of osmo primed . The result of the experiment revealed that, the maximum germination percentage (90.34%) was recorded from treatment combination (P_1T_0) 10%

PEG primed seed at 0 dSm^{-1} salt concentrations which was statistically similar with P_1T_1, P_2T_0 and P_2T_1. The minimum germination percentage (43.58%) was recorded from treatment combination P_2T_4 (hydro primed seeds at 20 dSm^{-1} salt concentrations). Similar findings were reported by Kaya *et al.* (2006) and Khajeh-Hosseini*et al*. (2003). Farooq*et al.* (2006), proposed that physiological changes exerted by osmo hardening such as metabolic activities proceed to repair and build up of nucleic acids, increase synthesis of proteins as well as repair membranes, and enhance the activities of anti-oxidative enzymes increased the starch hydrolysis and made more sugars available for embryo growth as a result, the germination capability and tolerance under stress condition (especially salt stress) of seeds can be promoted.

4.3.2 Shoot length (mm)

4.3.2.1 Effect of priming agents

Shoot length of different priming agents was significantly influenced by different salinity levels (Table.7). Results revealed that the highest shoot length (137.62 mm) was observed from P_1 (10% PEG primed) where the lower shoot length (115.36 mm) was observed from P_2 (Distilled water) Primed seeds under saline condition. It was stated by Kaur *et al*. (2005) that osmo and hydro priming of chickpea seeds with mannitol and water lightened the negative effects of water deficiency and salt stress on seedling development. The treatment of seeds with water, 2% and 4 % mannitol improved the length and biomass of roots and shoots of chickpea seedlings as compared to non-primed under salt stressed conditions and osmo primed showed more shoot length over hydro primed seeds. Similarly, seed priming with P solutions significantly improved fresh and dry weight and plant height of mungbean seedlings 21 days after sowing in the field experiment (Shah *et al*., 2012).

4.3.2.2 Effect of salinity levels

Shoot length of Mungbean variety primed with 10% PEG was significantly influenced by different salinity levels (Table 7). Result exposed that the shoot

length from primed seeds decreased significantly with increasing salinity level .Results revealed that the highest shoot length (149.6mm) was observed from seeds placed without salt (T_0) which is statistically similar with (T_1) Primed seeds placed with 5 dsm^{-1} NaCl where the lowest shoot length (88.16 mm) was observed from Primed seeds placed with 20 dsm^{-1} NaCl (T_4). Salinity has both osmotic and specific ionic effects on seedlings growth (Dioniso-Sese and Tobita, 2000). Similarly, toxic ion accumulation (Na^+ and Cl^-) negatively affect plant metabolism (Grieve and Fujiyama 1987). It has also been reported that salinity suppresses the uptake of essential nutrients like P and K (Nasim *et al.* ,2008).Sun *et al*. (2010) also concluded that PEG priming with moderate concentration resulted in higher tolerance to drought stress and maximize shoot and root length.

4.3.2.3 Intaraction effect of priming agents and salinity levels

Interaction between priming (osmo and hydro priming) and different salinity levels showed significant variation in case of shoot length (Table 8 and Appendix V). Treatment combination P_1T_0 showed highest tolerance capacity where as treatment combination P_2T_4 showed highest susceptibility. The result of the experiment revealed that, the highest shoot length (157.7 mm) was secured from treatment combination P_1T_0 (10% PEG primed seed under 0 dSm^{-1} salt concentrations) which is statistically similar with P_1T_1 and P_2T_0 and the lowest shoot length (73.66 mm) was secured from hydro primed seeds under 20 dSm^{-1} salt concentrations (P_2T_4). From treatment combination P_1T_1 (PEG primed seeds placed with 5 dsm^{-1} NaCl solution) shoot length was found 152.9 mm. Mohammadi and Amiri (2010) reported that the shoot length decreased in higher extent for salinity sensitive cultivar compare to the tolerance wheat cultivar might be due to more accumulation of Na^+ which retard the cell division and elongation process and ultimately reduce the shoot length. Under salinity stress condition the extent of shoot length decrease was lower of wheat seed primed with 10% PEG solution. The possible cause of lower reduction may be the osmo priming agent PEG which alleviate the adverse effect of salt

stress, increased the metabolic activity, trigger the cell division and elongation process consequently reduce the adverse effect of salt on shoot growth. Lee and Kim (2000) also excluded that, priming increased the metabolic activities of seed consequently produced the substantial shoot length. Nahar *et al*. (2017) and Baque *et al.* (2016) concluded that the highest shoot length of wheat was secured when the seed primed with 10% PEG solution. Moghanibashi *et al.* (2013) reported that, as salinity levels increased, shoot length reduces but the priming treatments clearly improved the parameter, so can be used to improve seed performance of sunflower under normal and stress conditions.

Table 7. Effect of osmo and hydro priming and different salt concentration on the germination and growth behavior of mungbean

Treatments	Germination percentage (%)	Shoot length (mm)	Root length (mm)	Shoot dry weight (mg)	Root dry weight (mg)
Effect of osmo and hydro priming					
P_1	74.92 a	137.62 a	90.33 a	42.66 a	15.30 a
P_2	67.36 b	115.36 b	79.55 b	29.11 b	11.23 b
LSD $_{(0.01)}$	3.40	6.61	4.69	2.12	0.85
CV (%)	4.60	5.03	5.31	5.70	6.18
Effect of different salt concentrations					
T_0	87.39 a	149.6 a	112.8 a	50.46 a	17.78 a
T_1	82.67 a	142.8 ab	103.9 b	45.70 b	16.70 a
T_2	72.22 b	133.9 b	92.14 c	38.03 c	14.69 b
T_3	63.54 c	118.0 c	71.87 d	27.82 d	11.15 c
T_4	49.88 d	88.16 d	43.96 e	17.41 e	6.00 d
LSD $_{(0.01)}$	5.38	10.46	7.41	3.36	1.35
CV (%)	4.60	5.03	5.31	5.70	6.18

P_1: Osmo priming P_2: Hydro priming

T_0: 0 dSm^{-1}(Control); T_1: 5 dSm^{-1} NaCl; T_2: 10 dSm^{-1} NaCl ,T_3: 15 dSm^{-1} NaCl ,T_4: 20dSm^{-1} NaCl

4.3.3 Shoot dry weight (mg)

4.3.3.1 Effect of priming agents

Significant variation was found on shoot dry weight of Mungbean genotype for osmo (PEG) and hydro priming affected by different salinity levels (Table 7). Decreased shoot dry weight was found with increased salinity level where no salinity level gave highest shoot dry weight. The results showed that the highest shoot dry weight (42.66 mg) was observed from primed seeds (P_1) done with 10% PEG placed under salinity stress, where the lowest shoot dry weight (29.11 mg) was observed from P_2 (hydro priming) under salinity stress . Khalil *et al.* (2000), observed that dry matter yield increased with each increment of priming under normal and stress condition. Inhibition of seedling dry weight due to salt stress should be overcome by using PEG 6000 as osmo priming treatments in soybean. Kumar *et al.* (2017) experimented on chickpea and found that in case of seedling dry weight it was higher (1.02 gm to 1.59mg) in PEG 20% seeds followed by Mannitol 4% when compared with control.

4.3.3.2 Effect of salinity levels

The shoot dry weight of osmo primed (PEG) Mungbean varities varied significantly by different salinity stress conditions (Table 7). There were different extent of reduction was observed of shoot dry weight of Mungbean genotypes with the increasing of salt stress. Mungbean genotype primed with 10% PEG showed better tolerance capability under different salt stress conditions .The maximum shoot dry weight (50.46 mg) was received from PEG priming at normal condition (0 dSm^{-1} salt concentrations) T_0 and the minimum shoot dry weight (17.41mg) was received at 20 dSm^{-1} salt concentrations (T_4). Mohammadi and Amiri (2010) reported that seedling dry weight was reduced when drought stress level were increased from 0 to 1.5 MPa.

4.3.3.3 Intaraction effect of priming agents and salinity levels

Intaraction between priming and different salinity level showed significant variation in shoot dry weight (Table 8 and Appendix V). Decreased shoot dry weight was observed with increased salinity level where as no salinity level gave highest shoot dry weight. The results showed that the highest shoot dry weight (55.32 mg) was observed from primed seeds placed without salt treatment combination P_1 T_0 which is statistically identical with treatment combination P_1T_1 (primed seeds placed with 5 dsm^{-1} NaCl solution) , where the lowest shoot dry weight (8.80 mg) was observed from treatment combination P_2T_4 (hydro primed seeds placed with 20 dsm^{-1} NaCl solution). According to Rahman (2014), decreased shoot dry weight was observed with increased salinity level where no salinity level gave highest shoot dry weight.

4.3.4 Root length (mm)

4.3.4.1 Effect of priming agents

Root length of Mungbean genotypes was significantly influenced by osmo and hydro priming at different salinity levels (Table 7). Result exposed that the root length from primed seeds with PEG decreased significantly with increasing salinity level where no salinity stress gave highest root length. Results indicated that the highest root length (90.33mm) was observed in osmo priming (P_1) done with 10% PEG under stress of salt. The lowest root length (79.55 mm) was observed from P_2 (hydro priming) under salt stress. Significant improvement in root and shoot length may be attributed to earlier germination induced by primed over non-primed seeds (Farooq *et al.* 2005), which resulted in vigorous seedlings with more root and shoot length than the seedlings from non-primed seeds. Boureima *et al.* (2011) stated that root length increased by 19.94% at 0.5Mpa in comparison with control.

4.3.4.2 Effect of salinity levels

Different salinity levels showed significant difference in respect of root length (Table 7). Highest root length was found 112.8 mm under normal condition (T_0) which is statistically similar with T_1 (primed seeds placed with 5 dsm^{-1} NaCl solution). Lowest root length was obtained 43.96 mm from T_4 (at 20 dSm^{-1} salt concentrations). Moderate drought stress increase root length of pearl millet by 15.8% (Radhoaune, 2008). Badiow *et al*, (2004) reported that the development of root system in response to water deficit suggests that expression of certain genes controlling root formation is stimulated by salt stress condition. However the reduction of radicle length due to excess moisture stress could be due to a cessation in cellular division and elongation at root level (Ashagre *et al*., 2014).

4.3.4.3 Intaraction effect of priming agents and salinity levels

Intaraction between priming and different salinity level showed significant variation in root length (Table 8 and Appendix V).Highest root length was recorded from treatment combination P_1T_0 (115.3 mm) which is statistically similar with treatment combination P_1T_1, P_2T_0 and P_2T_1. The lowest root length was showed in treatment combination P_2T_4 (33.95 mm).From this result it can be expressed that Osmo primed seed showed better performance than hydro primed seed under different salinity level. According to Ashagre *et al.*, (2014) ,increase in PEG $_{6000}$ concentrations decreased germination percentage and rate, while shoot and root lengths and shoot fresh and dry weights decreased beyond 60g/L and increased up to 120g/L PEG but further increase in stress negatively influenced cultivars tolerance.

4.3.5 Root dry weight (mm)

4.3.5.1 Effect of priming agents

Significant variation was also found for root dry weight $seedling^{-1}$ at different priming agents affected by different salinity levels (Table 7). Decreased root

dry weight was observed with increased salinity level where no salinity level gave highest root dry weight. Results showed that the highest root dry weight (15.30 mg) was observed from P_1 (PEG). The lowest root dry weight (11.23 mg) was found from P_2 (Hydro primed). Judicious doses of PEG (Polyethylene Glycol) showed better tolerance at drought stress condition than hydro-priming, while more doses of PEG had negative effects on germination (Sun *et al*., 2010).

4.3.5.2 Effect of salinity levels

Significant variation was found for root dry weight at different salinity levels (Table 7). Decreased root dry weight was observed with increased salinity level where no salinity level gave highest root dry weight. Results showed that the highest root dry weight was in T_0 (17.78 mg) when primed seeds placed without salt. Under salinity stress, the highest root dry weight (16.70 mg) was found from (T_1) primed seeds placed with 5dsm^{-1} NaCl solution. The lowest root dry weight 6.00 mg was found from (T_4) Primed seeds placed with 20 dsm^{-1} NaCl. According to Rahman (2014), the highest root dry weight (8.23 mg) was observed from V_2 (ESWYT-5) where primed seeds placed without salt. Under salinity stress, the highest root dry weight (8.01 mg) was found from V_2 (ESWYT-5) under primed seeds placed with 5dsm^{-1} NaCl solution. The lowest root dry weight (3.76 mg) was found from V1 (BARI Gom-28) under Primed seeds placed with 20 dsm^{-1} NaCl.

4.3.5.3 Intaraction effect of priming agents and salinity levels

Intaraction between priming and different salinity level showed significant variation in root dry weight (Table 8 and Appendix V). In case of root dry weight, osmo primed genotype showed better tolerance capability under different salt stress conditions and hydro primed showed the most susceptibility towards the salinity.The maximum root dry weight (19.07 mg) was received from treatment combination P_1T_0 (osmo primed seed under 0 dSm^{-1} salt concentrations) and the minimum root dry weight (2.87 mg) was received from

treatment combination P_2T_4 (hydro primed seed under 20 dSm^{-1} salt concentrations). Under salinity stress, the highest root dry weight (18.35 mg) was found from treatment combination P_1T_1 (osmo primed seeds placed with 5dsm^{-1} NaCl solution).Dry weight is the consequence of plant physiological and biological activity.Under salt stress condition marked reduction was observed of this parameter in comparatively sensitive genotypes. Priming presumably permitted some repairs of damaged to membrane caused by deterioration by salinity stress and exerted better germination pattern and higher vigor level than comparatively sensitive genotypes Ruan *et al.* (2002). Removing of obstacle speed up the germination ultimately increased shoot dry weight of wheat genotypes Barsa *et al.* (2005).

Table 8. Interaction effect of osmo and hydro priming and different salt concentration on the germination and growth behavior of mungbean

Treatment combinations	Germination percentage (%)	Shoot length (mm)	Root length (mm)	Shoot dry weight (mg)	Root dry weight (mg)
P_1T_0	90.34 a	157.7 a	115.3 a	55.32 a	19.07 a
P_1T_1	86.55 a	152.9 ab	107.0 a-c	51.63 a	18.35 ab
P_1T_2	74.45 cd	146.7 a-c	96.63 cd	44.81 b	16.18 c
P_1T_3	67.09 de	128.1 de	78.74 e	35.55 cd	13.77 d
P_1T_4	56.18 f	102.7 g	53.96 g	26.02 e	9.120 e
P_2T_0	84.45 ab	141.6 b-d	110.2 ab	45.60 b	16.49 bc
P_2T_1	78.78 bc	132.6 c-e	100.9 bc	39.78 c	15.05 cd
P_2T_2	70.00 d	121.1 ef	87.66 de	31.26 d	13.20 d
P_2T_3	60.00 ef	107.9 fg	65.00 f	20.09 f	8.53 e
P_2T_4	43.58 g	73.66 h	33.95 h	8.80 g	2.87 f
$LSD_{(0.01)}$	7.61	14.79	10.48	4.75	1.90
CV (%)	4.60	5.03	5.31	5.70	6.18

P_1T_0= PEG primed at 0 dS/m, P_1T_1= PEG primed at 5 dS/m, P_1T_2= PEG primed at 10 dS/m, P_1T_3= PEG primed at 15 dS/m, P_1T_4= PEG primed at 20 dS/m ,P_2T_0= Hydro primed at 0 dS/m, P_2T_1= Hydro primed at 5 dS/m, P_2T_2= Hydro primed at 10 dS/m, P_2T_3= Hydro primed at 15 dS/m, P_2T_4= Hydro primed at 20 dS/m

4.3.6 Relative water content (%)

4.3.6.1 Effect of priming agents

Relative water content of different Mungbean genotypes was significantly influenced by different different priming agents under salt stress (Table 9). Result exposed that the relative water content from primed seeds decreased significantly with increasing salinity level. Results indicated that the highest relative water content (76.65%) was observed from P_1 osmo primed seeds. The lowest relative water content (61.03%) was observed from P_2 hydro Primed seeds. The leaves obtained from plots having seed primed with $CaCl_2.2H_2O$ (0.5%) showed significantly highest relative water content (Assefa, 2008).

4.3.6.2 Effect of salinity levels

Relative water content of Mungbean genotypes was significantly influenced by different salinity levels (Table 9). The relative water content from primed seeds decreased significantly with increasing salinity level. Highest relative water content was found 88.20 % in T_0 (normal condition) which is statistically similar with T_1 (PEG primed seeds placed with 5 dsm^{-1} NaCl). Lowest data was obtained 35.14% from T_4 (20 dsm^{-1} NaCl solution). (Rahman (2014) concluded that the relative water content from primed seeds decreased significantly with increasing salinity level.Under salinity stress, the highest relative water content (88.03%) was found from primed seeds placed with 5 dsm^{-1} NaCl solution. The lowest relative water content (78.05%) was observed from Primed seeds placed with 15 dsm^{-1} NaCl solution.

4.3.6.3 Intaraction effect of priming agents and salinity levels

Intaraction between priming and different salinity level showed significant variation in relative water content (Table 10 and Appendix VI)**.**According to analyzed data in the case of relative water content, highest relative water content was showed 93.54% in treatment combination P_1T_0 (PEG primed seed in normal condition) which is statistically identical with treatment combination

P_1T_1 (osmo primed seed with 5 dsm^{-1} NaCl and similar with P_1T_2, P_2T_0. Lowest data recorded 23.23% from treatment combination P_2T_4 (hydro primed seed with 20 dsm^{-1} NaCl). According to, Flower (1998),Under salt stress condition tolerance plant can grow vigorously, minimize the salt uptake and maximize potential salt load per unit area by their compartmentalization technique and provide better water use efficiency thus plant growth not hampered. Moreover, the osmo priming agent (PEG) triggered this technique for optimum water use efficiency and higher the relative water content was recorded for salt tolerance wheat genotype. According to , Nayyar *et al*. (2006), RWC was significantly reduced under saline stress condition, but in case of primed seed with 10% PEG solution the RWC was comparatively higher.

4.3.7 Water saturation deficit %

4.3.7.1 Effect of priming agents

Osmo priming (P_1) and hydro priming (P_2) showed significant difference after water saturation deficit in table 9. P_2 (38.97) showed higher water saturation deficit than P_1 (23.35). This result was in agreement with the previous work of Abdullah (2017) and Asaduzzaman (2014). Nahar *et al.* (2017) reported that relative water content and water saturation deficit had an inverse relation between them. The enzymatic activities were lower in non-prime seed which result produced the weak and lean seedling on the other hand due to over priming time, ageing process was accelerated and produced weak and lean seedling which were failed to uptake enough water and provided more water saturation deficit value.

4.3.7.2 Effect of salinity levels

Different salinity levels showed significant difference in respect of water saturation deficit (Table 9).Water saturation deficit increased with increasing salinity level. Highest water saturation deficit was found 64 .86% in 20 dsm^{-1} NaCl level (T_4). Lowest data was obtained 11.80% from normal condition (T_0). According to Nahar *et al*. (2017), the water saturation deficit of primed wheat

genotypes was gradually increased with increasing the salinity stress. The result of the experiment revealed that, under different salinity levels wheat genotype showed magnitude of water saturation deficit lowest was (10.87%) at 0 dSm^{-1} saline concentration compare to susceptibility the water saturation deficit was maximum (55.12%) at 20 dSm^{1} salt concentration.

4.3.7.3 Intaraction effect of priming agents and salinity levels

Intaraction between priming and different salinity level showed significant variation in water saturation deficit (Table 10 and Appendix VI). There is an inverse relationship between relative water content and water saturation deficit. The result of the experiment revealed that, lowest (10.02%) water saturation deficit was found from treatment combination P_1T_0 which is statistically similar with treatment combination P_1T_1. Water saturation deficit is lowest at 0 dSm^{-1} saline concentration and the water saturation deficit was maximum (76.77%) at treatment combination P_2T_4 20 dSm^{-1} salt concentration. Water saturation deficit is the opposite character of relative water content. So, presumably the water saturation deficit was higher for salinity sensitive genotype and vice versa. Due to lack of compartmentalization technique, the salinity sensitive cultivars fail to uptake enough water necessary for running the physiological process smoothly under salt stress condition, thus there will be a huge water deficit occur in sensitive cultivars than the tolerance cultivars. Similar result was reported by Baque *et al.* (2002) but osmo priming might help to recover this physiological damage and minimize the water saturation deficit.

4.3.8 Water retention capacity

4.3.8.1 Effect of priming agents

Increasing the salinity stress level significantly decreased water retention capacity. However, this decreasing trend was more noticeable for hydro primed seeds than for osmo primed seeds. Maximum water retention capacity (7.45%) was noticed in osmo primed (P_1) and minimum (6.08%) was noticed in hydro

primed seeds (P_2). The result also coincides with the result of Sangakkara *et al.* (1996) .Osmo priming helps to activate the metabolic enzymes responsible for germination before germination takes place, so the seedling from osmo primed seed got advantages to retain enough water under salt stress condition.

4.3.8.2 Effect of salinity levels

Different salinity levels showed significant difference in respect of water retention capacity (Table 9). The water retention capacity of primed genotypes was decreased gradually with increasing the salinity stress. Under different salinity levels maximum tolerance capability was showed in normal condition T_0 (10.24 %) and Lowest data was obtained from T_4 (2.91%). According to Nahar *et al.* (2017), water retention capacity is decreased with incressing salinity level and found maximum at normal condition.

Table 9. Effect of osmo and hydro priming and different salt concentration on the water relation behavior of mungbean

Treatments	Relative water content (%)	Water saturation deficit%	Water retention capacity%	Coefficient of germination%	Vigor index
Effect of osmo and hydro priming					
P_1	76.65 a	23.35 b	7.45 a	18.78 a	175.93 a
P_2	61.03 b	38.97 a	6.08 b	14.70 b	138.67 b
LSD $_{(0.01)}$	3.86	1.90	0.39	1.06	10.38
CV (%)	5.40	5.85	5.54	6.09	6.35
Effect of different salt concentrations					
T_0	88.20 a	11.80 e	10.24 a	21.80 a	229.7 a
T_1	83.93 a	16.07 d	8.76 b	19.95 b	204.5 b
T_2	76.34 b	23.66 c	6.77 c	17.61 c	163.5 c
T_3	60.58 c	39.42 b	5.14 d	14.06 d	121.4 d
T_4	35.14 d	64.86 a	2.91 e	10.27 e	67.35 e
LSD $_{(0.01)}$	6.11	3.00	0.61	1.67	16.41
CV (%)	5.40	5.85	5.54	6.09	6.35

P_1: Osmo priming P_2: Hydro priming

T_0: 0 dSm^{-1}(Control); T_1: 5 dSm^{-1} NaCl; T_2: 10 dSm^{-1} NaCl ,T_3: 15 dSm^{-1} NaCl ,T_4: 20dSm^{-1} NaCl

4.3.8.3 Intaraction effect of priming agents and salinity levels

Combined effect of priming and different salinity level showed significant variation in case of water retention capacity (Table 10 and Appendix VI).The maximum water retention capacity was found in the treatment combination P_1T_0 (10.68%). Minimum was recorded from P_2T_4 (1.98%) . The result also coincides with the result of Sangakkara *et al.* (1996), osmo priming helps to activate the metabolic enzymes responsible for germination of seed before germination takes place, so the seedling from osmo primed seed got advantages to retain enough water under salt stress condition.

4.3.9 Coefficient of germination%

4.3.9.1 Effect of priming agents

Osmo priming (P_1) and hydro priming (P_2) showed significant difference after analysis of variance in table 9. P_1 (18.78%) showed higher coefficient of germination than P_2 (14.70%).

4.3.9.2 Effect of salinity levels

Germination coefficient of osmo primed genotypes significantly varied by different level of salinity stress conditions (Table 9). The germination coefficient of primed genotypes was decreased in different extent with increasing the salinity stress. Maximum coefficient of germination was 21.80% , found in normal condition (T_0). Lowest data was obtained 10.27%, from (T_4) 20 dSm^{-1} salt concentration .

4.3.9.3 Intaraction effect of priming agents and salinity levels

Combined effect of priming and different salinity level showed significant variation in case of germination coefficient (Table 10 and Appendix VI). Maximum germination coefficient value was recorded from treatment combination P_1T_0 (22.70%) which is statistically similar with P_1T_1, P_2T_0, P_1T_2 and P_2T_4. Lowest was found in treatment combination P_2T_4 (11.71%), hydro

primed seed at 20 dSm^{-1} salt concentrations. The reduction of germination rate with the increasing of salt concentration occurred for salt sensitive cultivars which caused reduction of coefficient of germination under salt stress condition. Similar results were reported by Akbarimoghaddam *et al.* (2011).But when the seed was primed with PEG, it reduced the negative impact of salt stress and maintains a improve performance of germination coefficient. Seed priming resulted from anti-oxidant increment as glutathione and ascorbate in the seed. These enzymes trigger germination speed via reduction of lipid peroxidation activity; so germination coefficient was higher in osmo primed salt tolerance genotypes compare to sensitive one.According to Moghanibashi (2013) ,osmo priming of Italian ryegrass and sorghum seeds with 20% PEG_{8000} for 2 d at 10°C enhanced germination coefficient of wheat genotypes under water stress, , cold stress and saline conditions .

4.3.10 Vigor index

4.3.10.1 Effect of priming agents

Osmo priming (P_1) and hydro priming (P_2) showed significant difference in vigor index (Table 9). P_1 (237.8) showed higher vigor index than P_2 (138.67). Safiatou, (2012) stated that seedling vigour is improved by using seed priming solutions in sorghum and Bambara groundnut. Also, highest seedling vigour was revealed by osmo-priming (Mannitol priming) in Bambara groundnut and by hydro-priming in sorghum.

4.3.10.2 Effect of salinity levels

Different salinity levels showed significant difference in respect of vigor index (Table 9). Highest vigor index was found in normal condition T_0 (229.7) Lowest data was obtained from T_4 (67.35) at 20 dSm^{-1} salt concentration. Moderate vigor index found in T_1 at 5 dSm^{-1} salt concentration. This result was supported by previous findings of Baque *et al*. (2016) and Maiti *et al.* (2013).

4.3.10.3 Intaraction effect of priming agents and salinity levels

Combined effect of priming and different salinity level showed significant variation in case of Vigor index (Table 10 and Appendix VI).The maximum vigor index was recorded from treatment combination P_1T_0 (246.8) which is statistically similar with P_1T_1 and lowest was found in treatment combination P_2T_4 (46.85). Vigor index was recorded 225.1 from treatment combination P_1T_1 (osmo primed seed with 5 dsm^{-1} NaCl solution). Moghanibashi, *et al.* (2013) reported that drought and salinity levels increased, vigour reduced but the priming treatments clearly improved the parameter under drought and salinity conditions so these treatments can be used to improve seed performance of sunflower under normal and stress. Sadeghi *et al*. (2013), reported that vigor of soybean plant was probably due to the reserve mobilization of food material, activation of enzymes by osmotic priming .

Table 10. Interaction effect of osmo and hydro priming and different salt concentration on the water relation behavior of mungbean

Treatment combinations	Relative water content (%)	Water saturation deficit%	Water retention capacity%	Coefficient of germination%	Vigor index
P_1T_0	93.54 a	6.46 f	10.68 a	22.70 a	246.8 a
P_1T_1	89.98 ab	10.02 f	9.53 b	21.84 ab	225.1 ab
P_1T_2	82.07 bc	17.93 de	7.52 c	19.71 bc	181.0 c
P_1T_3	70.63 d	29.38 c	5.68 d	16.47 de	138.9 d
P_1T_4	47.05 e	52.95 b	3.84 e	13.20 fg	87.84 e
P_2T_0	82.86 bc	17.14 e	9.81 b	20.90 ab	212.7 b
P_2T_1	77.89 cd	22.11 d	8.02 c	18.07 cd	184.0 c
P_2T_2	70.61 d	29.39 c	6.01 d	15.52 ef	146.0 d
P_2T_3	50.54 e	49.46 b	4.59 e	11.66 g	103.8 e
P_2T_4	23.23 f	76.77 a	1.98f	7.33 h	46.85 f
LSD (0.01)	8.64	4.24	0.87	2.37	23.20
CV (%)	5.40	5.85	5.54	6.09	6.35

P_1T_0: PEG primed at 0 dS/m, P_1T_1: PEG primed at 5 dS/m, P_1T_2=: PEG primed at 10 dS/m, P_1T_3: PEG primed at 15 dS/m, P_1T_4: PEG primed at 20 dS/m ,P_2T_0: Hydro primed at 0 dS/m,P_2T_1:Hydroprimed at 5 dS/m, P_2T_2= Hydro primed at 10 dS/m, P_2T_3= Hydro primed at 15 dS/m, P_2T_4= Hydro primed at 20 dS/m

CHAPTER V

SUMMARY AND CONCLUSION

The experiment was carried out at the Agronomy Laboratory of Wazed Mia Research Centre,of Sher-e-Bangla Agricultural University Dhaka, from 25 May to 15 August, 2018 to study induction of salt tolerance capability of Mungbean (*Vigna radiata L.*) through Polyethylene Glycol and hydro priming. The present study was conducted with three different experiments those laid out in Completely Randomized Design (CRD) with five replications.

In the 1st experiment, Mungbean seeds of BARI Mung 5 and BARI Mung 6 were pre-soaked in water, 5, 10, 15 and 20% PEG solution and untreated seeds were served as control. Results revealed that all the characters viz. germination percentage (GP), shoot length, root length, shoot and root dry weight, relative water content, water saturation deficit, water retention capacity, coefficient of germination and vigor index (VI) were significantly influenced by various PEG concentrations. All the parameters of both varieties gave the best results when seeds treated with 10% PEG solution compared to nonprimed and hydro primed seeds and decreased gradually with increasing PEG concentration.

In the 2nd experiment, BARI Mung 6 was primed in 3,6,9,12,15 and 18 hours under 10% PEG solution and distilled water. Results revealed that all the characters viz. germination percentage (GP), shoot length, root length, shoot and root dry weight, relative water content, water saturation deficit, water retention capacity, coefficient of germination and vigor index (VI) were significantly influenced by different priming time. Osmo priming showed better result than hydro priming. All the parameters increased with increasing priming time up to 9 hr after then decreased gradually. The highest GP, shoot length, root length, shoot and root dry weight, relative water content, water retention capacity, coefficient of germination and vigor index were obtained from seeds pre-treated with 10% PEG solution for 9 hr .Whereas the highest

water saturation deficit was obtained from when seeds were hydro primed for 3hr.

In the 3rd experiment osmo primed and hydro primed seeds were allowed to grow under (Normal) 0 dS/m, 5 dS/m, 10 dS/m, 15dS/m and 20 dS/m NaCl solution induced salt stress. Results revealed that under stress condition osmo priming showed better result than hydro primed seeds in case of all the characters viz. germination percentage, shoot length, root length, shoot and root dry weight, relative water content, water retention capacity, coefficient of germination and vigor index except water saturation deficit. Under normal condition (without stress) all the characters germination percentage, shoot length, root length, shoot and root dry weight, relative water content, water retention capacity, coefficient of germination and vigor index. Osmo primed seed under moderate stress condition showed better result than hydro primed seed in case of above all parameters except water saturation deficit.

Response of Mungbean varieties was different to pretreatments. Germination behavior and water relation behavior of both varieties gave the best results when seeds treated with 10% PEG solution compared to nonprimed and hydro primed seeds and decreased gradually with increasing PEG concentration. All the parameters except water saturation deficit gave the best result from seeds pre-treated with 10% PEG solution for 9 hr after that decreased gradually. Seeds pre-soaked with 10% PEG showed better performance than hydro primed seed in terms of germination behavior and water relation behaviours compared under salt stress. So, Mungbean seed primed with 10% PEG for 9 hr is considered as best priming concentration and priming time to induce salt tolerance capability (upto 10 dSm^{-1} NaCl) of Mungbean enhancing germination behavior and water relation behavior of Mungbean than hydro priming.

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APPENDICES

Appendix I. Analysis of variance of the data on germination and growth behavior of Mungbean as influenced by combined effect of different varieties and priming solution

Source of variation	df	Mean square of				
		Germination percentage	Shoot length	Root length	Shoot dry weight	Root dry weight
Variety (A)	1	285.58**	4491.25**	455.73**	388.88**	50.67**
Priming solution (B)	5	296.84**	5992.23**	1559.36**	694.01**	69.41**
Variety (A) X Priming solution (B)	5	14.94**	134.13**	5.84**	16.95**	0.66**
Error	48	20.84	45.74	9.67	3.55	0.66

**Significant at 1% level of significance

^{NS} Non significant

Appendix II. Analysis of variance of the data on growth and water relation behavior of Mungbean as influenced by combined effect of different varieties and priming solution

Source of variation	df	Mean square of				
		Relative water content	Water saturation deficit	Water retention capacity	Germination Coefficient	Vigor index
Variety (A)	1	851.95**	859.42**	4.42**	80.64**	11209.95**
Priming solution (B)	5	2319.54**	2318.62**	8.25**	29.54**	16046.89**
Variety (A) X Priming solution (B)	5	24.24**	24.32**	0.20**	0.17**	483.07**
Error	48	15.26	3.19	0.25	0.93	53.67

**Significant at 1% level of significance

^{NS} Non significant

Appendix III. Analysis of variance of the data on germination and growth behavior of Mungbean as influenced by combined effect of different priming agent and priming time

Source of variation	df	Mean square of				
		Germination percentage	Shoot length	Root length	Shoot dry weight	Root dry weight
Variety (A)	1	349.38**	2248.02**	880.21**	410.40**	19.23**
Priming solution (B)	5	1029.85**	3594.84**	869.67**	421.83**	47.22**
Variety (A) X Priming solution (B)	5	14.89**	58.07**	3.88**	2.49**	0.80**
Error	24	13.71	47.44	8.05	4.42	0.59

*Significant at 1% level of significance

NS Non significant

Appendix IV. Analysis of variance of the data on growth and water relation behavior of Mungbean as influenced by combined effect of different priming agent and priming time

Source of variation	df	Mean square of				
		Relative water content	Water saturation deficit	Water retention capacity	Germination Coefficient	Vigor index
Variety (A)	1	345.46**	346.02**	8.06**	92.06**	8606.58**
Priming solution (B)	5	1448.47**	1448.37**	12.18**	130.69**	14375.65**
Variety (A) X Priming solution (B)	5	2.76**	2.76**	0.07**	4.13**	69.64**
Error	24	14.35	2.98	0.26	1.33	76.02

**Significant at 1% level of significance

NS Non significant

Appendix V. Analysis of variance of the data on germination and growth behavior of Mungbean as influenced by combined effect of different priming agent and salt concentration

Source of variation	df	Mean square of				
		Germination percentage	Shoot length	Root length	Shoot dry weight	Root dry weight
Variety (A)	1	428.65**	3713.86**	872.43**	1378.37**	124.24**
Priming solution (B)	4	1361.74**	3592.96**	4557.90**	1079.64**	137.38**
Variety (A) X Priming solution (B)	4	14.29**	38.75**	56.70**	12.99**	3.80**
Error	20	10.72	40.54	20.34	4.18	0.67

**Significant at 1% level of significance

NS Non significant

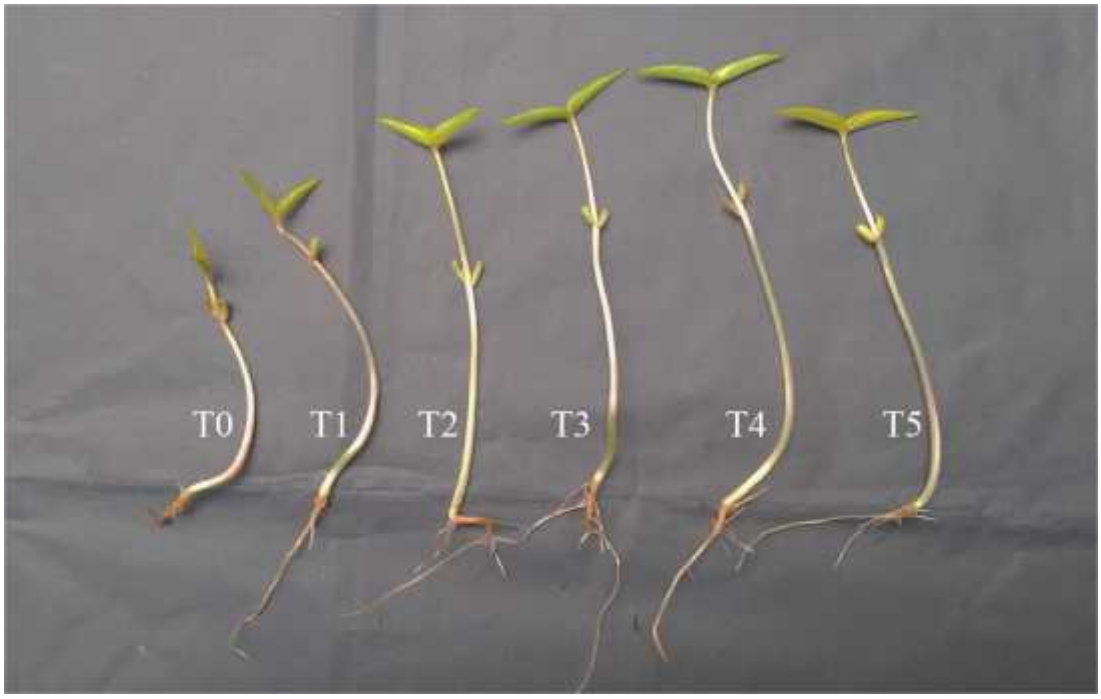
Appendix VI. Analysis of variance of the data on growth and water relation behavior of Mungbean as influenced by combined effect of different priming agent and salt concentration

Source of variation	df	Mean square of				
		Relative water content	Water saturation deficit	Water retention capacity	Germination Coefficient	Vigor index
Variety (A)	1	1831.29**	1831.13**	13.97**	125.18**	10411.19**
Priming solution (B)	4	2794.07**	2794.11**	50.52**	128.63**	25347.99**
Variety (A) X Priming solution (B)	4	53.03**	53.01**	0.23**	3.38**	18.03**
Error	20	13.82	3.33	0.14	1.04	99.76

**Significant at 1% level of significance

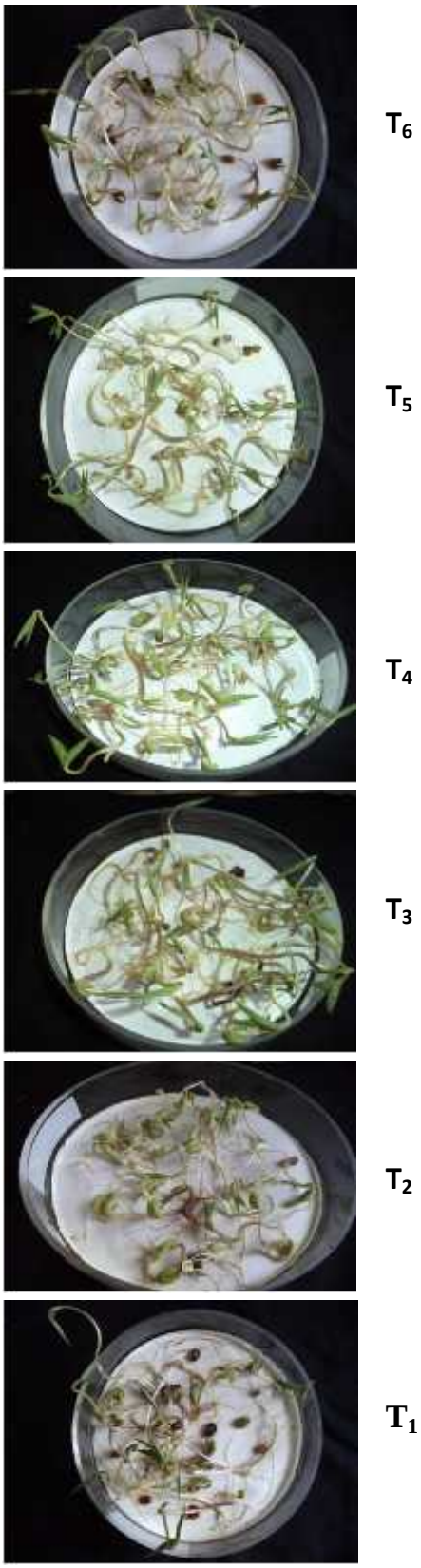
NS Non significant

PLATES



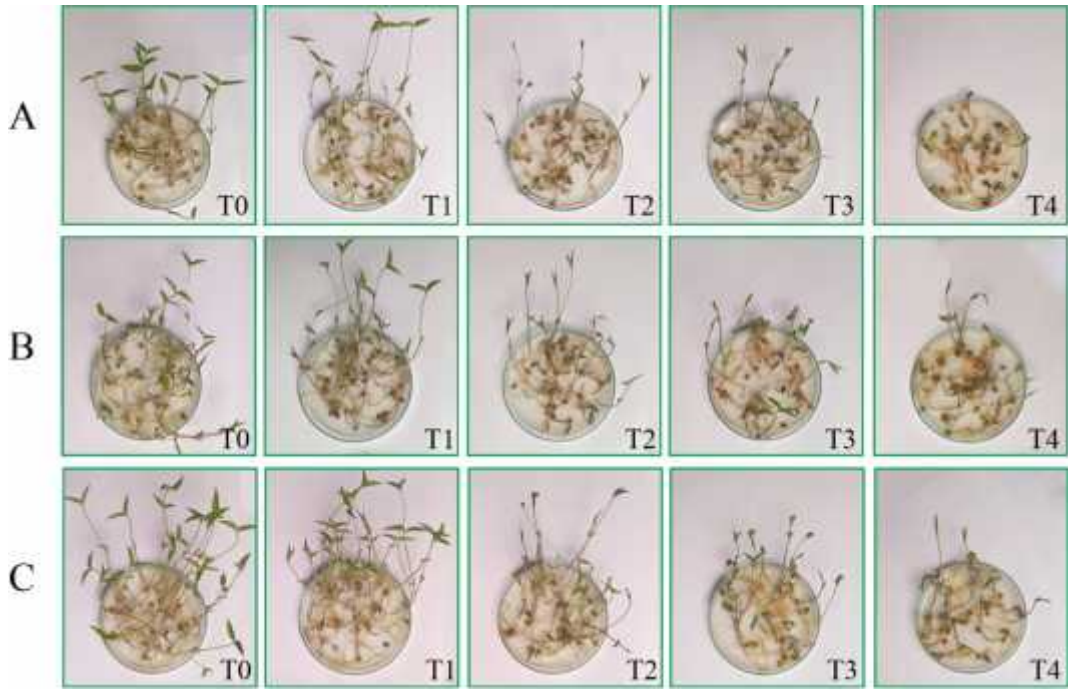
T_0 = Seeds without priming (control); T_1 = Seeds primed with 0% PEG for 9 hours; T_2 = Seeds primed with 5% PEG for 9 hours, T_3 = Seeds primed with 10% PEG for 9 hours; T_4 = Seeds primed with 15% PEG for 9 hours, T_5 = Seeds primed with 20% PEG for 9 hours

Plate 1: Effect of priming solutions on germination behavior of BARI Mung 6



T_1=3hr, T_2=6hr, T_3=9hr, T_4=12 hr, T_5=15 hr, T_6=18 hr

Plate 2: Effect of priming time on germination behavior of BARI Mung 6



T_0= Primed seeds placed without salt 0 dS/m (control), T_1= Primed seeds placed with 5 dS/m NaCl,T_2= Primed seeds placed with 10 dS/m NaCl,T_3= Primed seeds placed with 15 dS/m NaCl andT_4= Primed seeds placed with 20 dS/m NaCl

Plate 3: Effect of priming solution on germination behavior of BARI Mung 6 at different salinity level