INDUCTION OF DROUGHT TOLERANCE IN WHEAT THROUGH OSMO AND HYDRO PRIMING

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INDUCTION OF DROUGHT TOLERANCE IN WHEAT THROUGH OSMO AND HYDRO PRIMING

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CERTIFICATE

This is to certify that the thesis entitled, "**INDUCTION OF DROUGHT TOLERANCE IN WHEAT THROUGH OSMO 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 **MUNMUN NAHAR**, Registration No. **08-03028** 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
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Supervisor

Dedicated

To My

Beloved Parents

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INDUCTION OF DROUGHT TOLERANCE IN WHEAT THROUGH OSMO AND HYDRO PRIMING

Abstract

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. In the 1st experiment, wheat seeds of BARI Gom 27 and BARI Gom 28 were pre-soaked in 0, 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 index (GI) of wheat seed. The highest GP (95.55%), VI (201.00) and GI (43.73) were obtained from seeds of BARI Gom 27 pre-treated with 10% PEG solution compared to BARI Gom 28 (75.55%, 128.71 and 27.12 of GP, VI and GI, respectively) and then decreased gradually with increasing PEG concentration. In the 2nd experiment, BARI Gom 27 was primed in 0 to 18 hours under 10% PEG solution and distilled water. The highest GP (87.77%), VI (142.31) and GI (41.23) were obtained from seeds pre-soaked in 12 hours with 10% PEG solution compared to hydro-priming (84.44%, 133.83 and 36.62 of GP, VI and GI, respectively) and untreated control (57.77%, 84.85 and 29.75 of GP, VI and GI, respectively), and then decreased gradually with increasing priming time. In the 3rd experiment non primed and primed seeds (osmoprimed and hydroprimed) were germinated under 0, 5, 10, 15 and 20% PEG solution induced drought stress conditions. Results 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 hydroprimed seeds decreased drastically as drought stress increased but osmoprimed 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.

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LIST OF ACRONYMS

ABBREVIATIONS	ELABORATIONS
%	Percent
@	At the rate
°C	Degree centigrade
AEZ	Agro-Ecological Zone
BARI	Bangladesh Agricultural Research Institute
BAU	Bangladesh Agricultural University
BBS	Bangladesh Bureau of Statistics
CHO	Carbohydrate
cm	Centimeter
Conc.	Concentration
CRD	Completely Randomized Design
CV	Coefficient of Variation
cv.	Cultivar (s)
DMRT	Duncan's Multiple Range Test
DW	Dry weight
e.g.	exempli gratia (for example)
et al.	et alii (and others)
etc.	et cetera (and the rest)
FAO	Food and Agriculture Organization
FW	Fresh weight
GP	Germination Percentage
GI	Germination Index
g	Gram
h	Hour (s)
ha	Hectare
i.e.	id est (that is)
kg	Kilogram
LSD	Least Significant Difference
mm	millimeter
PEG	Poly Ethylene Glycol
ROS	Reactive Oxygen Species
VI	Vigour Index

Chapter I

Introduction

CHAPTER I

INTRODUCTION

Wheat is the most important cereal crop of the world. It ranks first in area (213600 thousand hectares) and third in production (576317 thousand metric tons) among the grain crops in the world (FAO, 2000). It is the second most important cereal crop in Bangladesh next to rice (BARI, 1993).

In irrigated land, winter wheat and transplanted aman is the dominant rotation. Planting of winter wheat is delayed after harvesting of transplanted aman. In addition, low precipitation and inadequate moisture around seed zone under rainfed conditions reduce grain yield potential (Cantliffe *et al.*, 1994).

Plants are exposed to various abiotic factors throughout the course of their growth and development (Zhao *et al.,* 2007). The major abiotic stresses to which plants are exposed include extreme temperature, drought or high salinity. These stresses are the most significant factors leading to substantial and unpredictable loss in crop production in agriculture (Jakab *et al.,* 2005). In most plants, drought or salinity causes a variety of biochemical, physiological and metabolic changes (Xiong and Zhu, 2002), which may result in oxidative stress and affect plant metabolism, performance and thereby the yield (Shafi *et al.,* 2009). Salt and osmotic stresses are also responsible for both inhibition or delayed seed germination and seedling establishment (Almansouri *et al.,* 2001). The physiological mechanisms through which plants respond to salinity and drought show high similarity, suggesting that both stresses must be perceived by the plant cell as deprivation of water (Tavili *et al.,* 2011).

Plant interaction with environmental stress factors is known to lead to the activation of various defense mechanisms resulting in a qualitative and/or quantitative change in plant metabolite production, activation of hormone signaling pathways regulated by abscisic acid, salicylic acid, jasmonic acid and ethylene, as well as reactive oxygen species (ROS) signaling pathways (Fujita *et al.,* 2006). Moreover, both environmental and biotic stresses can induce emissions of an array of organic compounds in any plant species, whereas the magnitude of emissions induced by given stress depends on stress tolerance, timing, duration and severity (mild vs. strong) of the stress (Niinemets, 2009).

Various methodologies were adapted from time to time to achieve tolerance to stresses. These include, conventional breeding methods such as selection and hybridization and modern methods such as mutation breeding, polyploidy breeding, genetic engineering, etc. The conventional breeding techniques have limitations like requirements of large man power, energy, etc. Attempts to generate plant varieties with improved salinity and drought tolerance using selection-based breeding strategies have proved largely unsuccessful mostly because of the well recognized complexity or multigenic nature of salinity and drought-tolerance traits (Cushman and Bohnert, 2000; Flowers *et al.,* 2000). Attempts were also made to produce transgenic plants which can withstand various kinds of stresses. Genetic engineering holds the potential of being reasonably fast and predictable in its consequences because of the targeted introduction of individual, heterologous traits into elite crop lines (Gust *et al.,* 2010). The incorporation of transgenes into breeding programs is not a simple process. Due to the effects such as pleiotropy and gene silencing, it is not possible to continue through the plant breeding process with the precision with which the selection of the gene was begun (Flowers *et al.,* 1997). Besides this major drawback, these methods are also expensive, cumbersome, and there exist biosafety regulations and restrictions which hinder the introduction of transgenic into the field. Due to the above-mentioned limitations of the available techniques, it has become imperative to think of an alternative solution to impart tolerance to plants against various stresses. The alternative solution would be more acceptable if it is simple, cost effective and can be adopted by the farmers without any complication and at the same time it should be effective in manifesting the tolerance.

Seed priming is considered as a promising approach to increase stress tolerance capacity of crop plants including drought. 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. The physiological state in which plants are able to faster or better activate defense responses or both is called the primed state of the plant (Beckers and Conrath, 2007). Seed priming can be accomplished through different methods such as hydro-priming (soaking in DW), osmo-priming (soaking in osmotic solutions such as PEG, potassium salts, e.g., KCl, K_2SO_4) and plant growth inducers (CCC, Ethephon, IAA) (Capron *et al.,* 2000; Chiu *et al.*, 2002; Harris *et al.,* 1999; Chivasa *et al.*, 1998).

Seed priming is also widely used to synchronize the germination of individual seeds (Taylor and Harman, 1990). Seed-priming technology has twofold benefits: enhanced, rapid and uniform emergence, with high vigor and better yields in vegetables and floriculture (Bruggink *et al.,* 1999) and some field crops (Basra *et al.,* 2005; Kaur *et al.,* 2005). According to McDonald (2000), primed seeds acquire the potential to rapidly imbibe and revive the seed metabolism thus enhancing the germination rate.

In many crops, seed germination and early seedling growth are the most sensitive stages of water limitation and the water deficit may delay the onset and reduce the rate and uniformity of germination, leading to poor crop performance and yield (Demir *et al.,* 2006). Therefore, the beneficial effects of priming may be more evident under unfavorable rather than favorable conditions (Parera and Cantliffe, 1994). Primed seeds usually exhibit an increased germination rate, greater germination uniformity, and at times, greater total germination percentage (Basra *et al.,* 2005). These attributes have practical agronomic implications, notably under adverse germination conditions (McDonald, 2000). Therefore, there is a strong interest in the seed industry to find suitable priming agent(s) that might be used to increase the tolerance of plants under adverse field conditions (Job *et al.,* 2000).

Objectives of the research work:

- To evaluate the effect of pre-sowing seed treatment with various concentrations of Polyethylene Glycol (PEG) on germination behavior of wheat in relation to drought tolerance.
- To optimize the priming time of the best priming chemical on germination behavior of wheat.
- To evaluate the effect of pre-sowing seed treatment with Polyethylene Glycol (PEG) and water on germination behavior of wheat under drought stress condition.

Chapter II

Review of Literature

CHAPTER II

REVIEW OF LITERATURE

A number of literatures relating to the performance of different priming such as hydropriming, osmopriming, halopriming etc. used alone or together with different priming time on different crops cultivation were available. But, performances of different priming on wheat, second important cereal crop of Bangladesh, were not available. The review of literature given below was based on the performance of different priming on different crops cultivation. The review includes reports of several investigators which appear pertinent in understanding the problem and which may lead to the explanation and interpretation of results of the present investigation.

İşeri *et al.* (2015) reported that organic and inorganic pre-treatments were applied to cucumber (*Cucumis sativus* L.) seeds. Salicylic acid (SA), mannitol, ascorbic acid (AscA), KCl, and $CaCl_2.2H_2O$ pre-treatments enhanced germination and seedling vigor. Seedlings obtained from the pre-treatd seeds with 0.2mM SA, 0.4mM AscA, 2% mannitol, 100mM KCl, and 50mM $CaCl_2$ were transferred to field. Under normal irrigation (NI), SA and AscA groups, and under drought (Dr) treatment groups had higher increase in height. SA group had higher fruit yield/plant under NI, whereas KCl, mannitol, $CaCl_2$ had lower inhibition in fruit yield/plant under Dr. KCl group yielded higher fruit weight under NI. All treatment groups maintained higher leaf relative water content. Under NI anthocyanin in leaves of SA, $CaCl_2$ and mannitol groups were higher. NI and Dr anthocyanin levels did not differ in these groups. Total carotenoid levels were higher in pre-treatment groups under NI and Dr. Seed priming may enable establishment of more drought tolerant crops with higher crop yield.

Özdemir and Sade (2015) conducted this study to determine effects of seed priming to physiological parameters (PP) at drought conditions (DC) on bread wheat. NC (irrigated as needed) and DC (soil saturated with 25% distilled water for 48 hours) pots. Seventh and fourteenth days after emergence physiological parameters [(chlorophyll content (CC), stomatal conductance (SC), photosynthetic efficiency (PE), proline content (PC), MDA (malondialdehyde)] were determined. MDA level at DC is (37, 62 nmol/g) higher than NC (32, 17 nmol/g). At NC except MDA and PC, all parameters increased. PC level of plant materials at DC (9, 44 µmol/g) was significantly higher than PC of NC (4, 76 µmol/g). Seedlings from primed seeds, better adapted to DC. Seedlings grew primed seeds have more CC, less MDA. CC of Altay 2000 at DC (38, 96 spad) significantly decreased compared to NC (48, 92 spad). The same situation was came across with Kirac 66 also (DC; 34, 69 spad; NC; 47, 38spad). Generally response of seedlings Altay 2000 to priming was better than Kirac 66. According to results, it is observed that seed priming is a practical method, can be used drought areas. Pre-treatment is an alternative method that can trigger resistance towards water deficit conditions.

Rouhi *et al.* (2015) conducted a study to evaluate the role of hydro and osmopriming on improvement of germination parameters of Tall Wheatgrass (Agropyronelongatum) under simulated drought stress by PEG. Germination was delayed by drought stress in primed and nonprimed seeds. With increasing the osmotic potential, all of the investigated traits began to decrease. Seeds were able to germinate at 0, −0.4 and −0.8 MPa concentrations of PEG but dearth seed germination was observed at −1.2 MPa. Osmopriming increased germination parameters under drought stress rather than hydropriming treatment. Finally, Tall Wheatgrass did have positive reaction in both priming treatments on germination parameters especially at lowest osmotic potentials (-1.2 MPa).

Khalilzadeh and Khanpaye (2014) mentioned that seed priming was used to reinforcement of barley seedling growth under water deficit stress in a greenhouse condition. Barley seeds were primed with humic acid, Pseudomonas Spp., Marmarin, distilled water (hydropriming) and none (as control) under four levels of water deficit stress (irrigation at 20 (I1), 40 (I2), 60 (I3) and 80% (I4) field capacity). Results indicated that all measured parameters were decreased with increasing the stress levels, except root length and root-shoot ratio. The highest value of root length (18.42 cm) and root-shoot ratio (2.84) was obtained in the I1 irrigation regime. However, I2, I3 and I4 irrigation regimes did not significantly affect on barley seedling traits, but I1 irrigation regime exhibited better growth. Seed priming with Pseudomonas affected root length, root and shoot dry weight, plant height and SPAD, significantly. Seed priming with Marmarin showed best results on SPAD. The maximum (0.37 g/plant) shoot dry weight was obtained from I2 irrigation regime with hydropriming treatment. Whereas, the lowest shoot dry weight (0.14 g/plant) was observed in I1 irrigation regimes in control condition. Hydropriming shows better response in root length, root and shoot and plant height and was equal with Pseudomonas, so it could be considered as a suitable substitute in organic agricultural systems.

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.

Ajirloo *et al.* (2013) conducted an experiment where seeds were primed for 20, 40 and 60 hours in seven priming media (PEG 5%, PEG 10%, KNO_3 1%, KNO_3 2%, KCl 2%, KCl 4% and distilled water as control). Results show that when seed primed by KNO_3 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.

Dastanpoor *et al.* (2013) reported that the germination of *Salvia officinalis* L. (sage) seeds is a problem of great concern that may be overcome by employing seed priming techniques. Seed priming is an efficient technique for improvement of seed vigor, increasing germination and seedling growth. Little information has been reported on seedling development of sage subsequent to seed priming. The influence of hydropriming treatments on seed parameters of S. officinalis L. was investigated. Seeds of sage were treated by hydropriming at three temperatures 10, 20, 30°C for 0, 12, 24 and 48 h. The hydropriming clearly improved the final germination percentage (FGP), mean germination time (MGT) and synchronized the germination of seeds at each three temperature. All the seed treatments resulted in germination enhancement except hydroprimed seeds for 48 h at temperature 30°C. However, hydropriming (12 h at 30°C) was most effective in improving seed germination that FGP was increased by 25.5% as compared to that of non-primed seeds.

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 osmopriming 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 osmopriming treatments in soybean. Among the materials used for osmopriming PEG 6000 has the greatest impact on mitigating the effects of drought stress on germination and early growth stages.

Jisha *et al.* (2013) reported that plants are exposed to any 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. 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 more quickly or aggressively. Moreover, plants raised from primed seeds showed sturdy and quick cellular defense response against abiotic stresses. Priming for enhanced resistance to abiotic stress obviously is operating via various pathways involved 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. 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 and its effects.

Maiti *et al.* (2013) observed that seed priming increases seedling vigour of several vegetable crops. The priming techniques improved seedling vigour, growth and yield of tomato and chilli although varieties showed variation in response to different treatments. Especially halopriming increased speed of emergence, seedling vigour index, root length and shoot length over hydropriming in tomato and chilli. At field level halo priming showed better performance than control and hydropriming. Halo priming caused early flowering in tomato (8149, 8152) and chilli (9357). Increased plant height is also noticed in halopriming with respect to tomato and chilli (8149, 8152, 9357 and 9366). Mostly importantly halopriming increased total yield 20 plants-1 in tomato and chilli. This is the first observation in the aspect of increased yield under field condition with respect to seed priming in vegetable crops.

Maiti *et al.* (2013) observed that seed priming is a useful technique for enhancing seed vigor, and thereby, improving overall germination and seedling development in crop species. In tomato variety germination percentage, early flowering, plant height and leaf length were increased in response to hydro-priming treatment. In okra and onion, seedling vigor was increased in osmo-priming. In case of watermelon, halo-priming and osmo-priming increased seedling vigor. In case of chilli, halo-priming increased seedling vigor. With respect to sponge gourd, osmo-priming increased seedling vigor. In case of ridge gourd, halo-priming increased seedling vigor. In general it is observed that the yield is increased in case tomato, chilli, and okra apart from seedling vigor. As the longevity of primed seeds is short, the priming technique may be used in increasing seed production of foundation seeds and breeder's seeds. In addition, seed priming could improve seedling vigor in seedling nursery.

Moghanibashi *et al.* (2013) 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 osmopriming 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.

Abbasi *et al.* (2012) conducted an experiment on germination of Agropyron elongatum where first factor was priming (control, hydropriming and osmopriming) and the second was osmotic potential levels (0, -0.3, -0.6, -0.9 and -1.2 MPa). Results indicated that hydropriming is a simple and useful technique for enhancing seedling emergence rate and percentage of Agropyron elongatum. In general, this study revealed that seed priming especially with hydropriming improved Agropyron elongatum seed performance under drought stress condition. However, the improvements were more obvious at the higher levels of drought stress.

Elif *et al.* (2012) mentioned that growth parameters and relative water content values of seedlings at normal conditions higher than drought. At the research highest fresh weight was determined at KH_2PO_4 treatment (32,4083 mg), similar results were determined dry weight (9,6206 mg) and seedling length (10, 3793 cm) at the same treatment also. According to results, priming is an alternative approach can trigger resistance at drought conditions.

Islam *et al.* (2012) conducted an experiment to study the efficacy of three osmopriming agents viz ., NaCl, KC1 and CaCb with control on seed germination and seedling growth of three T. aman rice varities viz., BRRI dhan40, BRRI dhan41 and BINA dhan7. Among the chemicals CaCb showed best as osmopriming agents and it enhanced all germination parameters. BRRI dhan41 showed the best performance. Better root length and shoot length were observed in control. Seeds of BRRI dhan41 when treated with NaCl showed the highest germination percentage (98.67%), germination energy (88%), germination speed (96.83%), but vigor index (10.63) was found highest in BRRI dhan40 when it was treated with KC1. BRRI dhan41 produced the largest root (8.56 cm) when seeds were treated with NaCl. BRRI dhan40 (10.51 cm) and BINA dhan7 (10.53 cm) produced the highest shoot length at controlled condition and CaCl2 treated seed, respectively.

Moghanibashi *et al.* (2012) reported that the effect of hydropriming 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, D50 and GI under all salinity and drought levels as compared with non-primed seeds. There was interaction between cultivar and priming on the germination rate and D50 as hydropriming was more effective in cultivar Urfloar. There was also interaction between priming and drought and salt stress on the Gmax, R50, D50 and GI. Primed seeds clearly produced higher Gmax, R50, D50 and GI than non-primed seeds under all drought and salinity levels. There was interaction among cultivar, drought and salt stress for all the parameters which were measured. Cultivar Urfloar clearly produced higher Gmax, R50, D50, GI, root and shoot length and their dry weight as compared with cultivar Blazar. The results suggested that hydropriming for 24 h was enhanced germination and seedling growth of sunflower under stress conditions. Therefore, this treatment may be used to improved seed performance of sunflower under normal and stress conditions.

Moradi *et al.* (2012) reported that seed priming increased germination percentage, germination speed, seedling length, root-shoot ratio and decreased mean germination time. The effectiveness of these treatments varied depending on the level of the stress (zero or -8 bar), and the priming condition including temperature, duration, type and osmotic potential of solution. The lower priming duration (ie., 12 and 24 h) improved germination under normal condition, while a higher priming duration (ie., 36 and 48 h) provided more protection when the seeds were exposed to drought stress. In this study, priming with urea had better results than other treatments. Results showed that seed priming with fertilizers might serve as an appropriate treatment for advancing germination under optimum and drought stress conditions.

Omid and Farzad (2012) observed the impacts of various concentrations of polyetylenglycol 6000 (0, -9, -11, -13 and -15 bar) and hydro priming on Mountain Rye germination characteristic and enzyme activity under drought stress. Analyze of variance for hydro priming showed that temperature × time of priming interaction was significantly for germination percentage (GP), normal seedling percentage (NSP), coefficient of velocity of germination (CVG), seedling vigor index (SVI), coefficient of allometry (AC) and seedling length (SL) under drought stress and for osmo priming showed that Concentration of PEG × Temperature × Time of priming interaction was significantly for all traits under drought stress. Results of interaction effects for hydro priming showed that the highest GP (53%) and NSP (23.5%) were attained from hydro priming for 16h at 15◦C and the highest CVG (0.21) and AC (0.49) were attained from hydro priming for 8h at 10◦C, also hydro priming for 8h at 15◦C increased SL (3.15) as compared to the unprimed. Osmo priming with concentration of -15 bar PEG for 24h at 15◦C increased GP (80.5 %), GI (17.9), NSP (45 %), SVI (257.85) and SL (5.73 cm) and decreased MTG as compared to the unprimed and other treatments of osmo priming. The highest CVG was attained from concentration of -9 bar PEG for 24h at 10◦C. the highest AC was attained from concentration of -9 bar PEG for 12h at 15◦C. Also osmo and hydro priming increased catalase (CAT) and ascorbate peroxidase (APX) as compared to the unprimed.

Sadeghi *et al.* (2011) conducted an experiment to evaluate the effect of seed Osmopriming by using PEG6000 priming media on germination behavior and seed vigor of soybean (cultivar 033). Seeds were primed with six levels of Poly ethylene glycol (PEG6000) as priming media (distilled water as control, -0.4, -0.8, -1.2, -1.6 and-2 MPa) for 6, 12, 24 and 48 hours at 25°C. Dry soybean seeds considered as a control treatment (non primed). Results of variance analysis made clear that different osmotic potential and priming duration had significant effect on germination percentage, mean germination time, germination index, and the time to get 50% germination, seed vigor and electrical conductivity of seeds. Also -1.2 MPa osmotic potential increased germination percentages, germination index and seed vigor meanwhile decreased mean germination time, the time to get 50% germination and electrical conductivity of seeds. Also it was observed that 12 h priming duration had most effect on studied traits as-1.2 MPa osmotic potential treatment. Generally primed seeds showed better condition than control treatment in aspect of studied criteria.

Abbasdokht *et al.* (2010) carried out an experiment in order to study of hydropriming and halopriming on germination and early growth stage of wheat (*Triticum aestivum*). Germination and early seedling growth were studied using distilled water (control) and under osmotic potentials of -0.4, -0.8 and -1.2 MPa for NaCl and polyethylene glycol (PEG 6000), respectively. Results showed that Hydroprimed seeds achieved maximum germination seedling dry weight, especially during the higher osmotic potentials. Minimum germination was recorded at untreated seeds (control) followed by osmopriming. Under high osmotic potentials, hydroprimed seeds had higher GI (germination index) as compared to haloprimed or untreated seeds. Interaction effect of seed treatment and osmotic potential significantly affected the seedling vigour index (SVI).

Khalil *et al.* (2010) observed that Phenology and dry matter are important traits being affected by seed priming and soil phosphorus (P_2O_5) application. Priming enhanced days to emergence, anthesis and increased dry matter (DM) production compared with non primed (control). Seed primed with 0.3% P_2O_5 solution took less time to anthesis (110 days). DM yield increased with each increment of priming and maximum DM yield (6051 kg ha-1) was obtained from seeds primed in 0.2% P_2O_5 solution. Water primed seed took less time to emergence (16 days). Soil P_2O_5 application enhanced days to heading, anthesis, maturity and increased DM yield, while days to emergence, spike m-2 and spike length were not affected. Earlier heading, anthesis, maturity and highest DM yield was recorded at 75 kg P_2O_5 ha-1.

Mohammadi and Amiri (2010) reported that for both cultivars germination percentage (GP), mean germination rate (MGR), radicle length (RL), plumule length

(PL) and seedling dry weight (SDW) were reduced when drought stress level were increased from 0 to 1.5 MPa. Although, the cultivars showed different responses to the increased drought stress level. For all of the traits under study, the best results due to the priming treatments were obtained at the drought stress levels higher than -0.6 or -0.9 MPa. So that, at the -1.5 MPa of drought stress level, GP, MGR, RL, PL and SDW were improved 128.62, 200, 223.08, 350 and 69.94%, respectively by KNO_3 and 21.44, 71.43, 219.23, 100 and 41.18%, respectively 3 by distilled water when compared with control. However, seeds primed with KNO_3 showed better performance 3 than those primed with distilled water. In general, this study revealed that seed priming especially with KNO_3 improved canola seed performance under drought stress condition. However, the improvements were more obvious at the higher levels of drought stress.

Yari *et al.* (2010) conducted an experiment in order to evaluate the effect of different seed priming techniques on germination and early growth of two wheat cultivars (Azar-2 and Sardari 101). Seeds were primed for 12, 24 and 36 hours at three temperature (20, 23 and 28°C) in seven priming media. Maximum seed germination percentage in cv.Azar-2 was observed when the seeds primed by PEG 20% for 12h and at 20°C. The most stem length was obtained for seeds osmoprimed with PEG 10% for 24h. Osmoprimed seeds with PEG 20% for 24h produced maximum radicle length of cv. Sardari. Maximum vigor index (VI) of cv. Azar-2 was obtained from seeds primed with KH_2PO_4 0.5% while the lowest germination percent, speed of germination and VI were observed in seeds which subjected to KCl 4% solution. Speed of germination was improved when the seed soaked water and PEG 10%. The most germination percent, VI and speed of germination were observed on 12h. Altogether 20°C treatment had better effects than other temperatures on germination attributes and seedling parameters.

Sun *et al.* (2010) reported that seeds with priming treatments, the contents of proline and SP were significantly higher than those in the corresponding controls, and the contents of SS and MDA were significantly lower than those in the controls. Seed priming accelerated the process of glucose metabolism, enhanced the activities of PAL, SOD, CAT, and POD in the stressed seeds of all cultivars. Compared to hydro-priming, priming with PEG in a proper concentration had a better effect on seed germination and seedling growth under drought stress, and the optimal priming concentrations of PEG were 20% for Gangyou 527 (indica hybrid rice) and 10%–15% for Nongken 57 (conventional japonica rice). Even higher concentrations of PEG had negative effects on seed germination. Moderate priming intensity improved metabolism of rice seed, germination indices, seedling quality, and drought tolerance of seedlings under drought stress for all cultivars. However, such effects had limited capability, and severe drought stress inhibited germination and caused damages of rice seedlings. Rice cultivars had significant impact on priming effect, and indica rice showed better performance than japonica rice.

Patanèa *et al.* (2009) reported that the effects of reduced water potential (ψ) in NaCl and pre-osmopriming in PEG, on seed germination and early radicle growth at different temperatures were assessed in the laboratory for sweet sorghum. Seed priming enhanced germination and shortened the delay in germination time due to the increase in saline stress, at suboptimal temperatures only. This effect could be explained by the faster water absorption occurring in primed seeds as compared to those unprimed, at these temperatures, regardless of salt concentration of the solution. Root growth was more sensitive than germination to salt stress. Indeed, the increase in NaCl concentration adversely affected root elongation at all temperatures. The beneficial effects of PEG-osmopriming were evident on root growth at all temperatures except the optimal one. From a practical point of view, when early sowings of sweet sorghum are requested, the use of primed seeds is suggested, as PEG-osmopriming is helpful in overcoming the negative effect imposed by reduced water potential upon seed germination under suboptimal thermal conditions.

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 hydropriming 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, hydropriming treatment was comparatively superior in laboratory tests. Invigoration of lentil seeds by hydropriming 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, hydropriming 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 hydropriming 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, hydropriming treatment was comparatively superior in the laboratory tests. Invigoration of lentil seeds by hydropriming 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 hydropriming is a simple, low cost and environmentally friendly technique for improving seed and seedling vigor of lentil.

Janmohammadi *et al.* (2008) 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. Hydropriming 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. Hydropriming could alleviate the effects of salinity and drought stress on germination and seedling early growth. This study indicated that hydropriming could be suitable seed invigoration treatment under saline and drought-prone environments.

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 (Triticosecalewitm., cv. Presto) 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 hydropriming is very pronounced particularly in improving germination and seedling growth in low stress.

Farooq *et al.* (2006) conducted an experiment to study the effectiveness of seed priming strategies on the improved agronomic characters of direct-sown rice. Seed priming improved germination and emergence, allometry, kernel yield, and its quality, whilst pre-germination displayed poor and erratic emergence of seedling followed by poor plant performance. Faster and uniform emergence was due to improved α-amylase activity, which increased the level of soluble sugars in the primed kernels. Osmohardening with KCl gave greater kernel and straw yield and harvest index, followed by that of $CaCl_2$, hardening and ascorbate priming. Improved yield was attributed principally to number of fertile tillers and 1000 kernel weight. A positive correlation between mean emergence time and days to heading, while a negative one between kernel yield and harvest index suggested long-term effects of seed priming on plant growth and development. The results suggest that physiological changes produced by osmohardening enhanced the starch hydrolysis and made more sugars available for embryo growth, vigorous seedling production and, later on, improved allometric, kernel yield and quality attributes.

Kayaa *et al.* (2006) reported that the treated seeds (control, KNO_3 and hydropriming) of sunflower (*Helianthus annuus* L.) cultivar Sanbro were evaluated at germination and seedling growth for tolerance to salt (NaCl) and drought conditions induced by PEG-6000 at the same water potentials of 0.0, −0.3, −0.6, −0.9 and −1.2 MPa. Results revealed that germination delayed in both solutions, having variable germination with different priming treatments. Germination, root and shoot length were higher but mean germination time and abnormal germination percentage were lower in NaCl than PEG at the same water potential. Seeds were able to germinate at all concentrations of NaCl but no seed germination was observed at −1.2 MPa of PEG treatments. NaCl had less inhibitor effect on seedling growth than the germination. It was concluded that inhibition of germination at the same water potential of NaCl and PEG resulted from osmotic effect rather than salt toxicity. Hydropriming increased germination and seedling growth under salt and drought stresses.

Sarwar *et al.* (2006) reported that studies were conducted to make possible use of waste saline land by treating the seeds with some simple chemicals. Chickpea seeds of variety CM88 were surface sterilized and soaked in water, mannitol (2% & 4%), K_2HPO_4 (0.5% & 0.8%) and KNO_3 (0.5%, 1.0%, 1.5% and 2.0%) for 24 hours. Treated seeds were sown in pots, where salinity was created with 75ml solution of NaCl, along with three different types of control experiments; non-treated seed in saline soil, non-treated seeds in non saline soil and water treated seed in non-saline soil. It was observed that root length and biomass of roots and shoots were better when treated with water and mannitol. As far as K_2HPO_4 and KNO_3 are concerned, lower concentrations of these chemicals (0.5% K_2HPO_4 and KNO_3) gave comparatively better results than higher concentrations. Chickpea seeds of variety CM98 treated with same chemicals were sown under field conditions, where EC of the soil was 2.99-9.4 at 0-15cm depth, 2.8-10.5 at 15- 30cm depth and pH was 7.7-9.5. Priming with mannitol and water improved plant survival and growth up to maturity under saline stress conditions.

Hu *et al.* (2005) conducted an experiment where a priming method called sand priming was developed using sand as a priming solid matrix. Seeds were mixed with sands that contained 3.8% (v/w) water and sealed in plastic box and then were primed at 18°C for 72 h. The results showed that the energy of germination, germination percentage, germination index and vigor index were improved in four varieties. Meanwhile, seedling height, root length, number of root and root dry weight were significantly higher than the nonprimed controls. Field experiments showed that the seed establishment and yield in sand primed seeds were significantly increased by 19.8% ~ 22.9% and by 9.8% ~ 31.2%, respectively as compared to soaked seeds without priming. It is indicated that sand priming method may help to improve seedling establishment in direct-sown rice and possible to be used in the field crop production.

Li and Zhang (2004) reported that seeds were subjected to different priming treatments such as water, PEG, and salicylic acid (SA) and then the seedlings were subjected to PEG (15%) stress. The different biochemical responses were studied 2 days after treatment. Under PEG stress, the root length was higher in PEG and SA-primed sets as compared to water-primed, while the shoot height had no change. SA-primed increased photosynthetic pigment content and Chla/Chlb of rice under PEG stress. Net photosynthetic rate and water use efficiency were higher in PEG and SA-primed sets as compared to water-primed under PEG stress. SA-primed enhanced the activities of superoxide dismutase and peroxidase, but decreased the activity of catalase under PEG stress. Proline content was lower in PEG-primed, but higher in SA-primed as compared to water-primed treatment. The study thus suggests the use of SA-priming as a more effective strategy to alleviate the PEG induced stress in rice.

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 conditions.

Chapter III

Materials and Methods

CHAPTER III
MATERIALS AND METHODS

This chapter describes the materials and methods that were used in conducting the experiment. It includes a short description of experimental site and duration of the experiment, materials used for the experiment, layout and design, collection of data and statistical analysis presented under the following headings:

3.1. Experimental site and period

The experiment was conducted under the laboratory condition of the department of Agronomy, Sher-e-Bangla Agricultural University, Dhaka from 29 August 2013 to 13 February 2014. Temperature range of the laboratory during the experiment was 26.2°C-33.4 °C and the relative humidity was 56-84%.

3.2. Materials used for the experiment

In this research work, seeds of the Wheat variety BARI Gom 27 and BARI Gom 28 collected from Bangladesh Agricultural Research Institute were used as planting material. Different priming chemicals such as PEG and distilled water were utilized for osmo and hydro priming. Different equipments such as growth chamber, electric balance, Petri dish, filter paper, micro pipette etc. were used for this study.

Characteristics of BARI Gom 27:

- It is a high yielding variety
- It is 2-3 days earlier in heading than Shatabdi
- It is resistant to stem rust and leaf rust
- It is moderately resistant to *Bipolaris* leaf spot
- It is moderately tolerant to terminal heat stress

Characteristics of BARI Gom 28:

- It is 7-8 days earlier in heading than Shatabdi
- It is tolerant to heat stress

3.3. Seed treatment

All seeds were surface sterilized with 2% Safex solution for 5 minutes, then rinsed with sterilized water and air dried at room temperature. After that seeds were used for priming.

3.4. Design and treatment of the experiment

Three different experiments were conducted in Completely Randomized Design (CRD) to achieve the desired objectives. The experiments were as follows:

Experiment 1. Effect of various Polyethylene Glycol (PEG) concentrations on the germination behavior of wheat

The experiment was employed with two factors such as variety (two varieties) and priming (six priming) with six replications.

Factor A: two varieties

V_1: BARI Gom 27

V_2: BARI Gom 28

Factor B: six priming

T_0: Non- primed seeds (Control)

T_w: Seeds primed with water for 12 h

T_5: Seeds primed with 5% PEG for 12 h

T_{10}: Seeds primed with 10% PEG for 12 h

T_{15}: Seeds primed with 15% PEG for 12 h

T_{20}: Seeds primed with 20% PEG for 12 h

All priming media were prepared in distilled water and duration of soaking for hydro and osmopriming were 12 h. After soaking seeds were air dried and placed in Petridish. For each replicate 30 seeds were placed in 12.5 cm Petridish on a layer of filter paper no. 102 moistened with 8 ml of distilled water.

Experiment 2. Optimization of pre-sowing priming time on the germination behavior of BARI Gom 27

This experiment was conducted with 11 treatments with 6 replications. The treatments are as follows:

T_0: Non primed seed (dry seed)

T_1: Seed primed with water for 6 h

T_2: Seed primed with 10% PEG for 6 h

T_3: Seed primed with water for 9h

T_4: Seed primed with 10% PEG for 9 h

T_5: Seed primed with water for 12 h

T_6: Seed primed with 10% PEG for 12 h

T_7: Seed primed with water for 15 h

T_8: Seed primed with 10% PEG for 15 h

T_9: Seed primed with water for 18 h

T_{10}: Seed primed with 10% PEG for 18 h

After priming seeds were air dried and placed in Petridish. For each replicate 30 seeds were placed in Petridish on filter paper moistened with 8 ml of distilled water.

Experiment 3. Effect of different level of drought stress on germination behavior of BARI Gom 27

The experiment was conducted with two factors such as various levels of priming and various levels of drought stress with six replications.

Factor A: levels of priming

T_0: Non primed seed (dry seed)

T_W: Hydro prime for 12 h

T_P: Prime with 10% PEG (osmo prime) for 12 h

Factor B: level of drought stress

P_0: only water

P_5: 5% PEG solution

P_{10}: 10% PEG solution

P_{15}: 15% PEG solution

P_{20}: 20% PEG solution

After seeds were hydro and osmo primed for 12 h, seeds were air dried and placed in Petridishs of different drought stress level. For each replicate 30 seeds were placed in 12.5 cm Petridish on a layer of filter paper no. 102 moistened with 10 ml of respective drought inducing solution and then next day another 10 ml of respective solution.

3.5 Parameter measurement

The data pertaining to following characters were recorded from each Petridish. Data were collected on the following parameters:

3.5.1. Germination percentage

Germination percentage was measured in the eighth day using the formula
GP (%) = (total number of germinated seeds/ total seed) x 100.

3.5.2. Plumule length (mm)

Plumule length was measured in the eighth day using millimeter scale.

3.5.3. Radical length (mm)

Radical length was measured in the eighth day using millimeter scale.

3.5.4. Seedling length (mm)

Seedling length was measured in the eighth day using millimeter scale by summing Plumule length and Radical length.

3.5.5. Vigour index (VI)

Vigour index (VI) was measured by using the formula of Abdul- Baki and Anderson (1970). VI= [GP (%) x seedlings length (mm)]/100.

3.5.6. Germination index (GI)

Germination index (GI) was measured according to the equation of Kader and Jutzi (2004). $GI= \Sigma (Ni /Ti)$. Ni: number of germinated seeds in the day and Ti: number of day after sowing.

3.5.7. Seedling dry weight (g)

Seedling dry weight (g) was measured by weighing the root and shoot after three days of oven drying.

3.6. Statistical analysis

The data collected on various parameters were statistically analyzed using MSTAT C package program to find out the statistical significance of the treatment effect. The mean values of all the treatments were calculated and analyses of variance for all the characters were performed by the F-test (variance ratio). The significance of the difference among the treatment combinations of means was estimated by least significance difference (LSD) at 5% level of probability.

Chapter IV

Results and Discussion

`CHAPTER IV

RESULTS AND DISCUSSION

The experiment was conducted to investigate the effect of seed priming and priming time on germination behavior of wheat in relation to drought tolerance. The data have been presented in Table 1 and Figure 1 to Figure 14 and a summary of the analysis of variance in respect of all the parameters have been shown in Appendix I to Appendix VI. The results of each parameter have been presented and discussed under the following headings

Experiment 1. Effect of various Polyethylene Glycol (PEG) concentrations on the germination behavior of wheat

4.1.1. Effect on germination percentage

Germination percentage of wheat was influenced by various Polyethylene Glycol (PEG) concentrations (Figure 1) and there was completely significant difference between control (non primed seeds) and primed seeds (Appendix I & II). Germination percentage (GP) was affected by hydro priming and different PEG concentrations. Germination percentage (GP) increases with increasing PEG concentration up to 10% then decreases gradually with increasing PEG concentration. The highest GP (95.55%) of BARI Gom 27 was obtained from seeds pre-treated with 10% PEG solution which is 17.43% and 13.95% higher over control and osmoprimed seeds, respectively. The highest GP (75.55%) of BARI Gom 28 was obtained from seeds pre-treated with 10% PEG solution which is 17.64% and 13.23% higher over control and osmoprimed seeds, respectively. Germination percent of BARI Gom 27 was higher compared to BARI Gom 28.

The result of the present study corroborates with the study of previous researchers (Ghassemi-Golezani *et al.*,2008a; Ajouri *et al*., 2004; Maiti *et al.*, 2013; Farooq *et al.*,2006; Sun *et al.*, 2010). 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. Hydropriming significantly improved germination rate (Ghassemi-Golezani *et al.*, 2008a) and is a useful technique for improving overall germination viz. tomato variety germination percentage was increased in response to hydro-priming treatment (Maiti *et al.*, 2013). Farooq *et al.* (2006) suggested that physiological changes produced by osmohardening enhanced the starch hydrolysis and made more sugars available for embryo growth which enhance germination. Sun *et al.* (2010) reported that optimal priming concentrations of PEG were 20% for Gangyou 527 (indica hybrid rice) and 10%–15% for Nongken 57 (conventional japonica rice). Even higher concentrations of PEG had negative effects on seed germination.

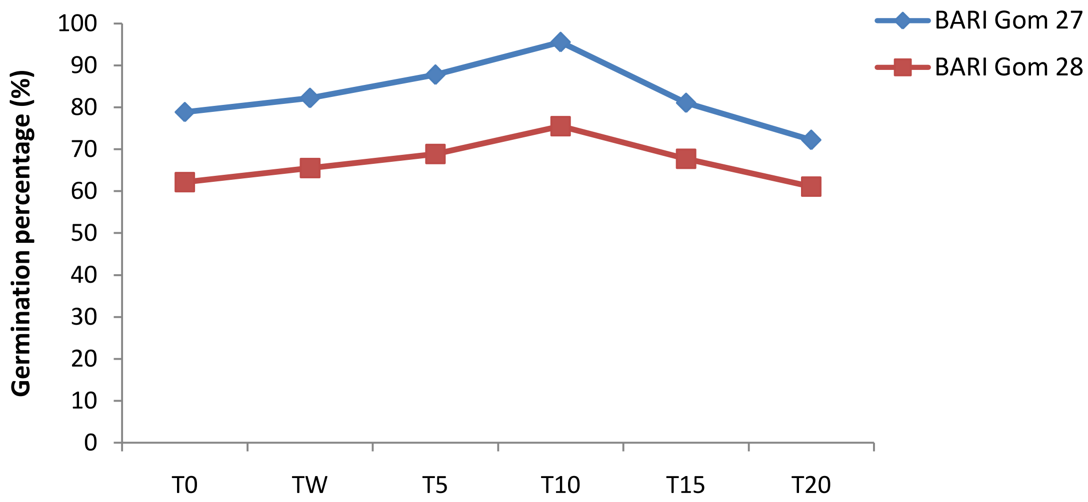

Figure 1. Effect of various Polyethylene Glycol (PEG) concentrations on the germination percentage of wheat (LSD $_{0.05}$ =8.973, 3.443)

Note: T_0: Non- primed seeds (Control), T_w: Seeds primed with water for 12 h,
T_5: Seeds primed with 5% PEG for 12 h, T_{10}: Seeds primed with 10% PEG for 12 h,
T_{15}: Seeds primed with 15% PEG for 12 h, T_{20}: Seeds primed with 20% PEG for 12 h.

4.1.2. Effect on plumule length (mm)

Plumule length (mm) of wheat was influenced by various Polyethylene Glycol (PEG) concentrations (Figure 2) and variance analysis results showed that there was significant difference between control (non primed seeds) and primed seeds (Appendix I & II). Plumule length was affected by hydro priming and different PEG concentration. Plumule length increases with increasing PEG concentration up to 10% then decreases gradually with increasing PEG concentration. The highest plumule length (97.7 mm) of BARI Gom 27 was obtained from seeds pre-treated with 10% PEG solution which is 27.05% and 23.57% higher over control and osmoprimed seeds, respectively. The highest plumule length (83.20 mm) of BARI Gom 28 was obtained from seeds pre-treated with 10% PEG solution which is 28.24% and 21.63%

higher over control and osmoprimed seeds, respectively. Plumule length of BARI Gom 27 was higher compared to BARI Gom 28.

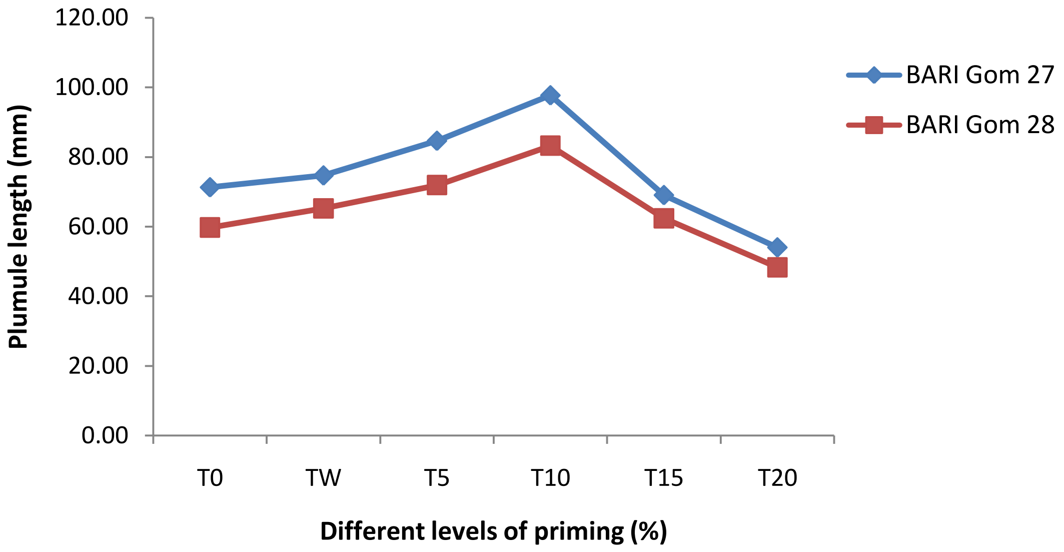

Figure 2. Effect of various Polyethylene Glycol (PEG) concentrations on the plumule length of wheat (LSD $_{0.05}$ = 0.5979, 1.029)

Note: T_0: Non- primed seeds (Control), T_w: Seeds primed with water for 12 h,
T_5: Seeds primed with 5% PEG for 12 h, T_{10}: Seeds primed with 10% PEG for 12 h,
T_{15}: Seeds primed with 15% PEG for 12 h, T_{20}: Seeds primed with 20% PEG for 12 h.

4.1.3. Effect on radical length (mm)

Radical length (mm) of wheat was influenced by various Polyethylene Glycol (PEG) concentrations (Figure 3) and variance analysis results showed that there was significant difference between control (non primed seeds) and primed seeds (Appendix I & II). Radical length was affected by hydro priming and different PEG concentrations. Radical length increases with increasing PEG concentration up to 10% then decreases gradually with increasing PEG concentration. The highest radical length (112.7 mm) of BARI Gom 27 was obtained from seeds pre-treated with 10% PEG solution which is 30.19% and 25.46% higher over control and osmoprimed seeds, respectively. The highest radical length (87.17 mm) of BARI Gom 28 was obtained from seeds pre-treated with 10% PEG solution which is 20.38% and 12.89% higher over control and osmoprimed seeds, respectively. Radical length of BARI Gom 27 was higher compared to BARI Gom 28.

This trend of the present results agrees to the study of previous researcher Sarwar *et al*. (2006) who reported that root length were better when treated with water and mannitol over control.

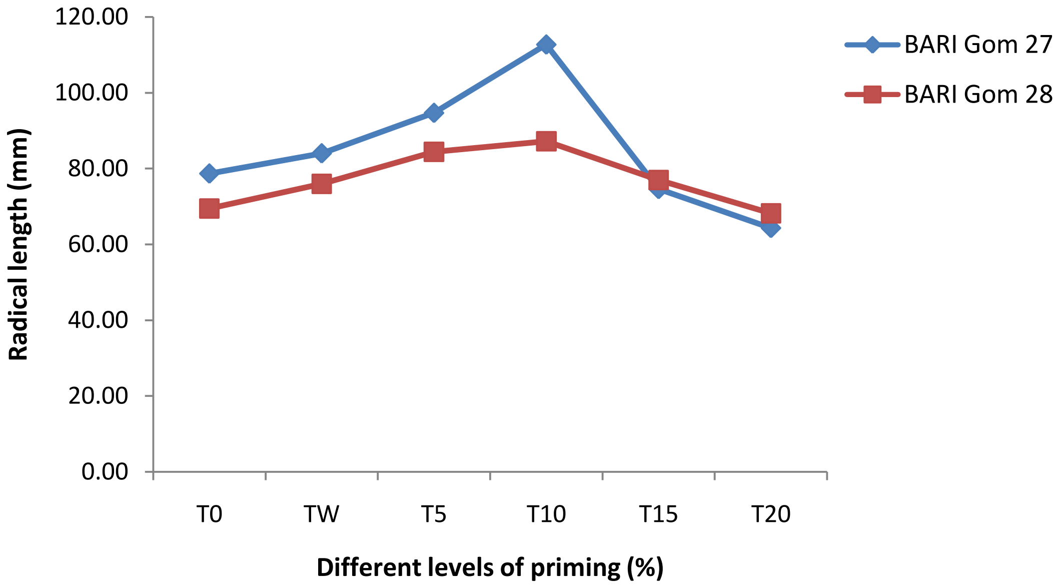

Figure 3. Effect of various Polyethylene Glycol (PEG) concentrations on the radical length of wheat (LSD $_{0.05}$ = 1.454, 0.7846)

Note: T_0: Non- primed seeds (Control), T_w: Seeds primed with water for 12 h, T_5: Seeds primed with 5% PEG for 12 h, T_{10}: Seeds primed with 10% PEG for 12 h, T_{15}: Seeds primed with 15% PEG for 12 h, T_{20}: Seeds primed with 20% PEG for 12 h.

4.1.4. Effect on seedling length (mm)

Seedling length (mm) of wheat was influenced by various Polyethylene Glycol (PEG) concentrations (Figure 4) and variance analysis results showed that there was significant difference between control (non primed seeds) and primed seeds (Appendix I & II). Seedling length was affected by hydro priming and different PEG concentrations. Seedling length increases with increasing PEG concentration up to 10% then decreases gradually with increasing PEG concentration. The highest seedling length (210.4 mm) of BARI Gom 27 was obtained from seeds pre-treated with 10% PEG solution which is 29.03% and 24.57% higher over control and osmoprimed seeds, respectively. The highest seedling length (170.4 mm) of BARI Gom 28 was obtained from seeds pre-treated with 10% PEG solution which is 24.23% and 17.19% higher over control and osmoprimed seeds, respectively. Seedling length of BARI Gom 27 was higher compared to BARI Gom 28.

This result is in agreement with the findings of several workers (Jisha *et al.*, 2013; Maiti *et al.*, 2013). Jisha *et al.* (2013) reported that overall growth of plants was enhanced due to the seed-priming treatments. Maiti *et al.* (2013) observed that seed priming increases seedling vigour of several vegetable crops. The priming techniques improved seedling growth of tomato and chilli although varieties showed variation in response to different treatments.

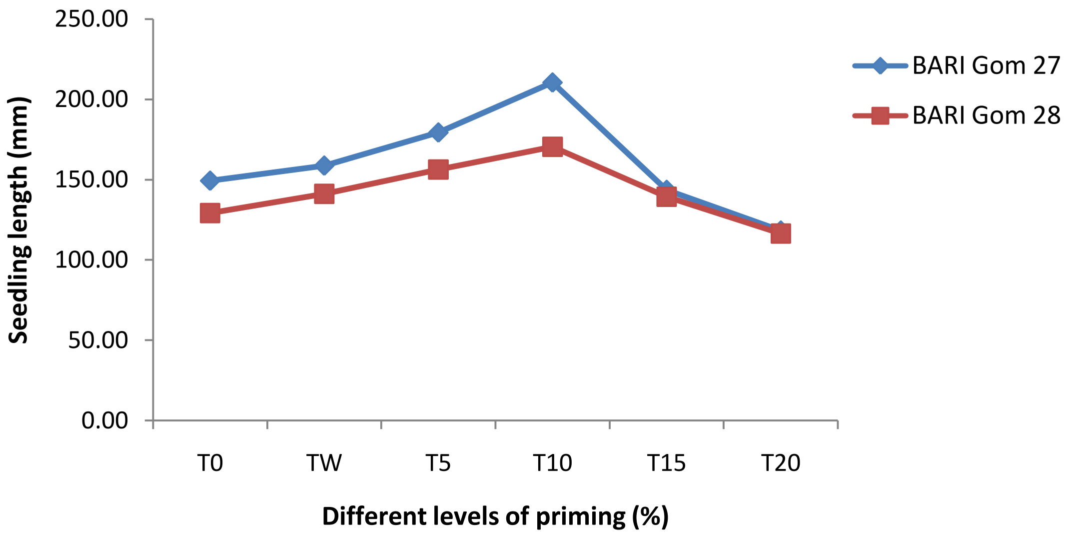

Figure 4. Effect of various Polyethylene Glycol (PEG) concentrations on the seedling length of wheat (LSD $_{0.05}$ = 1.926, 1.422)

Note: T_0: Non- primed seeds (Control), T_w: Seeds primed with water for 12 h,
T_5: Seeds primed with 5% PEG for 12 h, T_{10}: Seeds primed with 10% PEG for 12 h,
T_{15}: Seeds primed with 15% PEG for 12 h, T_{20}: Seeds primed with 20% PEG for 12 h.

4.1.5. Effect on vigour index (VI)

Vigour index (VI) of wheat was influenced by various Polyethylene Glycol (PEG) concentrations (Figure 5) and variance analysis results showed that there was significant difference between control (non primed seeds) and primed seeds (Appendix I & II). Vigour index was affected by hydro priming and different PEG concentrations. Vigour index increases with increasing PEG concentration up to 10% then decreases gradually with increasing PEG concentration. The highest vigour index (201) was obtained from seeds of BARI Gom 27 pre-treated with 10% PEG solution compared to BARI Gom 28 (128.71). The highest vigour index of BARI Gom 27 is 41.44% and 37.07% and of BARI Gom 28 is 37.59% and 28.16% higher over control and osmoprimed seeds, respectively.

This result is consistent with many scientists (Maiti *et al.*, 2013; Maiti *et al.*, 2011). Seed priming increases seedling vigour of several vegetable crops (Maiti *et al.*, 2013) and with respect to sponge gourd, osmo-priming increased seedling vigor (Maiti *et al.*, 2011).

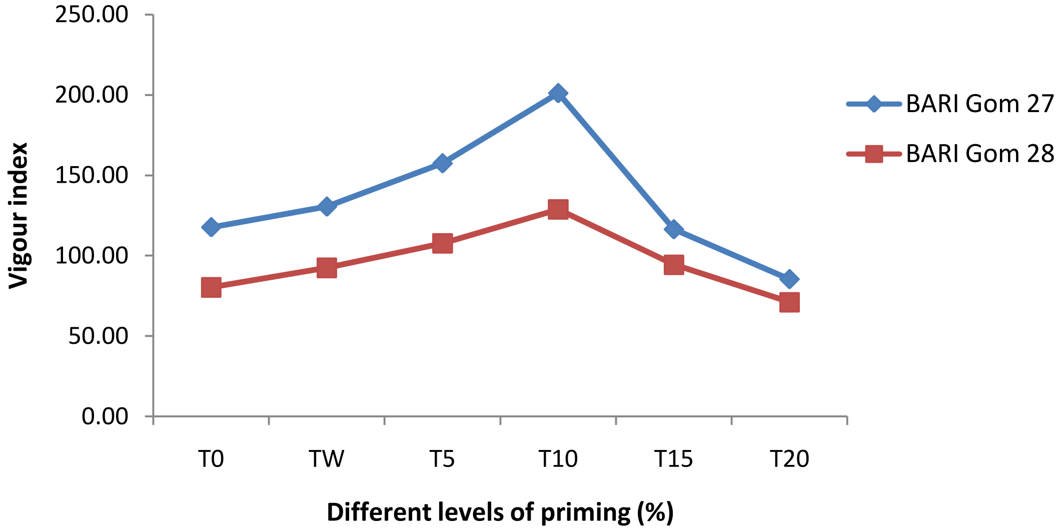

Figure 5. Effect of various Polyethylene Glycol (PEG) concentrations on the vigour index of wheat (LSD $_{0.05}$ = 12.88, 5.07)

Note: T_0: Non- primed seeds (Control), T_w: Seeds primed with water for 12 h, T_5: Seeds primed with 5% PEG for 12 h, T_{10}: Seeds primed with 10% PEG for 12 h, T_{15}: Seeds primed with 15% PEG for 12 h, T_{20}: Seeds primed with 20% PEG for 12 h.

4.1.6. Effect on germination index (GI)

Germination index (GI) of wheat was influenced by various Polyethylene Glycol (PEG) concentrations (Figure 6) and variance analysis results showed that there was significant difference between control (non primed seeds) and primed seeds (Appendix I & II). Germination index was affected by hydro priming and different PEG concentration. Germination index increases with increasing PEG concentration up to 10% then decreases gradually with increasing PEG concentrations. The highest germination index (47.73) was obtained from seeds of BARI Gom 27 pre-treated with 10% PEG solution compared to BARI Gom 28 (27.12). The highest germination index of BARI Gom 27 is 38.63% and 21.12% and of BARI Gom 28 is 17.95% and 14.42% higher over control and osmoprimed seeds, respectively.

This result is in agreement with the findings of Huns and Sung (1997) who reported that seed priming resulted anti-oxidant increment as glutathione and ascorbate in seed. These enzymes make more germination speed via reduction of lipid proxidation activity.

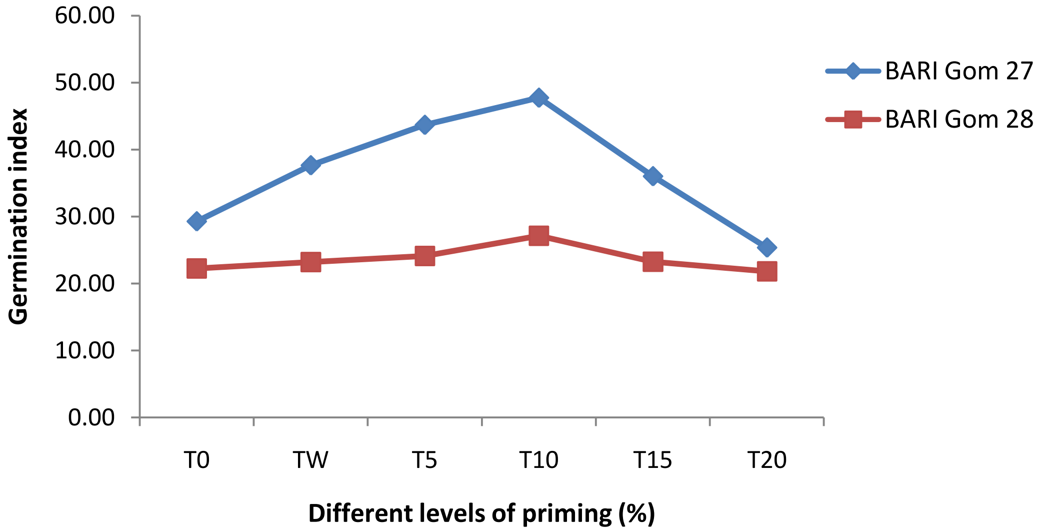

Figure 6. Effect of various Polyethylene Glycol (PEG) concentrations on the germination index of wheat (LSD $_{0.05}$ = 3.182, 1.163)

Note: T_0: Non- primed seeds (Control), T_w: Seeds primed with water for 12 h,
T_5: Seeds primed with 5% PEG for 12 h, T_{10}: Seeds primed with 10% PEG for 12 h,
T_{15}: Seeds primed with 15% PEG for 12 h, T_{20}: Seeds primed with 20% PEG for 12 h.

4.1.7. Effect on seedling dry weight (g)

Dry weight (g) of wheat was influenced by various Polyethylene Glycol (PEG) concentrations (Figure 7) and variance analysis results showed that there was no significant difference between control (non primed seeds) and primed seeds (Appendix I & II). Seedling dry weight was affected by hydro priming and different PEG concentration. Seedling dry weight increases with increasing PEG concentration up to 10% then decreases gradually with increasing PEG concentration. The highest seedling dry weight (0.105 g) of BARI Gom 27 was obtained from seeds pre-treated with 10% PEG solution which is 38.11% and 23.90% higher over control and osmoprimed seeds, respectively. The highest seedling dry weight (0.088 g) of BARI Gom 28 was obtained from seeds pre-treated with 10% PEG solution which is 43.18% and 31.81% higher over control and osmoprimed seeds, respectively. Seedling dry weight of BARI Gom 27 was higher compared to BARI Gom 28.

The result of the present study is also supported by the result of previous researchers (Khalil *et al.*, 2010; Ghassemi-Golezani *et al.*, 2008a; Sarwar *et al.*, 2006). Khalil *et al.* (2010) observed that dry matter yield increased with each increment of priming. Ghassemi-Golezani *et al.* (2008a) showed that hydropriming significantly improved root weights and Sarwar *et al.* (2006) reported that root length and biomass of roots and shoots were better when treated with water and mannitol.

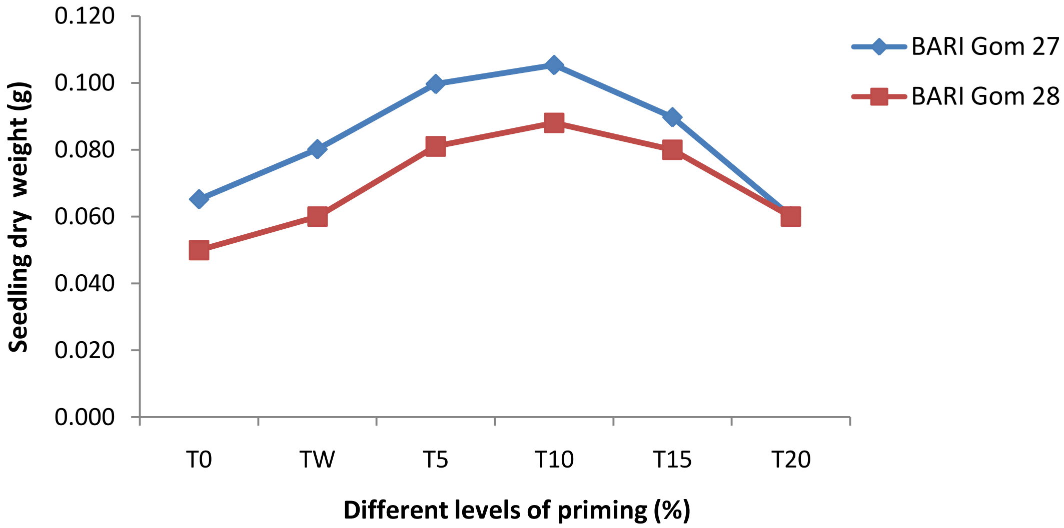

Figure 7. Effect of various Polyethylene Glycol (PEG) concentrations on the seedling dry weight of wheat (LSD $_{0.05}$ = 0.05753, 0.05131)

Note: T_0: Non- primed seeds (Control), T_w: Seeds primed with water for 12 h, T_5: Seeds primed with 5% PEG for 12 h, T_{10}: Seeds primed with 10% PEG for 12 h, T_{15}: Seeds primed with 15% PEG for 12 h, T_{20}: Seeds primed with 20% PEG for 12 h.

Experiment 2. Optimization of pre-sowing priming time on the germination behavior of BARI Gom 27

4.2.1. Effect on germination percentage

Germination percentage of wheat was influenced by different priming time (Table 1) and there was significant difference between control (non primed seeds) and primed seeds (Appendix III). Germination percentage (GP) increases with increasing priming time up to 12 h then decreases gradually. The highest GP (87.77%) was obtained from T_5 treatment, seeds pre-treated with 10% PEG solution for 12 h, which is statistically similar with treatment T_6 (84.44%), seeds pre-treated with water for 12 h. The GP given by T_5 treatment is 34.18 % higher over control.

The result of the present study corroborates with the study of previous researchers (Yari *et al.*, 2010; Sadeghi *et al.*, 2011; Dastanpoor *et al.*, 2013; Moradi *et al.*, 2012; Ajirloo *et al.*, 2013 and Ahammad *et al.*, 2014). Yari *et al.* (2010) reported that maximum seed germination percentage in cv.Azar-2 was observed when the seeds primed by PEG 20% for 12h. Priming duration had significant effect on germination percentage, mean germination time. It was observed that 12 h priming duration had most effect on these traits (Sadeghi *et al.*, 2011). Hydropriming (12 h at 30°C) was most effective in improving seed germination of *Salvia officinalis* L. that final germination percentage (FGP) was increased by 25.5% as compared to that of non-primed seeds (Dastanpoor *et al.,* 2013). Moradi *et al.* (2012) reported that lower priming duration (ie., 12 and 24 h) improved germination under normal condition, while a higher priming duration (ie., 36 and 48 h) provided more protection when the seeds were exposed to drought stress. Increasing of seed soaking duration improved rate of germination (Ajirloo *et al.*, 2013 and Rahman *et al.*, 2014).

4.2.2. Effect on plumule length (mm)

Plumule length (mm) of wheat was influenced by different priming time (Table 1) and variance analysis results showed that there was significant difference between control (non primed seeds) and primed seeds (Appendix III). Plumule length increases with increasing priming time up to 12 h then decreases gradually. The highest plumule length (80.2 mm) was obtained from T_5 treatment, seeds pre-treated with 10% PEG solution for 12 h, which is followed by treatment T_6 (77.67 mm), seeds pre-treated with water for 12 h. The plumule length given by T_5 treatment is 20.36% longer over control.

This trend of the present results agrees to the study of previous researchers (Yari *et al.*, 2010; Moghanibashi *et al.*, 2012). Yari *et al.* (2010) reported that most stem length was obtained for seeds osmoprimed with PEG 10% for 24h. The effect of hydropriming for 24 h increased shoot length of seed sunflower as compared with the control (Moghanibashi *et al.*, 2012).

4.2.3. Effect on radical length (mm)

Radical length (mm) of wheat was influenced by different priming time (Table 1) and variance analysis results showed that there was significant difference between control (non primed seeds) and primed seeds (Appendix III). Radical length increases with increasing priming time up to 12 h for osmopriming then decreases gradually. The highest radical length (82.03 mm) was obtained from T_5 treatment, seeds pre-treated with 10% PEG solution for 12 h. Treatment T_5 is followed by treatment T_4 (79.33 mm), seeds pre-treated with water for 12 h which is statistically similar with treatment T_6 (78.07 mm). The radical length with T_5 treatment is 21.58% longer over control.

This result of the study is consistent with many scientists (Yari *et al.*, 2010; Moghanibashi *et al.*, 2012; Ajirloo *et al.*, 2013). Yari *et al.* (2010) reported that osmoprimed seeds with PEG 20% for 24h produced maximum radical length of cv. Sardari. The effect of hydropriming for 24 h increased root length of seed sunflower as compared with the control (Moghanibashi *et al.,* 2012).According to Ajirloo *et al.* (2013), increasing of seed soaking duration improved radical length.

4.2.4. Effect on seedling length (mm)

Seedling length (mm) of wheat was influenced by different priming time (Table 1) and variance analysis results showed that there was significant difference between control (non primed seeds) and primed seeds (Appendix III). Seedling length increases with increasing priming time up to 12 h then decreases gradually. The highest seedling length (162.23 mm) was obtained from T_5 treatment, seeds pre-treated with 10% PEG solution for 12 h. Treatment T_5 is followed by treatment T_6 (155.73 mm), seeds pre-treated with water for 12 h which is statistically similar with treatment W_9 (155.40 mm). The seedling length given by T_5 treatment is 20.97% longer over control.

Some previous researcher indicated the same result of the present study and reported that increasing of seed soaking duration improved seedlings length (Ajirloo *et al.*, 2013).

4.2.5. Effect on vigour index (VI)

Vigour index (VI) of wheat was influenced by different priming time (Table 5). Variance analysis results showed that there was significant difference between control (non primed seeds) and primed seeds (Appendix III). Vigour index increases with increasing priming time up to 12 h then decreases gradually. The highest vigour index (141.31) was obtained from T_5 treatment when seeds pre-treated with 10% PEG solution for 12 h, followed by T_6 (133.83) in which seeds pre-treated with water for 12 h. The vigour index obtained in T_5 treatment is 39.96% higher over control.

This result is in agreement with the findings of several workers (Sadeghi *et al.*, 2011; Yari *et al.*, 2010)). Sadeghi *et al.* (2011) mentioned that priming duration had significant effect on seed vigor and also it was observed that 12 h priming duration had most effect on the studied trait. The highest VI was observed on 12h (Yari *et al.*, 2010).

4.2.6. Effect on germination index (GI)

Germination index (GI) of wheat was influenced by different priming time (Table 1). Variace analysis results showed that there was significant difference between control (non primed seeds) and primed seeds (Appendix III). Germination index increases with increasing priming time up to 12 h then decreases gradually. The highest germination index (41.23) was obtained from T_5 treatment (seeds pre-treated with 10% PEG solution for 12 h) followed by treatment T_6 (36.62) in which seeds pre-treated with water for 12 h which is statistically similar with treatment T_7 (36.18). The germination index obtained with T_5 treatment is 27.84% higher over control.

This result of the study is consistent with many scientists (Yari *et al.*, 2010; Sadeghi *et al.*, 2011; Moghanibashi *et al.*, 2012). Yari *et al.* (2010) reported that speed of germination was improved when the seed soaked in water and PEG 10%. The most speed of germination was observed on 12h. Priming duration had significant effect on germination index and it was observed that 12 h priming duration had significant effect (Sadeghi *et al.*, 2011). Moghanibashi *et al.* (2012) reported that the effect of hydropriming for 24 h increased germination index of seed sunflower as compared with the control.

4.2.7. Effect on seedling dry weight (g)

Seedling dry weight (g) of wheat was influenced by different priming time (Table 1). The results obtained from variance analysis showed that there was significant difference between control (non primed seeds) and primed seeds (Appendix III). Dry weight (g) increases with increasing priming time up to 12 h for osmopriming then decreases gradually. The highest seedling dry weight (0.088 g) was obtained from T_5

treatment (seeds pre-treated with 10% PEG solution for 12 h) followed by treatment T_4 (0.083 g) where seeds pre-treated with water for 9 h which is statistically similar with treatment T_3 (0.081 g). The seedling dry weight obtained by T_5 treatment is 34.09% higher over control.

The result of the present study is also supported by Moghanibashi *et al.* (2012) who reported that the effect of hydropriming for 24 h increased root and shoot weight of seed sunflower as compared with the control.

Table 1. Effect of various level of priming time on the germination behavior of BARI Gom27

Treatments	Germination percentage (%)	Plumule length (mm)	Radical length (mm)	Seedling length (mm)	Vigour index (VI)	Germination index (GI)	Seedling dry weight (g)
T_0	57.77 f	63.87 h	64.33 g	128.20 j	84.85 k	29.75 g	0.058 g
T_1	67.77 e	70.60 g	70.27 f	140.87 g	96.96 h	30.07 fg	0.065 f
T_2	76.66 c	73.33 ef	72.63 e	145.97 f	108.24 g	31.20 ef	0.080 bc
T_3	81.11 b	75.43 cd	75.60 d	151.03 de	116.82 f	32.45 d	0.081 b
T_4	82.22 b	76.07 c	79.33 b	155.40 bc	128.89 c	33.83 c	0.083 b
T_5	87.77 a	80.20 a	82.03 a	162.23 a	141.31 a	41.23 a	0.088 a
T_6	84.44 ab	77.67 b	78.07 bc	155.73 b	133.83 b	36.62 b	0.078 bcd
T_7	83.33 b	76.10 c	77.00 cd	153.10 cd	126.23 d	36.18 b	0.075 cde
T_8	82.22 b	74.40 de	75.61 d	150.01 e	121.00 e	33.77 c	0.073 de
T_9	72.22 d	72.60 f	65.40 g	138.00 h	90.66 i	31.65 de	0.073 e
T_{10}	64.44 e	69.40 g	64.13 g	133.53 i	88.28 j	29.01 g	0.072 e
LSD	3.55	1.365	1.823	2.463	1.215	1.139	0.005386
CV (%)	2.73%	1.09%	1.46%	0.99%	0.63%	2.01%	1.60%

In column, figures followed by different letter(s) indicate significantly different at 5% level of significance.

Note: T_0: Non primed seed (dry seed), T_1: Seed primed with water for 6 h, T_2: Seed primed with 10% PEG for 6 h, T_3: Seed primed with water for 9h, T_4: Seed primed with 10% PEG for 9 h, T_5: Seed primed with water for 12 h, T_6: Seed primed with 10% PEG for 12 h, T_7: Seed primed with water for 15 h, T_8: Seed primed with 10% PEG for 15 h, T_9: Seed primed with water for 18 h, T_{10}: Seed primed with 10% PEG for 18 h

Experiment 3. Effect of different levels of drought stress on germination behaviour of BARI Gom 27

4.3.1. Effect on germination percentage

Germination percentage of primed (hydroprimed and osmoprimed) and non primed wheat seed was influenced by different levels of drought stress (Figure 8). There was a significant difference between well watered and different level of drought stress levels (Appendix IV, V & VI). Germination percentage (GP) of osmoprimed and hydroprimed seeds was higher compared to nonprimed seeds at well watered and various levels of stress whereas osmoprimed seed gave best result. Germination percentage (GP) of nonprimed and hydroprimed seeds decreases drastically as drought stress increases but osmoprimed seeds showed tolerance capacity upto stress level induced by 10% PEG then significantly decreased with increasing drought stress. The highest GP of osmo, hydro and non primed seeds were 85.55%, 84.44% and 65.55%, respectively in well watered condition. Whereas in drought stress level created by 10% PEG, GP of osmo, hydro and non primed seeds were 4.05%, 52.01% and 118.52% lower in respect of well watered condition, respectively.

Similar results were also represented by several researchers (Yağmur and Kaydan, 2008; Abbasdokht *et al.*, 2010; Janmohammadi *et al.*, 2008; Zamirifar and Bakhtiari, 2014; Moghanibashi *et al.*, 2012; Moghanibashi *et al.*, 2013; Rouhi *et al.*, 2015; Razaji *et al.*, 2014; Ghiyasi and Tajbakhsh, 2013; Sun *et al.*, 2010; Mohammadi and Amiri, 2010; Chen and Arora, 2011). Osmopriming 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). Despite the negative effects of stress condition the priming treatment was effective in improving germination percentage in Presto (Yağmur and Kaydan, 2008) and in *Nigella sativa* (Zamirifar and Bakhtiari, 2014). Hydroprimed seeds achieved maximum germination especially during the higher osmotic potentials compared to untreated seeds (Abbasdokht *et al.*, 2010) and significantly improved germination under both stress and non-stress conditions (Janmohammadi *et al.*, 2008). Rouhi *et al.* (2015) mentioned that germination of Tall Wheatgrass was delayed by drought stress simulated by PEG in primed and nonprimed seeds. Osmopriming increased germination parameters under drought stress rather than hydropriming especially at lowest osmotic potentials. Razaji *et al.* (2014) concluded that priming resulted improvement in germination components and enzymes activity of rapeseed on drought stress condition and boost the resistance of rapeseed to drought stress condition. Compared to hydro-priming, priming with PEG in a proper concentration had a better effect on seed germination under drought stress although such effects had limited capability and severe drought stress inhibited germination (Sun *et al.*, 2010).

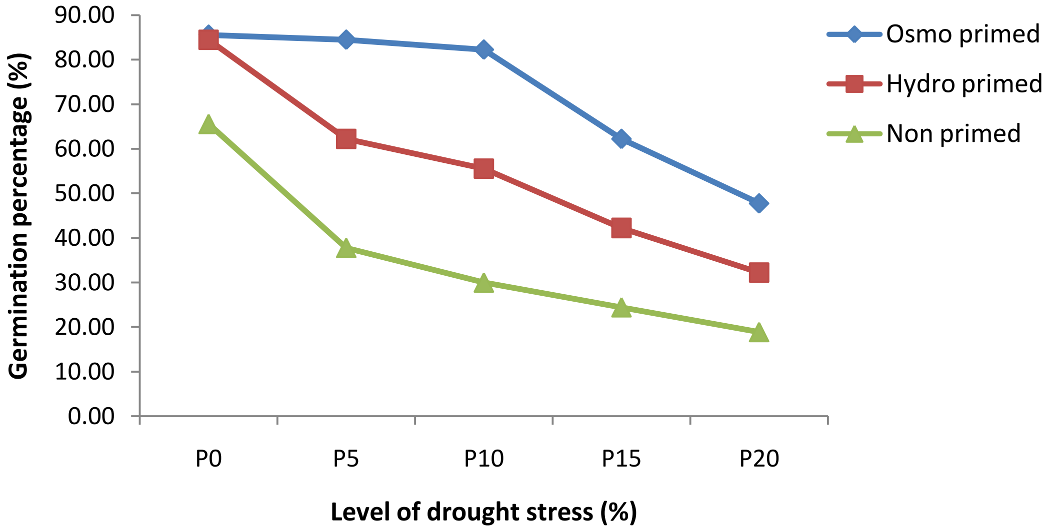

Figure 8. Effect of different levels of drought stress on germination percentage of primed and non primed seeds ($LSD_{0.05}$ = 1.985, 3.797, 4.135)

Note: P_0: only water, P_5: 5% PEG solution, P_{10}: 10% PEG solution, P_{15}: 15% PEG solution, P_{20}: 20% PEG solution

4.3.2. Effect on plumule length (mm)

Plumule length (mm) of primed (hydroprime and osmoprime) and non primed wheat seed was influenced by different levels of drought stress (Figure 9). Significant difference was observed between well watered and different drought stress levels (Appendix IV, V & VI). Plumule length of nonprimed seeds decreases drastically as drought stress increases but osmoprimed and hydroprimed seeds showed tolerance capability upto stress level induced by 10% PEG then significantly decreased with increasing drought stress. The highest plumule length of osmo, hydro and non primed seeds were 94.67 mm, 90.67 mm and 70.33 mm, respectively in well watered condition. Whereas in drought stress level created by 10% PEG, plumule length of osmo, hydro and non primed seeds were 9.23%, 30.14% and 283.63% lower in respect of well watered condition, respectively.

The result of the present study corroborates with the study of previous researchers (Moghanibashi *et al.*, 2013; Mohammadi and Amiri, 2010). Mohammadi and Amiri

(2010) reported that plumule length was reduced when drought stress level was increased. As drought and/or salinity levels increased, shoot length reduces but the priming treatments clearly improved the parameter under drought and salinity conditions so can be used to improve seed performance of sunflower under normal and stress conditions (Moghanibashi *et al.*, 2013).

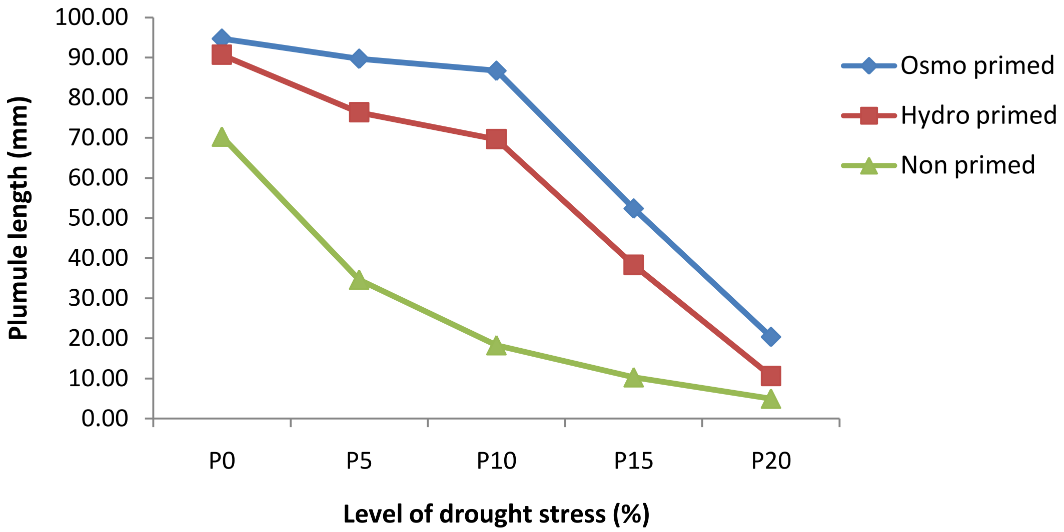

Figure 9. Effect of different levels of drought stress on plumule length of primed and non primed seeds ($LSD_{0.05}$ = 1.191, 1.518, 1.354)

Note: P_0: only water, P_5: 5% PEG solution, P_{10}: 10% PEG solution, P_{15}: 15% PEG solution, P_{20}: 20% PEG solution

4.3.3. Effect on radical length (mm)

Radical length (mm) of primed (hydroprime and osmoprime) and non primed wheat seed was influenced by different levels of drought stress (Figure 10). There was a significant difference was observed between well watered and different drought stress levels (Appendix IV, V & VI). Radical length of osmoprimed and hydroprimed seeds was higher compared to nonprimed seeds at well watered and various levels of stress whereas osmoprimed seed gave best result. Radical length of nonprimed seeds decreases drastically as drought stress increases but osmoprimed seeds showed a considerable tolerance capability upto stress level induced by 10% PEG then significantly decreased with increasing drought stress. The highest radical length of osmo, hydro and non primed seeds were 139.67 mm, 129.67 mm and 81.33 mm, respectively which were obtained from well watered condition. Whereas in drought stress level created by 10% PEG, radical length of osmo, hydro and non primed seeds were 6.35%, 46.24% and 130.19% lower in respect of well watered condition, respectively.

This trend of the present results agrees to the study of previous researchers (Moghanibashi *et al.*, 2013; Mohammadi and Amiri, 2010). Moghanibashi *et al.* (2013) reported that as drought and/or salinity levels increased, root length reduced but the priming treatments clearly improved the parameter of sunflower under normal and stress conditions. Mohammadi and Amiri (2010) mentioned that radicle length was reduced when drought stress level were increased from 0 to 1.5 MPa.

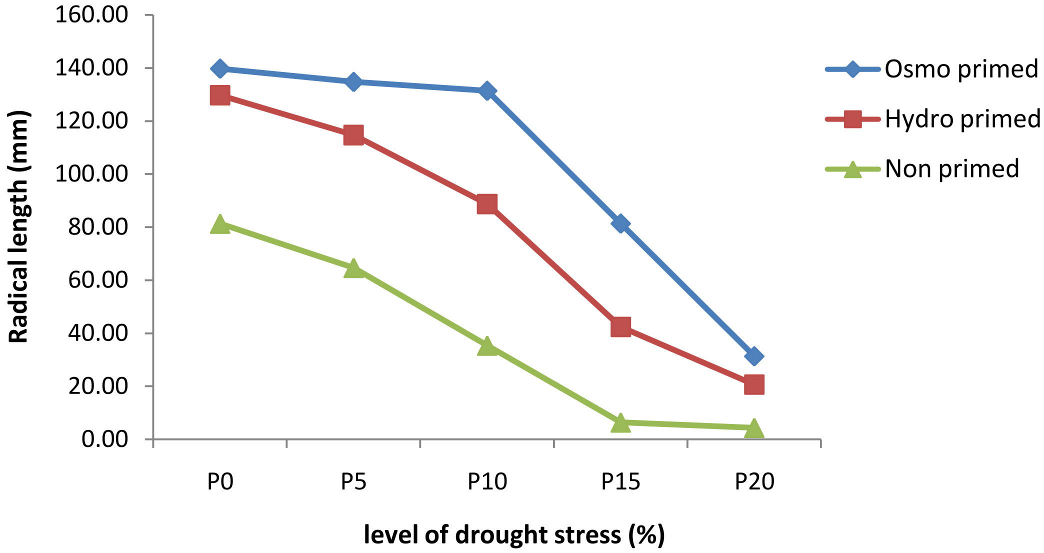

Figure 10. Effect of different levels of drought stress on radical length of primed and non primed seeds (LSD$_{0.05}$ = 1.975, 1.141,1.479)

Note: P_0: only water, P_5: 5% PEG solution, P_{10}: 10% PEG solution, P_{15}: 15% PEG solution, P_{20}: 20% PEG solution

4.3.4. Effect on seedling length (mm)

Seedling length (mm) of primed (hydroprime and osmoprime) and non primed wheat seed was influenced by different levels of drought stress (Figure 11). There was a significant difference observed between well watered and different drought stress levels (Appendix IV, V & VI). Seedling length of nonprimed seeds decreases drastically as drought stress increases but osmoprimed and hydroprimed seeds showed a considerable tolerance capability upto stress level induced by 10% PEG then significantly decreased with increasing drought stress. The highest seedling length of osmo, hydro and non primed seeds were 234.33 mm, 220.33 mm and 151.67 mm, respectively which were obtained from well watered condition. Whereas in drought stress level created by 10% PEG, seedling length of osmo, hydro and non primed seeds were 7.49%, 39.16% and 182.61% lower in respect of well watered condition, respectively.

This result of the study is consistent with many scientists (Yağmur and Kaydan, 2008; Kayaa *et al.*, 2006; Janmohammadi *et al.*, 2008; Ghiyasi and Tajbakhsh, 2013; Razaji *et al.*, 2014; Sun *et al.*, 2010). Hydropriming significantly improved length of seedling under both stress and non-stress conditions (Janmohammadi *et al.*, 2008; Kayaa *et al.*, 2006). Ghiyasi and Tajbakhsh (2013) mentioned that inhibition of seedling growth due to drought stress should be overcome by using PEG 6000 as osmopriming agent in soybean. Razaji *et al.* (2014) observed that increasing in drought stress catalase activity (CAT), peroxidase activity (POX) and Proline content increased as compared to control and concluded that priming resulted improvement in seedling growth and enzymes activity of rapeseed on drought stress condition and boost the resistance of rapeseed to drought stress condition. Sun *et al.* (2010) reported that compared to hydro-priming, priming with PEG in a proper concentration had a better effect on seedling growth under drought stress although such effects had limited capability and severe drought stress inhibited germination and caused damages of rice seedlings

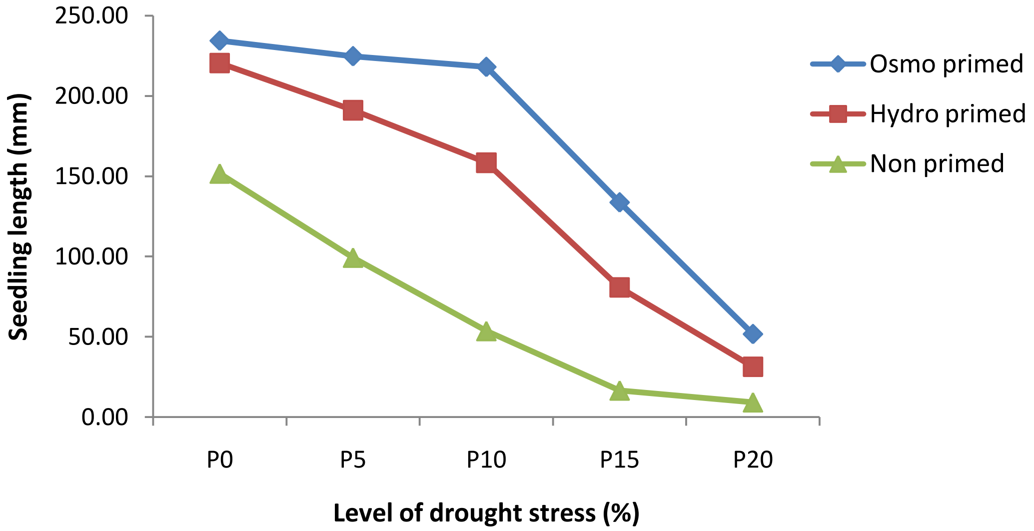

Figure 11. Effect of different levels of drought stress on seedling length of primed and non primed seeds ($LSD_{0.05}$ = 1.899, 1.594, 2.344)

Note: P_0: only water, P_5: 5% PEG solution, P_{10}: 10% PEG solution, P_{15}: 15% PEG solution, P_{20}: 20% PEG solution

4.3.5. Effect on vigour index (VI)

Vigour index (VI) of primed (hydroprime and osmoprime) and non primed wheat seed was influenced by different levels of drought stress (Figure 12). There was a significant difference observed between well watered and different drought stress levels (Appendix IV, V & VI). Vigour index of nonprimed and hydroprimed seeds decreases significantly as drought stress increases but osmoprimed seeds showed a considerable tolerance capability upto certain level. The highest vigour index of osmo, hydro and non primed seeds were 200.47, 186.04 and 99.41, respectively which were obtained from well watered condition. Whereas in drought stress level created by 10% PEG, vigour index of osmo, hydro and non primed seeds were 11.85%, 111.54% and 518.11% lower in respect of well watered condition, respectively.

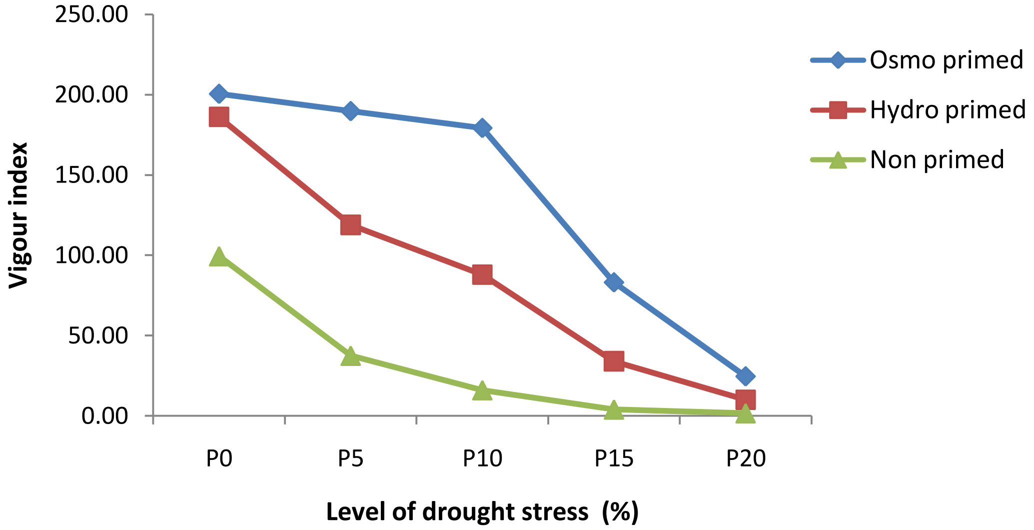

Figure 12. Effect of different levels of drought stress on vigour index of primed and non primed seeds ($LSD_{0.05}$ = 4.73, 4.727, 3.227)

Note: P_0: only water, P_5: 5% PEG solution, P_{10}: 10% PEG solution, P_{15}: 15% PEG solution, P_{20}: 20% PEG solution

This result is in agreement with the findings of several workers (Abbasdokht *et al.*, 2010; Janmohammadi *et al.*, 2008; Moghanibashi *et al.*, 2013; Ghiyasi and Tajbakhsh, 2013). Janmohammadi *et al.* (2008) reported that hydropriming significantly improved seedling vigour index under both stress and non-stress conditions. As drought and/or 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 conditions (Moghanibashi *et al.*, 2013). Ghiyasi and Tajbakhsh (2013) mentioned that inhibition of seedling vigor index due to drought stress should be overcome by using osmopriming treatments in soybean.

4.3.6. Effect on germination index (GI)

Germination index (GI) of primed (hydroprime and osmoprime) and non primed wheat seed was influenced by different levels of drought stress (Figure 13). Significant difference was observed between well watered and different drought stress levels (Appendix IV, V & VI). Germination index of osmoprimed and hydroprimed seeds was higher compared to nonprimed seeds at well watered and various levels of stress whereas osmoprimed seed gave best result. Germination index of nonprimed and hydroprimed seeds decreases drastically as drought stress increases but osmoprimed seeds showed tolerance capability upto stress level induced by 10% PEG then significantly decreased with increasing drought stress. The highest germination index of osmo, hydro and non primed seeds were 35.92, 30.89 and 22.92, respectively which were obtained from well watered condition. Whereas in drought stress level created by 10% PEG, germination index of osmo, hydro and non primed seeds were 12.14%, 47.76% and 96.23% lower in respect of well watered condition, respectively.

The result of the present study is also supported by previous researchers (Abbasdokht *et al.*, 2010; Janmohammadi *et al.*, 2008; Moghanibashi *et al.*, 2012; Moghanibashi *et al.*, Ghiyasi and Tajbakhsh, 2013). Under high osmotic potentials, hydroprimed seeds had higher GI as compared to untreated seeds (Abbasdokht *et al.*, 2010; Janmohammadi *et al.*, 2008). Moghanibashi *et al.* (2012) reported that as salinity and/or drought level increased, all of these parameters reduced under both conditions. Primed seeds produced higher GI under all salinity and drought levels as compared with non-primed seeds. Inhibition of germination index due to drought stress should be overcome by using osmopriming treatments in soybean (Ghiyasi and Tajbakhsh, 2013).

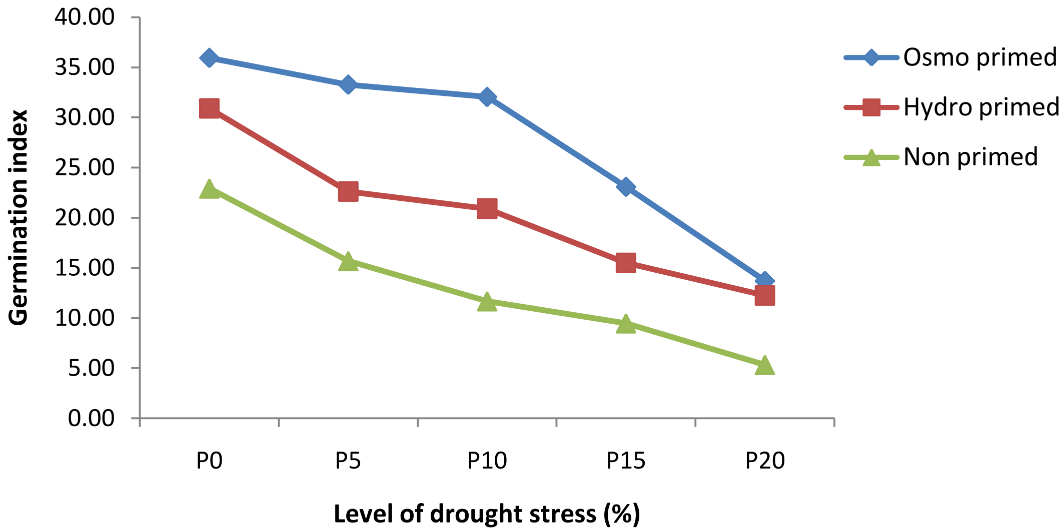

Figure 13. Effect of different levels of drought stress on germination index of primed and non primed seeds ($LSD_{0.05}$ = 1.647, 0.7921, 1.461)

Note: P_0: only water, P_5: 5% PEG solution, P_{10}: 10% PEG solution, P_{15}: 15% PEG solution, P_{20}: 20% PEG solution

4.3.7. Effect on seedling dry weight (g)

Seedling dry weight (g) of primed (hydroprime and osmoprime) and non primed wheat seed was influenced by different levels of drought stress (Figure 14). There was a significant difference observed between well watered and different drought stress levels (Appendix IV, V & VI). Seedling dry weight of osmoprimed and hydroprimed seeds was higher compared to nonprimed seeds at well watered and various levels of stress whereas osmoprimed seed gave best result. Seedling dry weight of nonprimed and hydroprimed seeds decreases drastically as drought stress increases but osmoprimed seeds showed tolerance capability upto stress level induced by 10% PEG then significantly decreased with increasing drought stress. The highest seedling dry weight of osmo, hydro and non primed seeds were 0.140 g, 0.129 g and 0.105 g, respectively which were obtained from well watered condition. Whereas in drought stress level created by 10% PEG, seedling dry weight of osmo, hydro and non primed seeds were 33.50%, 115.19% and 217.10% lower in respect of well watered condition, respectively.

This result of the study is consistent with many scientists (Abbasdokht *et al.*, 2010; Moghanibashi *et al.*, 2013; Ghiyasi and Tajbakhsh, 2013; Mohammadi and Amiri, 2010). Hydroprimed seeds achieved maximum seedling dry weight especially during the higher osmotic potentials compared to untreated seeds (Abbasdokht *et al.*, 2010).

Ghiyasi and Tajbakhsh (2013) mentioned that inhibition of seedling dry weight due to drought stress should be overcome by using PEG 6000 as osmopriming treatments in soybean. Mohammadi and Amiri (2010) reported that seedling dry weight was reduced when drought stress level were increased from 0 to 1.5 MPa.

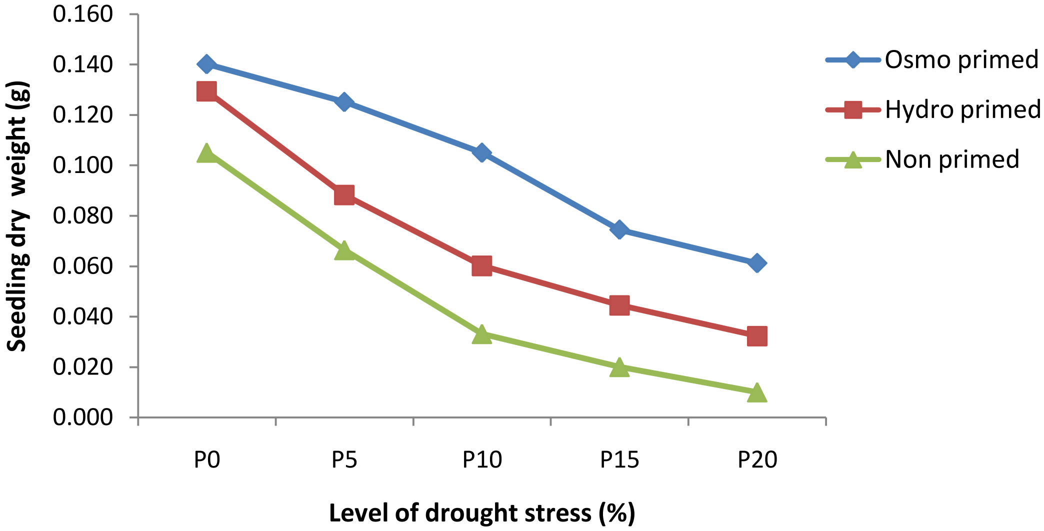

Figure 14. Effect of different levels of drought stress on radical length of primed and non primed seeds ($LSD_{0.05}$ = 0.0188, 0.0179, 0.0172)

Note: P_0: only water, P_5: 5% PEG solution, P_{10}: 10% PEG solution, P_{15}: 15% PEG solution, P_{20}: 20% PEG solution

Chapter V

Summary and Conclusion

CHAPTER V

SUMMARY AND CONCLUSION

A series of experiments 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 study induction of drought tolerance in wheat (*Triticum aestivum* L.) through osmo and hydro priming. The present studies were conducted with three different experiments those laid out in Completely Randomized Design (CRD) with six replications.

In the 1st experiment, wheat seeds of BARI Gom 27 and BARI Gom 28 were pre-soaked in 0, 5, 10, 15 and 20% PEG solution and untreated seeds were served as control. Results revealed that all the characters viz. germination percentage (GP), plumule length, radical length, seedling length, vigor index (VI) and germination index (GI) were significantly influenced by various PEG concentrations except seedling dry weight for both varieties. All the parameters of both varieties gave the best results when seeds treated with 10% PEG solution compared to nonprimed and hydroprimed seeds and decreased gradually with increasing PEG concentration. The highest germination percentage, plumule length, radical length, seedling length, vigor index, germination index and seedling dry weight were obtained from seeds of BARI Gom 27 pre-treated with 10% PEG solution compared to BARI Gom 28.

In the 2nd experiment, BARI Gom 27 was primed in 0 to 18 hours under 10% PEG solution and distilled water. Results revealed that all the characters viz. germination percentage, plumule length, radical length, seedling length, vigor index, germination index and seedling dry weight were significantly influenced by different priming time. All the parameters increased with increasing priming time up to 12 h then decreased gradually. The highest GP, plumule length, seedling length, vigor index and germination index were obtained from seeds pre-treated with 10% PEG solution for 12 h which is followed by seeds pre-treated with water for 12 h. Whereas the highest radical length and seedling dry weight were obtained from seeds pre-treated with 10% PEG solution for 12 h which is followed by seeds pre-treated with water for 9 h.

In the 3rd experiment non primed and primed seeds (osmoprimed and hydroprimed) were germinated under 0, 5, 10, 15 and 20% PEG solution induced drought stress conditions. Results revealed that all the characters viz. germination percentage, plumule length, radical length, seedling length, vigor index, germination index and seedling dry weight were significantly influenced by different stress level. Osmoprimed and hydroprimed seeds gave better result over nonprimed seeds at well watered and at various levels of stress whereas osmoprimed seed gave best result. Results showed that germination behavior and seedling growth of nonprimed and hydroprimed seeds decreased drastically as drought stress increased but osmoprimed seeds showed considerable tolerance capability upto stress level induced by 10% PEG then significantly decreased with increasing drought stress.

Conclusion

Response of wheat varieties was different to pretreatments. Germination behavior and seedling growth of both varieties gave the best results when seeds treated with 10% PEG solution compared to nonprimed and hydroprimed seeds and decreased gradually with increasing PEG concentration. All the parameters gave the best result from seeds pre-treated with 10% PEG solution for 12 h after that decreased gradually. Seeds pre-soaked with 10% PEG and distilled water showed better performance in terms of germination behavior and seedling growth compared to unprimed seed under drought stress. So, wheat seed primed with 10% PEG for 12 h is considered as best priming concentration and priming time to induce drought tolerance capability for enhancing germination behavior and seedling growth of wheat under a certain level of drought stress conditions.

Recommendation

In this study when seeds primed with 10% PEG for 12 h gave the best results in respect of germination behavior and seedling growth of wheat. Under drought stress conditions, it is recommended that seeds be primed with 10% PEG for 12 h. It is suggested that further study should be carried out with different priming agent and field experiments should be done in different location with seeds treated with various priming agents.

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