INDUCTION OF DROUGHT TOLERANCE CAPABILITY OF BARI GOM 30 (*Triticum aestivum* L.) THROUGH UREA AND HYDRO PRIMING

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CERTIFICATE

This is to certify that the thesis entitled 'INDUCTION OF DROUGHT TOLERANCE CAPABILITY OF BARI GOM 30 (Triticum aestivum L.) THROUGH UREA 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 (MS) in AGRONOMY, embodies the result of a piece of bona fide research work carried out by MST. HABIBA SULTANA, Registration Number: 19-10201, under my supervision and guidance. No part of the thesis has been submitted for any other degree or diploma.

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



Dated: Place: Dhaka, Bangladesh Prof. Dr. Md. Abdullahil Baque Department of Agronomy Supervisor

Dedicated to My

Nurturers,

Especially My

Parents

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INDUCTION OF DROUGHT TOLERANCE CAPABILITY OF BARI GOM 30 (*Triticum aestivum* L.) THROUGH UREA AND HYDRO PRIMING

ABSTRACT

The experiment was conducted under the laboratory condition at the Department of Agronomy, Sher-e-Bangla Agricultural University, Dhaka from August 2019 to July 2020 to study the induction of drought tolerance capability in wheat through urea and hydro priming. In the 1st experiment, wheat seeds of BARI Gom 30 were surface sterilized with 75% alcohol for 5 minutes. The seeds were then pre-soaked in 0%, 2%, 4%, 6%, 8% and 10% urea solution and in distilled water for 9 hours and untreated seeds were used as control treatment. The results showed that seed priming influenced the germination percentage, growth parameters (seedling fresh weight, root length, shoot length, shoot dry weight, root dry weight and vigor index) and water relation behaviours (relative water content, water retention capacity and water saturation deficit) of wheat. The highest germination percentage (98.21%), root and shoot length (162.07 mm and 174.90 mm), seedling fresh weight (275.19 mg), root and shoot dry weight (12.27 mg and 16.59 mg), vigour index (330.93), water retention capacity (18.53%) and relative water content (93.15%) were obtained from the seeds treated with 6% urea solution and these values then gradually decreased with increasing urea concentration. The highest water saturation deficit (19.73%) was obtained from control treatment and lowest (6.85%) obtained from seeds treated with 6% urea solution. In 2nd experiment, seeds primed with 6% urea for 9 hours, hydro primed (distilled water for 9 hours) and non-primed seeds were placed under drought stress condition as induced by 0%, 5%, 10%, 15% and 20% PEG solution. Seeds treated with 6% urea solution showed better result compared to hydro primed and control (non-primed) seeds with up to 10% PEG concentrations in all parameters and this value decreased with further increasing concentrations of PEG solution. Hydro priming also gave better result than non-primed seeds in all parameters but not better than 6% urea priming solutions. So, seeds primed with both 6% urea solution and distilled water had the better performance compared to non-primed seeds under drought stress condition. The result thus suggested that seeds primed with 6% urea solution for 9 hours considered as best priming concentration for inducing drought tolerance in wheat seeds for germination, water relation behaviour and all other related growth parameters.

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Acronym		Full meanings
AEZ	=	Agro-Ecological Zone
AOSA	=	Association of Official Seed Analysts
%	=	Percent
⁰ C	=	Degree Celsius
BARI	=	Bangladesh Agricultural Research Institute
BBS	=	Bangladesh Bureau of Statistics
CV%	=	Percentage of coefficient of variance
Cm	=	Centimeter
CRD	=	Completely Randomized Design
et al.	=	And others
FAO	=	Food and Agriculture Organization
GI	=	Germination Index
GP	=	Germination percentage
Intl.	=	International
LSD	=	Least Significant Difference
Mg	=	Milligram
Mm	=	Milimeter
Res.	=	Research
Sci.	=	Science
SAU	=	Sher-e-Bangla Agricultural University
Technol.	=	Technology
VI	=	Vigor Index

CHAPTER I

INTRODUCTION

Wheat (*Triticum aestivum*) is the most important cereal grain crop in the world. It is mostly consumed in the form of bread. It is a grass plant that produces a "head of wheat" with edible seeds. Its origins can be traced back to the Levant, a region of the Near East. It is now grown all over the world. Wheat is one of the most important vegetable protein sources in human food. It has higher protein content than major cereals like maize or rice. Wheat is the second most important human food crop, trailing only maize in terms of total production. Now, worldwide wheat production stands at 770.4 tons, up 3.9 percent from the previous year (FAO, 2021). It is the staple food of more than 36% of the world's population, containing around 55% of the carbohydrates and 20% of the food calories ingested as part of the human diet (Hasanuzzaman *et al.*, 2017). Wheat production covers over 30% of the world's land area (Lobell and Gourdji, 2012).

National wheat production in 2021 was 1.03 M metric tons, a 4.24% decrease from the previous year (USDA, 2021). However, due to growing wheat under rainfed conditions, Bangladesh's wheat yield is very low when compared to other wheat producing countries around the world (Bazzaz, 2013). Wheat production in Bangladesh must be increased to meet the ongoing food crisis. Due to overpopulation, Bangladesh's ability to increase cultivated land is limited. As a result, increasing total production as yield per unit area is the primary way to meeting the food demand.

By 2050, the total world population will be around 9-10 billion, necessitating increased food production to feed this massive population (Waraich *et al.*, 2011). Wheat productivity is suffering as a result of various abiotic and biotic stresses. Global climate change as a result of unpredictable environmental conditions is now a threat to current and future agriculture, causing both biotic and abiotic stresses that reduce plant growth and yield attributes (Hasanuzzaman *et al.*, 2012). There are various abiotic stresses; drought stress takes an important position due to its nature of demolition and losses to crop yields. To meet this challenge, increasing yield potential by reducing various types of biotic and abiotic stresses such as drought is critical (Tuteja *et al.*, 2012). Every year, plants are subjected to a variety of environmental disasters such as drought, salinity, temperature extremes, toxic metals, and so on, resulting in a 50% yield reduction.

The indeterminacy of the global climate, combined with precarious rainfall patterns, is one of the primary causes of the world's rapid onset of drought stress. Inadequate water supply for extended periods has a negative impact on yield and productivity, as well as phenology, growth, and reproduction. Due to decreased water flow from the xylem to the other cells, cell division, elongation, and expansion were inhibited. Plant height, leaf area, stem extension, and root proliferation are all reduced by drought stress. Plants generally adapt to drought stress by inducing a variety of morphological, physiological (osmotic adjustment and cell membrane stability), biochemical (proline, auxins, and ethylene), and molecular mechanisms such as stress-responsive proteins (Basu *et al.*, 2016). In order to prevent plant tolerance to various stresses, a substitute solution must be developed due to the limitations of the current techniques. The alternative solution will be more acceptable if it is simple, affordable, and readily accepted by the farmers. It should also be very feasible and effective in illuminating the tolerance.

Seed priming is thought to be a promising method for improving crop plant's ability to withstand stress including drought. It is the process of treating seeds with natural and artificial compounds prior to germination in order to induce specific physiological behavior in plants. The primed state of the plant is the physiological condition in which plants can better or faster activate defense responses or both (Beckers and Conrath, 2007). Seed priming induces the early emergence of seedlings through the regulation of metabolic processes in the early phases of germination under drought stress (Farooq *et al.*, 2017). It is a simple and affordable method to overcome the negative effects of abiotic stresses (Ibrahim *et al.*, 2016; Wojtyla *et al.*, 2016). Seed priming prevents seeds from absorbing enough water for radical protrusion and retards the seeds in the lag phase (Taylor *et al.*, 1998). Seed priming has been commonly used to minimize the time between seed sowing and seedling emergence and thereby to ensure synchronize emergence (Parera and Cantliffe, 1994). The effects of priming are collaborated with repairing and building up of nucleic acids, increased synthesis of proteins as well as repairing of the membranes (McDonald, 2000).

Seed priming techniques includes hydro priming (soaking in DW), osmo-priming (soaking in osmotic solutions of PEG, manitol, KCl, MN, Urea, NaCl) (Eivazi, 2012; Farooq *et al.*, 2013; Ghiyasi and Tajbakhsh, 2013), hardening with plant growth inducers (CCC, Ethephon, IAA) (Eivazi, 2012) and hormonal priming (Khan *et al.*, 2009). Various priming agents including ascorbic acid, salicylic acid, kinetin, CaCl₂,

abscisic acid (Khan *et al.*, 2012) and urea (Moradi *et al.*, 2012) are frequently reported in literature for chemical priming of seeds. There are reports that osmopriming with urea, in addition to osmotic effects, have nutritional effects on germination and therefore enhance seed germination under stress condition (Mauromicale and Cavallaro, 1996). Al-Mudarsi and Jutzi (1999) observed that the seeds of sorghum and millet primed with urea had higher germination (40%) in compared to non-primed seeds.

Drought stress inhibited the growth and yield of crops, if the plants formerly exposed to one type of stress (stress priming/hardening) may develop tolerance to another kind of stress through promoted production of secondary metabolites (Tabassum *et al.*, 2017; Farooq *et al.*, 2017). Seed osmo priming with urea and PEG in comparison with hydro priming more effectively improved seed germination of wheat in optimum and drought stress conditions. In the optimum conditions, the effect of priming with PEG for most of traits are the same or weaker than that of urea, while this effect is more pronounced under drought conditions and these indices in primed seeds with urea compared to PEG are higher (Moradi *et al.*, 2012).

In Bangladesh, very few information related to seed priming with osmotic priming agenst for inducing drought tolerance in wheat or other crops is rare. Therefore, the present research will be undertaken with the following objectives:

- To evaluate the effect of pre-sowing seed treatment with various concentration of urea (CH₄N₂O) on germination behavior, seedling growth and water relation behavior of wheat in relation to drought stress, and
- ii. To better understanding of the physiological mechanism involved during seed germination, seedling growth and water relation behavior under drought stress condition.

CHAPTER II

REVIEW OF LITERATURE

Wheat is a most important cereal crop in Bangladesh but drought stress has a significant negative impact on wheat production. Under various stress conditions, pre-treating seeds improves germination, seedling growth, and water relation behaviors. The most important method for escaping that unfavorable situation is seed priming. Various authors have studied the available literature on seed priming for various legumes and other crops, as follows:

2.1 Effect on seed priming

Seed priming is a form of seed preparation in which seeds are pre-soaked before planting (Ahammad *et al.*, 2014). Organic and inorganic pre-treatments can apply to cucumber (*Cucumis sativus* L.) seeds and increases seed germination (Isheri *et al.*, 2015). According to Parmoon *et al.* (2013), priming allowed the start of biochemical processes and sugar metabolism, which acted as hydrolysis inhibitors during the first and second stages of germination prior to the emergence of the radical. According to reports, seed priming relieves physiological and pathological stresses and causes the utilization, activation, and enhancement of various cellular defense responses and resistance (Conrath *et al.*, 2002). Under PEG-stimulated drought stress, hydro and osmo priming improved the germination parameter of tall wheat grass (*Agropyron elongatum*) (Rouhi *et al.*, 2015).

Munns and Tester (2008) observed that Poly ethylene glycol (PEG) is most commonly used to induce the osmotic stress in plants and also increases the growth of leaves, stems, leaf area, number of tillers, development of new leaves, lateral buds, branches and root growth (Taiz and Zeiger, 2006). Arruje *et al.* (2013) conducted that seed priming with sodium silicate (SS) enhances seed germination and seedling growth under water-deficit stress induced by polyethylene glycol (PEG). The objective of this study was to evaluate the effects of priming with sodium silicate on improvement of germination percentage and seedling growth of wheat under simulated drought stress. Result indicated that seed priming with different concentrations of sodium silicate and hydro priming increases final germination percentage and improves seedling growth comparing with non-primed seeds under drought stress condition.

Poor wheat stand establishment is a major cause of yield loss, particularly in rainfed and poorly irrigated environments. Seeds of eight wheat cultivars (*Triticum aestivum* L.) including irrigated and rainfed varieties, belonging to three different climates of Iran (Hamedan as cold, Karaj as temperate and Sarpolzohab as warm climates) were primed using osmo primig treatments through use of Urea, NaCl and PEG6000 as well as hydro priming treatments through tap and distilled water. In each climate while employing the related cultivars, a separate experiment was conducted using a split plot factorial in a randomized complete block design of three replications. Main plots were representatives of two sowing dates (timely, and late), while subplots included combinations of cultivar and priming treatments. Results showed that Urea and PEG osmo priming treatments as well as tap water hydro priming treatments caused an increase in emergence rate (both two sowing dates) in all rainfed cultivars of all climates as well as in Alvand irrigated cultivar of cold climate (Hamedan) zone (Aboutalebian *et al.*, 2008).

2.2 Effect on germination related attributes

2.2.1 Total Germination

Seed priming is a pre-sowing seed treatment in water or another osmotic solution that allows the seed to absorb water and aids germination while preventing radical protrusion through the seed coat. Hydro priming and halo priming are the most popular and functional priming treatments. Seed priming is a simple, low-cost, and effective process that also involves controlled hydration followed by re-drying and is very effective in reducing salinity effects in many crops (Wahid *et al.*, 2007; Afzal *et al.*, 2011). Seed priming improved the germination percentage and seedling growth by reducing emergence time and increased yields in field crops including rice (Farooq *et al.*, 2006; Afzal *et al.*, 2006; Afzal *et al.*, 2011). Sun *et al.* (2010) reported that PEG priming with moderate concentration showed higher tolerance to drought stress than hydro-priming, though higher concentrations of PEG had negative effects on seed germination. Baque *et al.* (2016) concluded that seed priming with Polyethylene Glycol (PEG) helps to increase germination behavior on wheat seed.

Seed priming induces crop growth and higher yields in a range of crops. Harris *et al.* (2007) reported that seed priming helps to better establishment and growth, earlier flowering, increase seed tolerance to adverse environment and greater yield in maize.

Seed priming has a beneficial effect in many field crops such as wheat, sweet corn, mungbean, barley, lentil, cucumber etc. (Sadeghian and Yavari, 2004). Roy and Srivastava (1999) concluded that soaking wheat kernels in water enhanced their germination rate under saline conditions. Seed priming increased on field emergence its rate and early seedling growth of maize crop and improved the field stand and plant growth at vegetative and maturity of maize (Nagar *et al.*, 1998). Afzal *et al.* (2006) reported that wheat cultivar treated with different priming agents i.e. Abscisic acid (ABA), Salicylic acid (SA) and ascorbic acid and were sown under normal and saline condition (15 dSm⁻¹), and under saline conditions these treatment reduced the time for 50% germination, increased final germination count, and significantly increased the fresh and dry weight but ascorbic acid did not gives such result.

The effect of priming sorghum and pearl millet seeds with fertilizers was investigated. Two experiments were conducted on M35-1 (sorghum) and ICMH 356 (pearl millet), and ICSV 745 (sorghum) and Barmer (pearl millet) seeds, respectively. Treatments included nitrogen, phosphorous and potassium-based treatments which showed that 7.5 g l^{-1} of urea substantially enhanced the final germination percentage of seeds. The second experiment included 10 priming treatments with 7.5 g l^{-1} urea mixed with other fertilizers. Results indicated that soaking seeds for 3 days in urea + one of several fertilizers including N, P or K-based nutrient compositions or micro elements significantly increased germination percentage and speed but did not affect seedling growth at 15 or 60 days after sowing. It is concluded that seed priming treatments with fertilizers may serve as an appropriate treatment for advancing germination of the species studied (Al-Mudaris and Jutzi, 2001).

Seed priming is used to increase the germination percentage and seed vigor. Primed seeds have great potential to grow under stressful conditions. It has strong resistance against disease and insect attack. Primed seeds have much growth potential and give more production as compared to non-primed seeds. It showed that more yield and uniformity as compare to non-primed seeds. Seed germination process occurs in the three phases. First phase in which seed uptake the water rapidly is called the imbibitional phase, second phase which just change in the water content and third or last phase is radical emergence. Primed seeds completed first two phases during priming process so immediately germinate after sowing (Aymen and Hannachi, 2012).

Seed priming is a method to improve germination and seedling establishment under stress conditions. The effect of seed priming in chemical solutions such as urea and KNO₃, on protein and proline content, germination, and seedling growth responses of four maize (Zea mays L.) hybrids under drought and salt stress conditions was studied in a controlled environment in 2010. Treatments included stress type and intensity at five levels: moderate drought (MD), severe drought (SD), moderate salt (MS), severe salt (SS), and control (C1, without stress), three seed priming types including water (C2, as control), KNO₃, and urea (as chemical priming), and four maize hybrids including Maxima, SC704, Zola, and 307. The results showed that the highest germination percentage (Ger %), germination rate (GR), seedling length (SL), radical length (RL), and seedling to radical length ratio (S/R) were achieved in no stress treatments and most proline content in SD treatment. Urea priming led to more Ger%, GR, and SL compared to other primers and treatment under KNO₃ priming resulted in higher RL compared to other primers. Chemical priming had no effect on S/R and proline content. Also, in terms of most traits, no difference was found among the four hybrids. Results showed that salt stress could affect GR and RL more than the drought stress. Drought stress affected germination percentage and S/R more than the salt stress. Both stresses decreased all measured parameters, except protein and proline content which were increased remarkably, and more under drought compared to salt stress. Based on proline content, hybrid 304 appeared to be more resistant to stress than other hybrids. Generally, KNO₃ and urea alleviated effects of both stresses and led to increased germination and seedling growth as well as the root length. Therefore, priming could be recommended for enhancing maize growth responses under stressful conditions (Anosheh et al., 2011).

NaCl and Urea osmo priming increased flowering speed in first sowing date of Azar2 (rainfed cultivar of Hamedan) and Chenab (rainfed cultivar of Sarpolzohab) respectively. In rainfed cultivars of cold climate zones, PEG and Urea osmo priming significantly increased germination percentage and leaf area index. Number of spikes per unit area was the most important factor among yield components that was significantly increased by osmo priming treatments in Hamedan cultivars (both irrigated and rainfed cultivars) as well as in Karaj rainfed cultivar and for both sowing dates. This resulted in an increase in yield. Meanwhile harvest index was not affected by priming treatments. It seems that the desired effects of osmo priming would be on

the increase in the presence of such environmental growth limiting factors as low temperatures and water deficit (Aboutalebian *et al.*, 2008).

According to Basra *et al.* (2003) germination percent, seedling emergence and seedling establishment improved by using seed priming techniques. In fact priming enhances a range of biochemical changes in the seed which increases the germination process i.e., breaking of dormancy, hydrolysis or metabolism of inhibitors, imbibition and enzymes activation (Ajouri *et al.*, 2004). Arrjue *et al.*, (2013) set an experiment on seed germination and seedling growth of wheat seed primed under water deficit conditions. They found that the germination percentage (GP) (normal seedling percentage (NSP), germination index and seedling length were more in osmo priming conditions than non-primed seeds. They also found that seed priming treatments not only improved the seed germination but also enhanced the wheat seedling growth under water-deficit stress induced by PEG. Beneficial effects on seed germination and seedling vigor indicated an improvement in water deficit stress tolerance of treated seeds.

According to Tabassum *et al.* (2017), osmo priming the seed from a terminally drought stressed source increased the number of grains per spike (14%), 100-grain weight (32%), and grain yield (21%). Osmo priming the seed from a well-watered crop increased the number of productive tillers (21%) and the harvest index (10%) compared to the non-primed control. Faijunnahar *et al.* (2017) conducted an experiment and found that 10% PEG is sufficient to improve wheat genotype germination, seedling growth, and water relation behavior.

Koirala *et al.* (2019) conducted an experiment from December 2017 to May 2018 to depict reliable and effective technique of seed priming in spring rice variety. The laboratory analysis was performed in the Regional Seed Testing Laboratory, Sundarpur, Kanchanpur under controlled conditions. The completely randomized design with 6 treatments- without priming, water/ hydro-priming, 2% urea priming, 2% DAP priming, 2% MOP priming and 2% ZnSO4 priming with 4 replications was used. Two different experimental setups were made for germination and emergence test using germination paper roll and sand tray method. Meanwhile, the parameters like germination percentage, germination energy, vigor index, root length and shoot length at 7 DAS and emergence percentage were remarkably affected. However, the parameters like the speed of germination and shoot length, fresh weight and dry weight

at 10th, 15th and 20th DAS were not affected significantly by the priming treatments. The priming treatment with 2% MoP solution gave the best results for germination percentage (93.5%), germination energy (92.5%), vigor index (138.1) and emergence percentage (93.0%). 2% urea priming treatment resulted in longest root length (12.04 cm) and shoot length (7.97 cm) at 7 DAS. This study suggests seed priming with 2% MOP solution as an easy and effective technique for improving the germination, emergence and seedling parameters of spring rice, variety Hardinath-1.

Salehzade *et al.* (2009) conducted a study to enhance germination and seedling growth of wheat seeds using osmo priming treatments where seeds were osmo primed with PEG-8000 solution for 12 hours. By doing these treatments the seedling stand establishment parameters improved.

2.2.2 Mean germination time

The mean germination time (MGT) is a measure of the rate and spread of germination. MGT is used to compare specific pairs or groups of means and to assess seed vigor. Accelerated germination of primed seeds may be due to an increased rate of cell division (Bose and Mishra, 1992). Priming treatment reduces planting time and seed emergence while also protecting seeds from biotic and abiotic stress during the critical phase of seedling establishment. According to Basra et al. (2005), priming reduced mean germination time compared to non-primed seed. MGT is affected by imbibition duration and internal metabolic activities after imbibition. Harris et al. (1999) reported that early emergence and maturity in seed priming treatment could be due to advancement in metabolic state. Seed priming improves plant stand and provide benefits in term of maturity was concluded by Musa et al. (1999). Seed priming increased earlier emergence of seedlings by 1-3 days and also increased plant stand and initial growth vigor. Priming of seeds increased 47% grain yield advantage and showing positive effects of priming. Seed priming treatment influenced the MGT compared with control seeds at all of the germination temperatures (Yucel, 2012). In generally, seeds primed for 24hrs reduced hours required reaching 50% germination compared with the seeds primed for 12hrs. Kumar et al. (2002) conducted an experiment and reported that priming of finger millet seeds for 8hrs in water resulted in an increased mean plant height by 9 cm, reduced mean time to 50 per cent flowering and maturity by about 6 days and finally increased grain yield. Narayanareddy and Biradarpatil (2012)

conducted experiment to study the influence of seed priming on sunflower. The seeds treated with polyethylene glycol (PEG) with CaCl₂ and water hydrations treatments recorded significantly higher germination (71.30%), speed of germination (31.56), root length (12.12 cm), shoot length (12.24 cm), seedling dry weight (165.85 mg), seedling vigor index (1738) and lower electrical conductivity (0.470 dS/m).

Arif *et al.* (2008) conducted an experiment and observed that priming improved seed establishment in soybean, possibly because pre-germination metabolic activities were completed earlier, preparing the seed for radical protrusion. As seed priming time was increased, grain yield decreased. In comparison to 12 and 18 hours, seed priming for 6 hours resulted in faster and better emergence, as well as higher soybean grain yield. Priming reduced the optimum and ceiling temperatures for germination, aided in germination time advancement, and had no effect on the final percentage emergence (Finch-Savage *et al.*, 2004). Janmohammadi *et al.* (2008) described hydro priming as a suitable, cheap and easy seed invigoration treatment for inbreed lines of maize, when germination is affected by salinity and drought stress. Hydro priming showed result in the earlier germination of desert cacti (Dubrovsky 1996), *Allium porrum* (Ashraf and Bray, 1993), pyrethrum (*Tanacetum cinerariifolium*) (Li *et al.*, 2011), and coriander (Rithichai et *al.*, 2009). Moradi Dezfuli *et al.* (2008) revealed hydro primed seeds for 36 h had lowest values (T50 and MGT).

Ghassemi-Golezani *et al.* (2008) investigated the seed germination properties of lentils using osmo and hydro priming techniques. They discovered that seed priming improved lentil germination and field performance when compared to unprimed seed, but that the effect of different priming was also significant. When compared to the control and seed priming with PEG, hydro-priming resulted in higher seedling emergence in the field. Water priming increased the rate of seedling emergence. As a result, hydro-priming is a simple and effective method for increasing lentil seed germination and seedling emergence in the field.

Park *et al.* (1997) reported that the primed seeds of soybean resulted in good germination and stand establishment in the field trials. Tavili *et al.* (2011) reported that speed of germination of Bromus increased with seed priming treatments rather than that of control. Fotia *et al.* (2008) reported that osmotic seed priming of maize caryopses resulted in more homogenous and faster seed germination as compared to the control.

Elkoca *et al.* (2007) also determined that hydro priming treatment in chickpea enhanced faster and more synchronous germination compared with the unprimed seeds.

2.2.3 Seed germination index

The Germination Index (GI) is an estimate of how long (in days) it takes for a certain germination percentage to occur. Subsequent germination/emergence counts are treated similarly, and the sum of the values is called the Germination Index (GI). Assefa *et al.* (2010) reported that seed priming with GA₃ enhance emergence and germination rate of soybean.

Abro *et al.* (2009) reported that sodium silicate application resulted in higher germination percentage and germination index of wheat seedlings and ultimately leads to improved yield. Mishra and Dwibedi (1980) reported positive effect of seed priming with potassium and distilled water on growth, dry matter accumulation, grain and straw yield in 12 wheat varieties under rain-fed conditions.

Moradi *et al.* (2012) conducted an experiment and found that seed priming increased germination percentage, seed germination index, seedling length, root-shoot ratio, and decreased mean germination time. The usefulness of these treatments varied depending on the level of stress (zero or -8 bar) and the priming condition, which included temperature, duration, solution type, and osmotic potential. Lower priming durations (i.e., 12 and 24 h) improved germination under normal conditions. Higher priming durations (36 and 48 h) provided more protection when the seeds were exposed to drought stress. Priming with urea produced better results than other treatments in this study, and seed priming with fertilizers may be an appropriate treatment for advancing germination under optimum and drought stress conditions. Seed priming enhanced speed of germination, better crop stand and increased yields in different situations for a lot of crops (Rashid *et al.*, 2006). Seed priming hasten germination index and helps to buildup germination metabolites or osmotic regulations during priming (Arif, 2005).

Kachuei and Dehkohne (2014) conducted an experiment to assess the priming effect on the yield and growth way of wheat kinds with seeds (prime) treats with urea, water and lack of priming into factorial in a randomized complete block design on the dry farming kinds (sardary and Azar 2) for three times in Imam Qais region of Chaharmahal va Bakhtiari Province in 91-92. In this study, the yield component such as number of grains per spike, number of spikes per unit area, grain weight, grain yield, biological yield and harvest index were evaluated. The result showed that the priming has significant increase in the number of raceme per unit of area and seed yield. Also, the priming effect has significant increase in the number of seed per spike and seed yield in Azar 2 and it has significant increase in weight of 1000 seed in Sardary too. Totally as a result, the Azar2 and hydro primong in dry farming cultivation of Imam Qais region showed best results rather than of other compounds in this study.

2.2.4 Coefficient of velocity

The coefficient of germination velocity provides an indication of the rate of germination. Its value increases as the number of germinated seeds increases and the time required for germination decreases (Jones and Sanders, 1987).

2.2.5 Energy of emergence

Seed priming helps to enhance rapid and uniform emergence and also help to achieve high vigor, which leading to better stand establishment and yield. Hu *et al.* (2005) conducted an experiment. In this priming experiment a priming method called sand priming was practiced where sand was used as a priming solid matrix. Seeds were mixed with sands that contained 3.8% (v/w) water and sealed in plastic box after that they 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. Otherwise, seedling height, root length, number of root and root dry weight were significantly higher compare to the non-primed controls. Field experiments showed that the seed establishment and yield in sand primed seeds were significantly increased by $19.8\% \sim 22.9\%$ and by $9.8\% \sim 31.2\%$, respectively as compared to soaked seeds without priming. Therefore these findings suggest that sand priming method may useful way to improve seedling establishment in direct-sown rice and possible to be used in the field crop production.

Khalil *et al.* (2010) reported that seed priming enhances speed and uniformity of germination and induces several biochemical changes in the seed that are required to start the germination process such as breaking of dormancy, hydrolysis or mobilization of inhibitors, imbibition and enzyme activation. Seed priming induces many of the metabolic processes involved in the early phases of germination, and primed seeds seedlings emerge faster, grow more vigorously, and perform better in adverse conditions (Cramer, 2002). Germination is trigged by priming and persists following

the re-desiccation of the seeds (Asgedom and Becker, 2001). Priming method resulted in more germination speed mainly in drought stress, saline stress and low temperatures in sorghum, sunflower and melon (Sivritepe *et al.*, 2003; Kaya *et al.*, 2006; Foti *et al.*, 2002).

Seed priming techniques such as hydro priming, hardening, osmo priming, osmo hardening, hormonal priming and hydro priming used to enhance germination and more vigorous plants and better drought tolerance in many field crop like wheat (Baque *et al.*, 2016), chickpea (Kaur *et al.*, 2002), sunflower (Kaya *et al.*, 2006), cotton (Casenave and Toselli, 2007) triticale (Yagmur and Kaydan, 2008).

The objective of this study was to investigate the effect of priming wheat seeds (Triticum aestivum, Giza171 local wheat genotype) with bulk cellulose (BC) and nano cellulose crystals (NC) aqueous solutions and their urea loaded formulations on different germination parameters. BC was extracted from rice straw. NC was prepared from BC through bleaching, acid hydrolysis, and flocculation processes. Priming treatments were prepared using six BC and NC concentrations, three urea concentrations, and their combinations in a completely randomized design with three replicates. Seeds were soaked in the treatment solutions for 3 hours, and then germinated on wet filter papers in Petri dishes for 7 days at 28°C. Results revealed that NC treatments significantly increased root length and surface area, and vigor index compared to BC treatments which had significant positive effect on germination percentage, root fresh weight, and root radius. The concentration of 0.5% BC or 0.3%NC recorded the significantly highest values of all parameters except root radius. Although application of 0.5% urea significantly increased root fresh weight and root radius, significant reduction in germination percentage, root length, and vigor index was observed. According to the statistical analysis of the vigor index data obtained, priming wheat seeds using 0.3% NC without urea loaded is the most efficient treatment to enhance germination of wheat seeds (Sherif et al., 2022).

Baque *et al.* (2016) were conducted a lab experiment to find out the effect of different levels of drought stress on germination behavior of BARI Gom 27. Non primed and primed seeds (osmo primed and hydro primed) 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 non-primed seeds. The drought tolerance capability of non-primed and hydro primed seeds decreased drastically as drought stress increased, but osmo primed seeds showed considerable tolerance capability up to 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.

Dey *et al.* (2013) set an experiment at the Seed Laboratory of the Department of Agronomy, Bangladesh Agricultural University, Mymensingh during the period from January to April 2012 to study the effect of hydro priming on field establishment of seedlings obtained from primed seeds of Boro rice cv. For this experiment BRRI dhan29 seeds were soaked in water for 0, 24, 30, 36, 42, 48, 54 and 60 hours. They examined that priming treatments had significant effect on germination and other growth parameters of rice seedlings. The highest germination, vigor index, population m^2 , length of shoot and root and their weights were found at 15 and 30 DAS. The lowest mean germination time was observed from hydro priming of seeds with 30 hours soaking. On the contrary, no priming treatment showed the lowest germination, vigor index, population m^2 , and the highest mean germination time.

Yousaf *et al.* (2011) conducted an experiment about effects of seed priming with 30 mM NaCl on various growth and biochemical characters of 6 wheat varieties (Tatara-96, Ghaznavi-98, Fakhri Sarhad, Bakhtawar-92, Pirsabaq-2004 and Auqab- 2000) under 4 salinity levels (0, 40, 80 and 120 mM), the effects of varieties and salinity were significant ($P \le 0.05$) and of seed priming was non-significant (P > 0.05) on plant height (cm), root length (cm) and shoot chlorophyll contents.

Yuanyuan *et al.* (2010) conducted an experiment for disclosing the effects of seed priming with water and polyethylene glycol (PEG) on physiological characteristics in rice (*Oryza sativa* L.), the seeds of 4 rice cultivars were treated with H_2O and different concentrations of PEG before germination. Primed or non-primed (control) seeds were then germinated under drought stress conditions simulated with PEG in a serious of concentrations. 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.

A common problem with vegetable production in saline areas is poor crop stand, but for black cumin (Nigella sativa L.) germination data are limited and inconsistent. The effects of chemical priming with Urmia lake salt and urea solutions for 16 h at 30°C on seed germination and seedling growth of black cumin (Nigella sativa L.) were studied under various osmotic stress levels. For a more detailed assessment of chemical priming, the effects of hydro priming for 16 h at 30°C were also studied. A seed lot that was not exposed to any treatment, except disinfection, was used as control. Osmotic stress levels were -2, -4, -6, and -8 bar, which were achieved with polyethylene glycol 6000 (PEG 6000). Seed germination of black cumin was reduced by 16.2%, 33.8%, 50.9%, and 74.9% under osmotic potential -2, -4, -6, and -8 bar, respectively, compared with non-stressed control. Improved germination index values, reduced mean germination time, and increased coefficients of velocity of germination were observed under osmotic stress in primed seeds compared with non-primed control. Averaged over priming treatments, priming improved the final germination percentage by 10.5%, 24.3%, 45.5%, and 74.6% under osmotic potential -2, -4, -6, and -8 bar, respectively. Post-germination growth was also inhibited under low osmotic potential compared with the non-stressed control. Nevertheless, priming improved length and weight of black cumin seedlings and enhanced peroxidase and catalase activity at all osmotic potential levels compared with non-primed seeds. Higher seedling vigor indices were recorded in seedlings from primed seeds with decreasing osmotic potential levels than nonprimed seeds. Urmia lake salt priming had the greatest impact on improving seed germination and vigor indices, especially under osmotic stress conditions. Although seed priming did not completely eliminate the symptoms of osmotic stress in black cumin germination, it is an efficient method to mitigate the impact of osmotic stress on germination of this species (Ghiyasi et al., 2019).

2.3 Effect on growth related attributes

2.3.1 Shoot length

Seed priming increased germination percentage, germination speed, seedling length, root-shoot ratio and decreased mean germination time reported by Moradi *et al.* (2012). Hydro priming improves shoot length of Nerica and also helps to increase seedling growth of it (Mamun *et al.*, 2018). Results showed that seed priming with fertilizers might serve as an appropriate treatment for advancing germination under optimum and drought stress condition.

A field experiment was conducted by Gupta and Singh (2012) to find out the effects of seed priming on chickpea. The treatments consisted of seed priming (seed soaking in water for 8hrs). The results showed that the growth parameters of chickpea were significantly affected by seed priming. Soaking 14 chickpea seeds in water for about 8hrs significantly influenced plant height and nodule dry weight in comparison to non-primed seeds.

Singh *et al.* (2017) conducted and experimented and reported that hydro priming and osmo-priming treatments on shoot length provide significant variation. They showed that 20% Polyethylene glycol (PEG) for 24hrs (13.14cm) gives better effect on rest of the treatments except at 100 ml distilled water for 12hrs (12.11 cm) and 20% Polyethylene glycol (PEG) for 12hrs (12.77 cm) on pea (*Pisum sativum*).

Zamirifar and Bakhtiari (2014) observed an experiment and investigate that effects of seed priming, germination percentage and rate, radical and hypocotyl length and dry weight, root and shoot length, root and shoot and leaves dry weight, leaf number and leaf area per plant. 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.

Akbari *et al.* (2007) observed an experiment and reported that auxin treatments increased the hypocotyl length, seedling fresh and dry weight and hypocotyl dry weight

in wheat seed. In case of wheat seed hydro priming has resulted in 3 to 4-fold increases in root and shoots length in comparison with seedlings obtained from non-primed seeds in drought condition (Kaur *et al.*, 2002). Kumar *et al.* (2017) conducted an experimented on chickpea and reported that higher shoot length has recorded in case of osmo primed seeds than that of non-primed seeds. Among different osmo priming treatments 20% PEG showed the highest shoot length followed by 4% mannitol and control showed the lowest shoot length.

2.3.2 Root length

Seed priming help to early and rapid emergence, stand establishment, higher water use efficiency, deeper roots, increasing in root growth, uniformity in emergence, germination in wide range of temperature, break of seed dormancy (Farhoudi and Sharifzadeh, 2006).

Eivazi (2012) conducted an experiment for evaluating the effects of seed priming on wheat cultivars. Arrangement of treatments were Zarrin, Shariar, Sardary and Azar cultivars as A factor, and priming treatments including distilled water (DW), osmotic solutions (10% PEG, 2.5% KCl, 4% MN, 10% Urea, 5% NaCl W/V) and plant growth inducers (20 ppm IAA, 1000 ppm CCC) with non-primed seed as a control established B factor. During the second year of field experiment two separate treatments were done under drought stress and well watered conditions. Drought stress was withheld by irrigation at booting stage of plants. Maximum amount of absorbed water was determined in cultivar Shariar, 15.5 g DW. Seed weight of all cultivars increased the most when primed with CCC and IAA. Irrespective of the cultivar seedlings related traits revealed that treatment with CCC increased plumule and radical dry weights (11.5 and 8.0 mg) and their lengths (17.2 and 17.8 cm). In opposite, urea pretreatment had negative effects on seedlings growth. All priming treatments increased grain yield and its components, chlorophyll content and nitrogen absorbed under field and green house conditions in four cultivars in comparison to control. Plants arising from seeds primed with potassium chloride under drought stress had the lowest percentage of variation for traits such as relative water content (-9.3%), total dry matter (-10.7%) and grain yield (-4.0%) in comparison with well watered plants. Potassium chloride improved drought tolerance at all wheat cultivars. There were significant correlations between grain yield at primed with KCl and following wheat traits: number of spikes per square meter (0.91^{**}) , number of grains per spike (0.92^{**}) and total dry matter (0.79^{*}) . Therefore, it seems that these traits could be used as indirect criteria for selection of high grain yield for primed seed.

Riedell *et al.* (1985) observed an experiment and reported that GA3 treated soybean seeds recorded better field performance and its stimulation effect in enzymes formations which are important in the early phase of germination which helps for a fast radical protrusion in many field crops.

Higher concentrations of PEG 6000 decreased germination percentage and rate, while shoot and root lengths and shoot fresh and dry weights decreased beyond 60g/L and increased up to 120g/L PEG but further increase in stress negatively influenced cultivars tolerance was founded by (Ashagre *et al.*, 2014).

2.3.3 Shoot dry weight and root dry weight

Seed priming gives higher plant dry weight and seed yield (Harris *et al.*, 2004). The increase in the dry weight and grain yield of mungbean was due to better emergence and better performance per plant (Parera and Cantliffe, 1994). Kumar *et al.* (2017) experimented on chickpea and found that in case of seedling dry weight it was higher (1.02 mg to 1.59mg) in PEG 20% seeds followed by mannitol 4% when compared with control. Laghari *et al.* (2016) found that, shoot and root dry weight (mg) has affected by temperature regimes, hydro-priming periods showed highly significant where as their interaction was significant for shoot dry weight. The maximum mean shoots dry weight mg (54.74) was recorded at hydro-priming period 4 hours whereas the lower shoot dry weight mg (7.898) was observed at hydro-priming period 4 hours whereas the lower root dry weight mg (5.496) found at no priming or check.

Hamayun *et al.* (2010) investigated the adverse effects of drought stress on growth, yield and endogenous phyto hormones of soybean. Polyethylene glycol (PEG) solutions of elevated strength (8% and 16%) were used for drought stress induction. Drought stress period span for two weeks each at pre and post flowering growth stage. They reported that growth and yield attributes of soybean was adversely affected by PEG induced drought stress. Soybean plants were found to be more susceptible to an early drought stress as compared to drought stress at a later growth stage. The level of endogenous growth hormones was also affected by drought stress, as the contents of

plant growth promoting hormone (gibberellin) declined, while those of JA and ABA increased under drought stress. It shows that JA and ABA are concerned with plant stress and reaffirms their role in plant resistance to abiotic stress. SA is related to systemic acquired resistance (SAR) of plant and an increase in the quantity of endogenous SA shows that soybean become more susceptible to injuries and pathogens under drought stress.

Amoghein *et al.* (2013) conducted an experiment on the effect of osmo priming and hydro priming on the different index of germination & early growth of wheat under salty stress. They reported that the simple effect of priming for all the characteristics under study, except of shoot dry weight and simple effect of salinity for all the characteristics under study in the experiment at 1% level was significantly simple effect of seed soaking time (4 hours) only on hypocotyle length was significantly. Interaction of salinity on seed priming for root dry weight, longest root on the 5% level showed a statistical significant difference. Also shoot dry weight had a positive and significant correlation with the first and second leaf length, root number and root longest at the %1 level.

Khalil *et al.* (2010) observed that phenology and dry matter are important traits being affected by seed priming and soil phosphorus (P₂O₅) application. Priming enhanced days to emergence, anthesis and increased dry matter (DM) production compared with non-primed (control). Seed primed with 0.3% P₂O₅ 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% P₂O₅ solution. Water primed seed took less time to emergence (16 days). Soil P₂O₅ 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 P₂O₅ ha⁻¹.

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 hours. 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 non-primed controls. Field experiments showed that the seed establishment and yield in sand primed seeds were significantly increased by $19.8\% \sim 22.9\%$ and by $9.8\% \sim 31.2\%$, respectively as compared to soaked seeds without priming. It is indicated that sand priming method may help to improve 18 seedling establishment in direct-sown rice and possible to be used in the field crop production.

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

Gholami *et al.* (2009) conducted an experiment and found that increase of the synthesis of the hormone gibberellin, which Trigg the activity of α -amylase and other germination specific enzymes like protease and nuclease involved in hydrolysis and assimilation of the starch enhance dry weight of the shoot and dry weight.

2.3.4 Vigor index

Eisvand *et al.* (2010) experimented that the effects of hormonal priming on physiological quality and antioxidant enzymes of aged seeds of tall wheatgrass were evaluated under control and drought (-0.5MPa) conditions. Rate of germination, vigour index and growth of root, shoot and seedling were declined by stress conditions. According to the results, hormonal priming improved physiological quality of deteriorated seeds of tall wheatgrass under drought and control conditions. Germination percentage and rate of germination of primed seeds were higher than non-primed seeds under drought condition. 50ppm of auxin increased germination of naturally aged seeds by 18% under drought condition. Likewise, cytokinin treatment resulted in the highest vigour index. Auxin decreased root length and increased number of seminal roots. For other hormones, seed priming by 100 ppm of gibberellin, 50 ppm of cytokinin, and 50 ppm of abscisic acid (ABA) improved seed performance under control and drought conditions.

Sadeghi *et al.* (2011) performed an experiment to evaluate the effect of seed osmo priming by using PEG 6000 priming media on germination behavior and seed vigor of soybean (cultivar 033). Seeds were primed with six levels of poly ethylene glycol (PEG 6000) 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. Experimental units were arranged factorial in a completely randomized design with three replications. 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.

Umair *et al.* (2010) also reported that seed priming significantly improved the germination rate and vigor of the mungbean seedlings. It is also reported that seed priming improve the antioxidant enzymes activity which decrease the adverse effects of Reactive Oxygen Species (ROS) (Del Ryo *et al.*, 2002). Afzal *et al.* (2011) observed that the priming-induced salt tolerance was associated with improved seedling vigour, metabolism of reserves as well as enhanced K⁺ and Ca²⁺ and decreased Na⁺ accumulation in wheat plants. Seed priming is used for improvement of germination speed, germination vigor, seedling establishment and yield (Talebian *et al.*, 2008). Afzal *et al* (2005) also found that the priming-induced salt tolerance was associated with improved seedling vigor.

Omid and Farzad (2012) observed the impacts of various concentrations of poly ethylene glycol 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.

Safiatou (2012) reported that priming improved seedling vigour and seedling vigour index increased by using seed priming methods in sorghum and Bambara groundnut. Also, highest seedling vigour was achieved by osmo priming (Mannitol priming) in Bambara groundnut and by hydro-priming in sorghum. Farnia and Shafie (2015) found that the probable reason for the highest vigour index might be due to photosynthetic capacity treated with bio fertilizers increases due to increased supply of nutrition.

Amjad et al. (2007) set an experiment to evaluate the influence of seed priming using different priming agents (distilled water, NaCl, salicylic acid, acetyl salicylic acid, ascorbic acid, PEG-8000 and KNO₃) on seed vigour of hot pepper cv. They found that all priming treatments significantly improved seed performance over the control. KNO₃ primed seeds excelled over all other treatments; decreased time taken to 50% germination, increased root and shoot length, seedling fresh weight and vigour over all other priming agents. Seeds were primed in water (hydro priming) and NaCl (1% solution) (halo priming) and sown in pots at different salinity levels [1.17 (control), 3, 5 and 7 dS m⁻¹], along with unprimed seeds. Emergence rate (ER), final emergence percentage (FEP), reduction percentage of emergence (RPE), shoots length, number of secondary roots, seedling fresh weight and vigor were significantly improved by both priming treatments over the control; halo priming was more effective than hydro priming. Number of secondary roots was maximum in halo primed and unprimed seeds. Post-harvest seed enhancement treatments improve germination and seedling vigor (Taylor and Harman, 1998). The effect of priming on seedling vigor and productivity of tomato, chilli, cucumber and cabbage during post-rainy seasons demonstrating that priming improved germination and seedling development and yield of these vegetable species (Maiti et al., 2009).

2.4 Effect on water relation behaviours

Relative water content is influenced by seed quality and seed priming technique. Significantly higher relative water content was recorded in leaves obtained from plots sown with higher quality seeds as compared to those obtained from plots sown with lower quality seeds. Baque *et al.* (2002) observed that primed seed showed the maximum relative water content, higher water retention capacity and exudation rate.

CHAPTER III

MATERIALS AND METHODS

This chapter describes the materials and methods that were used in conducting the experiment. A short description of the experimental site, temperature and humidity of the laboratory room, experimental materials, treatments and design, methods of the study, data calculation procedure and data analysis are discussed at this section.

3.1 Experimental site and duration

The experiment was carried out at the laboratory condition of the department of Agronomy which situated in the Central Laboratory, Sher-e-Bangla Agricultural University (SAU), Sher-e-Bangla Nagar, Dhaka-1207, from August 2019 to July 2020. During the experimentation the temperature and the relative humidity ranges of the laboratory were recorded 25.6°C-33.2°C and 57.5 to 86.7% respectively. It was situated in 24.09°N latitude and 90.26°E longitudes.

3.2. Materials used for the experiment

For this experiment, seeds of the wheat variety BARI Gom 30 were collected from Bangladesh Wheat and Maize Research Institute. The collected seeds were free from all types of visible defects, disease symptoms and pest infestations. Two priming agents *viz.* urea and distilled water were used for nutrient priming and hydro priming respectively. In this experiment PEG (Polyethylene Glycol) was used as a factor which helps to create drought stress condition. Different equipment such as growth chamber, electric balance, petri dish, filter paper, micro pipette, electric balance, oven, paper bag, wash bottle, beaker, forceps etc. were used in this study.

Characteristics of BARI Gom 30

- It is a short duration crop
- Crop duration 102 to 108 days
- It is tolerant to leaf rust and leaf spot disease (blight)
- It is heat tolerant crop

3.3 Chemical used for seed priming

Urea and distilled water were used as priming agents. Polyethylene Glycol (PEG) 6000 was used for inducing drought stress. The 75% alcohol was used for treating seed.

3.4 Experimental design and treatments

Two different experiments were conducted in Completely Randomized Design (CRD) with four replications to achieve the desired objectives. The experiment comprises of

- a) In the first experiment seven levels of priming agent solutions *viz*. distilled water, 0%, 2%, 4%, 6%, 8% and 10% urea solutions.
- b) In the second experiment, three treatments viz. control, hydro priming and urea solutions against five levels of drought stress viz. 0%, 5%, 10%, 15%, and 20% with Polyethylene Glycol (PEG) 6000.

3.5 Experimental details

The entire study was conducted under two different experiments.

3.5.1 First experiment

Study on the germination, growth and water relation behaviors of BARI Gom 30 at different concentrations of priming agents (Urea and Distilled water).

3.5.1.1 Weight of seeds

The 200 g seeds were weighed from the total seed from BARI Gom 30 variety to avoid the unnecessary loss of seeds. Remaining seeds were kept in refrigerator at airtight condition to protect from external damage.

3.5.1.2 Surface treatment

All seeds were surface sterilized with 75% alcohol for 5 minutes then sterilized seeds were rinsed 2 minutes with distilled water for 3 times to reduce the effect of alcohol from the seed surface. Then, seeds were dried in room temperature to remove excess moisture and retain the normal condition.

3.5.1.3 Treatments and design

The experiment was employed with one wheat variety and priming (seven priming) with four replications.

Wheat variety

BARI Gom 30

Seven types of priming solution

 T_0 = Seeds without priming (control)

- T_1 = Seeds primed with distilled water for 9 hours
- T_2 = Seeds primed with 2% urea solution for 9 hours

 T_3 = Seeds primed with 4% urea solution for 9 hours

 T_4 = Seeds primed with 6% urea solution for 9 hours

 T_5 = Seeds primed with 8% urea solution for 9 hours

 T_6 = Seeds primed with 10% urea solution for 9 hours

Every priming media were prepared in distilled water and duration of soaking for hydro and osmo/nutrient priming were 9 hours (Keya, 2018). After soaking seeds were primarily dried by kitchen tissue paper and then air dried, placed in Petri dish. For each replication 30 seeds were placed in 12.5 cm Petri dish on a layer of filter paper no. 102 moistened with 8 ml of distilled water.

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

3.5.1.4 Preparation of priming solutions

a) Urea (CH₄N₂O) preparations (2%, 4%, 6%, 8% and 10%)

The 2% urea solution was prepared by mixing 5 g of urea at 250 ml distilled water. Similarly, 10 g, 15 g, 20 g and 25 g urea were mixed with 250 ml of distilled water to prepare 4%, 6%, 8% and 10% solution of urea, respectively.

b) Distilled water

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

3.5.1.5 Priming technique

For the BARI Gom 30 variety, urea priming and hydro priming were performed. Surface sterilized seeds were divided into three groups: one for priming with distilled water, another for priming with urea, and a third for control (non-primed). Urea primed seeds were divided into five parts and they were soaked for 9 hours in 2%, 4%, 6%, 8%, and 10% urea separately, while hydro primed seeds were soaked in distilled water for 9 hours. To avoid evaporation loss, various petri dishes with covers were used. After 9 hours, all of the seeds were rinsed in the same water. The primed seeds were carefully rinsed three times with distilled water and then wiped dry with tissue paper to remove excess water. Following that, all seeds were air dried separately for 72 hours to restore normal conditions.

3.5.1.6 Experimental set up

Initially, thirty seeds were selected randomly from each treatment and then placed them in a 120 mm diameters petri dishes and whatman no.1 papers were used as growth media. Whatman no.1 was kept saturated by spraying distilled water. All petri dishes were placed at the laboratory room maintaining room temperature 25°C under normal light which helps to faster germination of wheat seeds. This procedure was repeated for ten days. The emergence of 2 mm radicle indicates that germination has occurred. The germination process was observed every 24 hours and the germination progress was recorded as data for future work. Abnormal seedlings had shorter, thicker, and spirally formed hypocotyls, as well as had stunted primary rooted seedlings (ISTA, 2003). At the time of data collection, abnormal, rotted, dead, and fungus-infected seeds were carefully removed with forceps. At the tenth day of germination, five saplings from each treatment were selected at random. The root and shoot were separated, and the weight and length of fresh seedlings were measured with an electric balance and recorded. The root and shoot of each treatment were then emerged in distilled water separately, and the petri dish was covered with a thick cloth to avoid sunlight, which helps in reducing evaporation loss. After 24 hours, the root and shoots were removed from the water and wiped clean with tissue paper. The turgid weight of root and shoot from each treatment was then recorded separately. The root and shoot were then wrapped in brown paper and placed in the oven to dry. The seedlings were then dried

in a 75°C oven for 72 hours. Following that, the oven dried weight of root and shoot were carefully measured and recorded.

3.5.1.7 Achievement from the first experiment

From the first experiment, 6% urea solution gave the best result. So, the 6% urea (15 g of urea) solution mixed with 250 ml distilled water by using a stirring machine was used for the next experiment to evaluate the best result under drought stress conditions imposed in the study.

3.5.2 Second experiment

Study on the germination, seedlings growth and water relation behaviors of primed seed (BARI Gom 30) under drought (Polyethylene Glycol) stress condition.

3.5.2.1 Weight of seeds

The 200 g seed sample was taken from the seed lot of from BARI Gom 30 variety. Remaining seeds were kept in refrigerator at airtight condition to protect from external damage.

3.5.2.2 Surface treatment

All seeds were surface sterilized with 75% alcohol for 5 minutes then sterilized seeds were rinsed 2 minutes with distilled water for 3 times to reduce the effect alcohol from the seed surface. Then, seeds were dried in room temperature to remove excess moisture and retain the normal condition.

3.5.2.3 Treatments and design

Three levels of priming treatments were applied separately in BARI Gom 30 against five droughts stress conditions including control as follows:

- P_0 = Control (no seed priming)
- P₁= Hydro priming (seeds primed with distilled water) and
- P_2 = Seeds primed with 6% urea solution

Here, Primed seeds were placed in 5 levels of drought (Polyethylene Glycol) stress conditions including control as follows:

 $T_0 = 0\%$ PEG concentration

 $T_1 = 5\%$ PEG concentration

 $T_2=10\%$ PEG concentration

 $T_3 = 15\%$ PEG concentration

 $T_4=20\%$ PEG concentration

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

3.5.2.4 Priming solutions and time

The 6% urea solution and distilled water used for BARI Gom 30 due to its best performance in the first experiment. Seeds were soaked in 6% urea solution and distilled water for 9 hours.

3.5.2.5 Preparation of priming solutions

a) Urea (CH4N2O) solutions (6% urea)

The 6% urea solution was prepared by dissolving 15 g of urea at 250ml distilled water by using a stirring machine.

b) Distilled water

Distilled water was collected from Agricultural Chemistry and Soil laboratory of Shere-Bangla Agricultural University.

3.5.2.6 Preparation of drought stress solutions

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

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

3.5.2.7 Priming technique

Urea solution priming and hydro priming were done for BARI Gom 30 variety. Surface sterilized seeds were divided into three parts of which one for distilled water priming, one for 6% urea concentrations priming and the last one for control (non-primed). For urea priming seeds were soaked in 6% urea and hydro priming seeds were soaked in distilled water for 9 hours separately. Different petri dishes with cover were used for avoiding evaporation loss. After 9 hours, all the seeds were rinsed from water at same

time. The primed seeds were rinsed with distilled water for 3 times carefully and then wiped out with tissue paper to remove excess water. After that all seeds are separately air dried for 72 hours to back the normal conditions.

3.5.2.8 Experimental set up

Initially, thirty seeds were selected randomly from each treatment and then placed them in petri dishes (120 mm diameter). Whatman no.1 papers were used as growth media by saturating with PEG solutions. Seeds were kept in different petri dishes and all seeds were sprayed separately by (5%, 10%, 15% and 20%) PEG separately. Here, PEG used as drought stress inducing agent. The dishes were placed at the laboratory room condition under normal light for faster germination of seeds. This process was continued for 10 days. Emergence of 2mm radicle indicates as germination occurred. Every day the germination of seeds was monitored and the progress was recorded as data for next work. Shorter, thicker and spiral formed hypocotyls and stunted primary rooted seedlings were considered as abnormal seedlings (ISTA, 2003). Abnormal, rotted, dead and seed attacked by fungus were taken off by using forceps very carefully at time of data recorded. At 10 day of germination five saplings from each treatment were selected randomly. Then root and shoot were separated and root and shoot weight and length of fresh plant were taken by using electric balance and data recorded. Then root and shoot of each treatment were emerged in distilled water separately and cover all the petri dish by using a thick cloth to avoid sunlight which helps to reduce evaporation loss. After 24 hours root and shoots were picked from water and wiped out by tissue paper. Then turgid weight of root and shoot from each treatment were taken separately and data recorded. Then root and shoot were packed in brown paper separately for oven dry. Then seedlings were dried in oven at 75°C for 72 hours. After that oven dry weight root and shoot were taken carefully and data recorded.

3.5.2.9 Relative water content, water saturation deficit and water retention capacity

At 10th days of germination test, five seedlings were selected randomly from each treatment and fresh weight and length was measured immediate after removing roots. Then, the shoots and roots were soaked at distilled water at room temperature in the dark for 24 hours and Shoots and roots turgid weight was measured after removing the excess water by gently wiping with tissue paper. Then shoots were packed in brown

paper and oven dried at 75°C for 72 hours for measuring dry weight. The fresh, turgid and dry weights of shoots were utilized to calculate relative water content (%), water saturation deficit (%) and water retention capacity (Baque *et al.*, 2002).

3.6 Parameter measurement

The data referring to following characters were recorded from each Petri dish. Data were collected on the following parameters-

3.6.1 Germination percentage

Germination percentage was measured in the 10th days using the formula

 $GP(\%) = \frac{\text{Total number of germinated seeds}}{\text{Total seeds}} \times 100$

3.6.2 Shoot length (mm)

Shoot length was measured in the 10th days using millimeter scale.

3.6.3 Root length (mm)

Root length was measured in the 10th days using millimeter scale.

3.6.4 Seedling length (mm)

Seedling length was measured in the 10th days using millimeter scale by summing shoot length and Root length.

3.6.5 Total germination (TG %)

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

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

3.6.6 Mean germination time (MGT)

Mean germination time (MGT) was calculated according to the equation of Moradi Dezfuli *et al.* (2008).

$$MGT = \frac{\sum Dn}{\sum n}$$

Where,

n = number of seeds germinated on day D, and

D = number of days counted from the beginning of germination

3.6.7 Germination index (GI)

Germination index (GI) was calculated as described in the Association of Official Seed Analysts (AOSA, 1983) as the following formula:

Germination index = $\frac{Gt}{Tt}$

Where,

Gt = number of seeds germinated on day t and Tt = the number of germinated seeds at time Ti.

3.6.8 Coefficient of velocity (CV)

Coefficient of velocity (CV) = (number of germinated seeds per day) is measured according to the method described by Scott *et al.* (1998).

$$CV = \frac{\sum Ni}{\sum TiNi} \times 100$$

Where, Ti= number of days after sowing and Ni = number of seeds germinated on ith day.

3.6.9 Vigor Index (VI)

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

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

Here, TG = total germination

3.6.10 Relative Water Content (RWC %)

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

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

3.6.11 Water Saturation Deficit (%)

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

Water Saturation Deficit (WSD) = 100 - RWC

3.6.12 Water Retention Capacity (WRC)

Water retention capacity was calculated from the turgid and dry weights of shoots by using the following formula used by (Baque *et al.*, 2002). Water retention capacity expressed in percentage.

Water Retention Capacity (RWC) = $\frac{\text{Turgid weight}}{\text{Dry weight}}$

3.6.13 Shoot and root dry weight (mg)

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

3.7 Data analysis techniques

The data obtained for different parameters were statistically analyzed to observe the significant difference among the treatments. The mean value of all the parameters was calculated and analysis of variance was performed. The significance of difference among the treatments means was estimated by the least significant difference (LSD) test at 1% and 5% level of significance. A computer software MSTAT-C was used to carry out the statistical analysis (Gomez and Gomez, 1984). Drawings were made using Excel software.

CHAPTER IV

RESULTS AND DISCUSSION

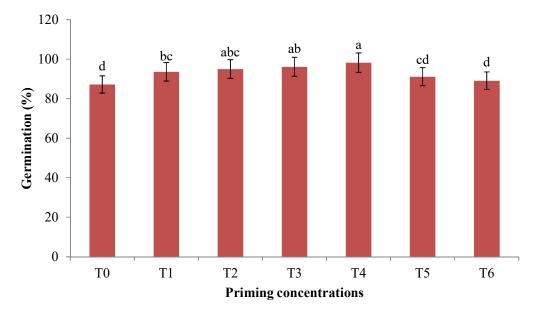
The experiment was conducted to study the 'Induction of drought tolerance capability in wheat through urea and hydro priming'. The result of the germination and growth parameters of wheat which was influenced by different concentration of priming solutions (urea), hydro priming (distilled water) in drought stress (PEG) condition have been presented and discussed in this chapter. The result of the experiment have been summarized and presented by appropriate table and figures. The statistical analysis of variance in respect of all parameters has been shown in Appendix I to Appendix XII.

Experiment no. 1

Study on the germination and water relation behaviors of BARI Gom 30 at different concentrations of priming agents (Urea and Distilled water).

4.1.1 Effect on total germination (%)

Germination percentage of wheat was statistically influenced by different urea concentrations (Fig. 1) and significant differences was observed between control (nonprimed seeds) and primed seeds. Germination percentage of variety BARI Gom 30 increased with increasing priming concentrations with urea up to 6% then decreases gradually with increasing urea concentrations. Treatment T₄ (primed with 6% urea concentration) showed the maximum germination percentage (98.21%) and then gradually decreases due to increasing urea. The germination percentage of hydro priming (distilled water) was found T_1 (93.58%) and lowest germination percentage T_0 (control) was found 87.20%. The result of the study was the agreement with the findings of Keya (2018); Ahammad et al., (2014); Faijunnahar et al., (2017); and Baque et al., (2016). According to Ajouri et al., (2004) priming induces a wide range of biochemical changes in the seed that required promoting the germination process i.e., breaking of dormancy, hydrolysis or metabolism of inhibitors, imbibitions and enzymes activation. Hydro priming significantly induced germination rate (Ghassemi-Golezani et al., 2008). Hydro priming is a useful technique for improving entire germination viz. tomato variety germination percentage was increased in response to 25 hydro-priming treatments (Maiti et al., 2013).

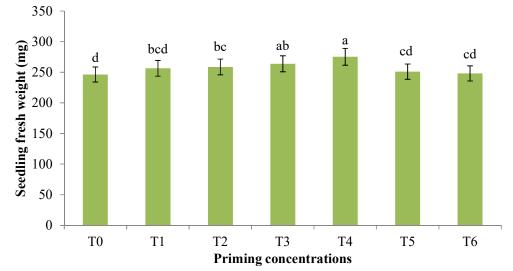


 T_0 = Seeds without priming (control), T_1 = Seeds primed with distilled water for 9 hours, T_2 = Seeds primed with 2% urea solution for 9 hours, T_3 = Seeds primed with 4% urea solution for 9 hours, T_4 = Seeds primed with 6% urea solution for 9 hours, T_5 = Seeds primed with 8% urea solution for 9 hours and T_6 = Seeds primed with 10% urea solution for 9 hours

Figure 1. Effect of priming concentrations on total germination percentage (%) of BARI Gom 30 (LSD_(0.01) = 4.19)

4.1.2 Effect on seedling fresh weight (mg)

Seedling fresh weight of wheat was statistically influenced by different urea solutions (Fig. 2) and significant differences were observed between control (non-primed) and primed seeds. Seedling fresh weight of variety BARI Gom 30 increased with increasing priming concentrations of urea up to 6% and then gradually decreases with increasing concentrations of urea. Treatment T₄ (primed with 6% urea concentration) showed higher seedling fresh weight comparing with others treatment concentrations. The fresh weight of seedlings in T₄ was found 275.19 mg and after that decreased with increasing urea concentrations. The seedling fresh weight (246.26 mg) was observed in control (T₀) treatment. Seed priming with distilled water increased shoot length and seedling weight of Nerica (Mamun *et al.*, 2018).



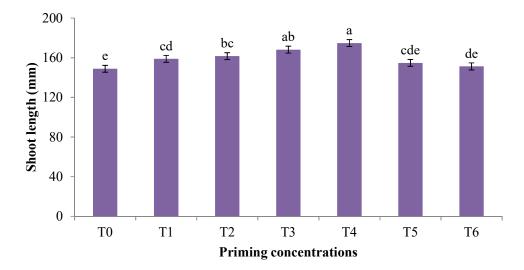
 T_0 = Seeds without priming (control), T_1 = Seeds primed with distilled water for 9 hours, T_2 = Seeds primed with 2% urea solution for 9 hours, T_3 = Seeds primed with 4% urea solution for 9 hours, T_4 = Seeds primed with 6% urea solution for 9 hours, T_5 = Seeds primed with 8% urea solution for 9 hours and T_6 = Seeds primed with 10% urea solution for 9 hours

Figure 2. Effect of priming concentrations on seedling fresh weight (mg) of BARI Gom

 $30 (LSD_{(0.01)} = 11.41)$

4.1.3 Effect on shoot length (mm)

Shoot length (mm) of wheat was statistically affected by different urea solutions (Fig. 3) and observed significant differences between control (non-primed) and primed seeds. Shoot length of BARI Gom 30 increased with increasing priming concentrations of urea up to 6% and then decreased gradually with increasing urea concentrations. The T₄ (primed with 6% urea concentration) showed higher shoot length (174.90 mm) and then decreased due to increasing urea concentrations. The shoot length of hydro priming (T₁) treatment was found (158.89 mm) and lowest shoot length (148.94 mm) result was found in control (T₀). The study was corroborates with the study of previous researcher Kumar *et al.* (2017) who reported that osmo priming of seed increased shoot length than the non-primed seed.

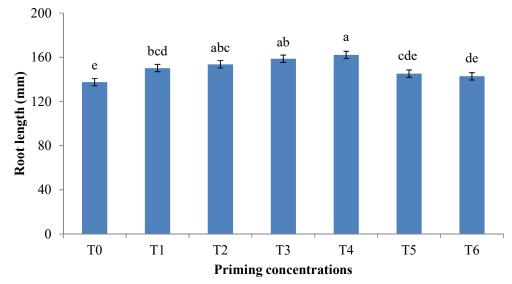


 T_0 = Seeds without priming (control), T_1 = Seeds primed with distilled water for 9 hours, T_2 = Seeds primed with 2% urea solution for 9 hours, T_3 = Seeds primed with 4% urea solution for 9 hours, T_4 = Seeds primed with 6% urea solution for 9 hours, T_5 = Seeds primed with 8% urea solution for 9 hours and T_6 = Seeds primed with 10% urea solution for 9 hours

Figure 3. Effect of priming concentrations on shoot length (mm) of BARI Gom 30 (LSD_(0.01))= 7.94)

4.1.4 Effect on root length (mm)

Significant variation on root length (mm) of wheat was observed due to different urea solution between control (non-primed) and primed seeds (Fig. 4). Root length of variety BARI Gom 30 increased with increasing priming concentration up to 6% urea and then decreased gradually with increasing urea concentrations. The T₄ (primed with 6% urea concentration) showed higher root length (162.07 mm) and then decreased due to increasing urea concentrations. The root length of hydro priming (T₁) treatment was found 150.16 mm and lowest root length (137.47 mm) was found in control (T₀) treatment. The result of this study was the agreements with findings of Eivazi (2012) who reported that root length were better when treated with water and osmo priming over control. Seed priming with GA₃ increases field performance and fast radical protrusion in soybean plants (Riedell *et al.*, 1985).

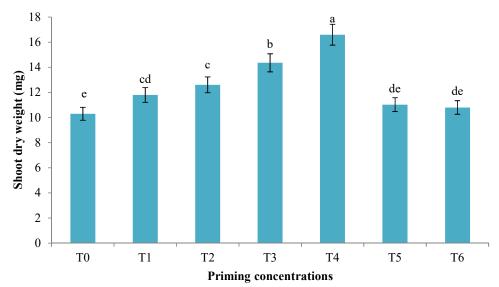


 T_0 = Seeds without priming (control), T_1 = Seeds primed with distilled water for 9 hours, T_2 = Seeds primed with 2% urea solution for 9 hours, T_3 = Seeds primed with 4% urea solution for 9 hours, T_4 = Seeds primed with 6% urea solution for 9 hours, T_5 = Seeds primed with 8% urea solution for 9 hours and T_6 = Seeds primed with 10% urea solution for 9 hours

Figure 4. Effect of priming concentrations on root length (mm) of BARI Gom 30 $(LSD_{(0.01)})= 8.55$).

4.1.5 Effect on shoot dry weight (mg)

Seedling shoot dry weight of wheat was statistically influenced by different urea concentrations (Fig. 5) and significant difference was observed between control (nonprimed) and primed seeds. Seedling shoot dry weight of variety BARI Gom 30 increased with the increasing priming concentrations of urea up to 6% and then gradually decreased with increasing concentrations of urea. Treatment T_4 (primed with 6% urea concentrations) showed higher seedling shoot dry weight comparing with others urea concentrations. The higher shoot dry weight of seedlings in T_4 was found 16.59 mg and after that decreased with increasing percentage of urea concentrations. The seedling shoot dry weight in case of hydro priming (T_1) was found 11.79 mg. The lower seedling shoot dry weight (10.30 mg) was found in control (T_0) treatment. The result of the present study was also agreed by Harris *et al.* (2004) who revealed that seed priming gives higher plant dry weight and seed yield.

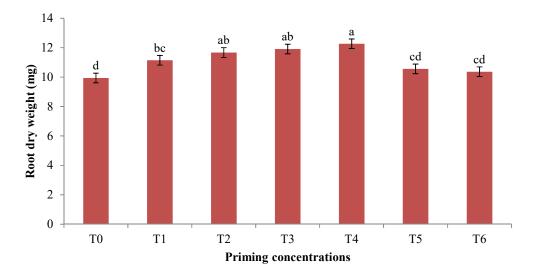


 T_0 = Seeds without priming (control), T_1 = Seeds primed with distilled water for 9 hours, T_2 = Seeds primed with 2% urea solution for 9 hours, T_3 = Seeds primed with 4% urea solution for 9 hours, T_4 = Seeds primed with 6% urea solution for 9 hours, T_5 = Seeds primed with 8% urea solution for 9 hours and T_6 = Seeds primed with 10% urea solution for 9 hours

Figure 5. Effect of priming concentrations on seedling shoot dry weight of BARI Gom $30 (LSD_{(0.01)})= 1.30$

4.1.6 Effect on root dry weight (mg)

Priming concentrations had a significant influence on seedling root dry weight for urea, hydro primed and non-primed seeds (Fig. 6). This effect was very low in urea priming seeds than control. Hydro priming (distilled water) also showed significant result. Seedling root dry weight of variety BARI Gom 30 increased with increasing priming concentrations of urea up to 6% and then gradually reduced with increasing concentrations of urea. From the result of the experiment, the treatment T_4 (primed with 6% urea concentration) showed higher seedling root dry weight comparing with others concentrations. The higher root dry weight (12.27 mg) of seedlings was recorded in T_4 (primed with 6% urea concentrations) treatment and after that decreased with increasing percentage of urea concentrations. The seedling root dry weight in case of hydro priming (T_1) was found 11.14 mg. The lower root dry weight of seedling (9.94 mg) was found in control (T_0) treatment which was lowest of all the treatments. The result of this study corroborates with the study of Baque *et al.* (2016) who reported that the effect of nutrient and hydro priming for 12 hours increased root and shoot dry weight of wheat as compared with the control.



 T_0 = Seeds without priming (control), T_1 = Seeds primed with distilled water for 9 hours, T_2 = Seeds primed with 2% urea solution for 9 hours, T_3 = Seeds primed with 4% urea solution for 9 hours, T_4 = Seeds primed with 6% urea solution for 9 hours, T_5 = Seeds primed with 8% urea solution for 9 hours and T_6 = Seeds primed with 10% urea solution for 9 hours

Figure 6. Effect of priming concentrations on seedling root dry weight of BARI Gom

 $30 (LSD_{(0.01)} = 1.08)$

4.1.7 Effect on relative water content (RWC) (%)

Statistically significant variation was observed on relative water content (RWC) of wheat due to different solution of urea between control (non-primed) and primed seeds (Table 1). Relative water content (RWC) of variety BARI Gom 30 increased with increasing priming concentrations of urea up to 6% and then gradually decreased with the increasing concentrations of urea. Highest result was found in T₄ (primed with 6% urea concentration) treatment comparing with others concentrations. The Relative water content (RWC) of seedlings in T₄ was found 93.15% and after that decreased with increasing percentage of urea concentrations. Relative water content (RWC) in case of hydro priming (T₁) was found 86.39%. The lowest relative water content (RWC) was found in control (T₀). It was found 80.27% which was lowest of all the treatments. This result is similar with the findings of Faijunnahar *et al.* (2017) who reported that osmo and hydro primed seedlings can give better water use efficiency thus plant growth not hampered than non-primed seeds under stress condition.

4.1.8 Effect on water saturation deficit (WSD) (%)

Water saturation deficit (WSD) of wheat variety BARI Gom 30 was significantly influenced by different urea solutions (Table. 1). Water saturation deficit (WSD) of variety BARI Gom 30 decreased with the increasing priming concentrations of urea up to 6% and then gradually increased with increasing concentrations of urea. Treatment T₄ (primed with 6% urea) showed lower water saturation deficit (WSD) comparing with others concentrations. The lowest water saturation deficit of seedlings in T₄ was found 6.85% and after that increased with increasing percentage of urea concentrations. Water saturation deficit in case of hydro priming (T₁) was found 13.61%. The highest water saturation deficit was found in control (T₀). It was found 19.73% which was highest of all the treatments. This result is similar with the findings of Faijunnahar *et al.*, (2017) and Baque *et al.* (2002)who reported that osmo and hydro primed seedlings can give better water use efficiency thus plant growth not hampered than non-primed seeds under stress condition.

4.1.9 Effect on water retention capacity (WRC) (%)

Statistically significant influence was observed on water retention capacity (WRC) of wheat variety BARI Gom 30 due to different urea solutions, hydro primed and nonprimed seeds (Table 1). Water retention capacity (WRC) of variety BARI Gom 30 increased with increasing priming concentrations of urea up to 6% then gradually reduced with increasing concentrations of urea. The T₄ (primed with 6% urea concentrations) showed higher water retention capacity (WRC) comparing with others concentrations. Water retention capacity (WRC) of seedlings in T₄ was found 18.53% and after that decreased with increasing percentage of urea concentrations. Water retention capacity (WRC) of seedlings in T₄ was found 18.53% and after that decreased with increasing percentage of urea concentrations. Water retention capacity (WRC) in case of hydro priming (T₁) was found 14.67%. The lowest water retention capacity was found in control (T₀). It was found 11.76% which was lowest of all the treatments. This result was similar with the findings of Faijunnahar *et al.* (2017) and Baque *et al.* (2016). They revealed that priming helps to activate the metabolic enzymes responsible for seed germination before germination occurs, so hydro and osmo primed seedlings can uptake more water retention capacity.

4.1.10 Effect on vigor index (VI)

Priming with various concentrations of urea showed significant variation on vigor index (Table 1). Seedling vigor index of variety BARI Gom 30 increased with the increasing priming concentrations of urea up to 6% and then gradually decreased with the increasing concentrations of urea. Highest seedling vigor index was found in T_4 (6% urea concentrations) treatment which was 330.93. The seedling vigor index in case of hydro priming (T₁) was found (289.21). The lowest seedling vigor index was found in control (T₀). It was found (249.83) which were lowest of all the treatments. The result of this study corroborates with the study of Keya (2018) who reported that the effect of nutrient and hydro priming for 9 hours increased vigor index of wheat as compared to the control. Osmo priming increased seedling vigor of various vegetable crops and concerning sponge gourd was reported by Maiti *et al.* (2009).

Treatments	Relative water content (%)	Water saturation deficit (%)	Water retention capacity (%)	Vigor index
To	80.27 d	19.73 a	11.76 e	249.83 e
T 1	86.39 bc	13.61 cd	14.67 c	289.21 c
T 2	87.71 a-c	12.29 de	15.15 c	299.42 c
Тз	90.42 ab	9.58 e	16.81 b	314.27 b
T4	93.15 a	6.85 f	18.53 a	330.93 a
T5	84.67 b-d	15.33 bc	13.83 cd	273.34 d
T ₆	82.00 cd	18.00 ab	12.61 de	261.98 de
LSD(0.01)	5.79	2.72	1.42	13.02
CV%	3.35	9.99	4.83	2.26

 Table 1. Effect of priming concentrations on the growth and water relation behaviors of BARI Gom 30

In a column means having similar letter(s) are statistically identical and those having dissimilar letter(s) differ significantly as per 0.01 level of probability

 T_0 = Seeds without priming (control), T_1 = Seeds primed with distilled water for 9 hours, T_2 = Seeds primed with 2% urea solution for 9 hours, T_3 = Seeds primed with 4% urea solution for 9 hours, T_4 = Seeds primed with 6% urea solution for 9 hours, T_5 = Seeds primed with 8% urea solution for 9 hours and T_6 = Seeds primed with 10% urea solution for 9 hours

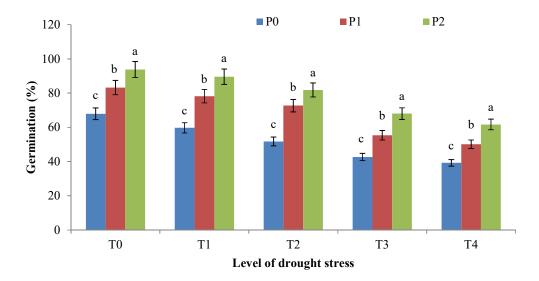
4.2 Experiment 2

Study on the germination and growth behaviors of primed seeds (BARI Gom 30) under drought (Polyethylene Glycol) stress condition

The result of the study showed that every treatment was affected by the experimental factors and there was completely significant difference between primed seeds and nonprimed seeds. Nutrient priming (Urea) and hydro priming increased the germination parameters (germination percentage, germination index and germination time), growth parameters (shoot length, root length, total seedling fresh weight, dry weight and vigor index) and water relation behaviors (relative water content, water saturation deficit and water retention capacity) of wheat than non-primed seeds in drought stress condition. The increasing rate of drought stress in culture media reduces in germination parameters, growth parameters and water retention behaviors in primed seeds as well as non-primed seeds. But the decrease was more prominent in non-primed seeds than nutrient and hydro priming.

4.2.1 Effect on germination percentage

Different drought stress levels showed significant difference in germination percentage under the present study (Fig. 7). Results found that germination from primed and nonprimed seeds decreased gradually with increasing rate of drought stress level. But germination percentage of P_2 (6% urea solution priming) and P_1 (hydro priming) were higher compared with control seeds (without priming). The treatment P_2 (6% urea priming) seed had maximum germination percentage (93.83%) followed by P_1 (83.23%). On the other hand, P₀ (control) seed had (67.94\%) germination percentage found from T_0 (0% PEG) treatment. In T_1 (5% PEG) treatment germination percentage of P_2 , P_1 and P_0 seeds were 89.62%, 78.21% and 59.76%, respectively. On the other hand, minimum germination percentage of P_2 , P_1 and P_0 seeds were found in T₄ (PEG 20%) treatment 61.70%, 50.12% and 39.25%, respectively. Aymen and Hannachi (2012) reported that priming improved germination components and enzyme activity in rapeseed under drought stress conditions and increased rapeseed resistance to drought stress. Baque et al. (2016) reported that in drought stress conditions, the germination percentage of priming wheat seeds yielded a higher result than without priming seeds.



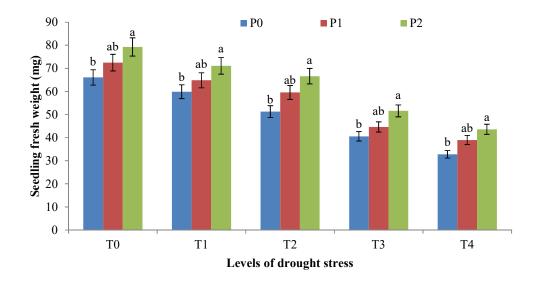
 P_0 = Control (no seed priming), P_1 = Hydro priming (seeds primed with distilled water) and P_2 = 6% urea solution (seeds primed with 6% urea solution)

 $T_0\!\!=0\%$ PEG, $T_1\!\!=5\%$ PEG, $T_2\!\!=10\%$ PEG, $T_3\!\!=15\%$ PEG and $T_4\!\!=20\%$ PEG

Figure 7. Effect of drought stress on germination (%) of BARI Gom 30 $(LSD_{(0.01)}= 9.28, 10.33, 8.94, 8.44 \text{ and } 8.70 \text{ at } T_0, T_1, T_2, T_3 \text{ and } T_4$, respectively)

4.2.2 Effect on seedling fresh weight (mg)

Different drought stress levels showed significant difference in seedling fresh weight (Fig. 8). Results found that seedling fresh weight from primed and non-primed seeds decreased gradually with increasing rate of drought stress level. But seedling fresh weight of P₂ (6% urea priming) and P₁ (hydro priming) was higher compared with P₀ (control) seeds. The maximum seedling fresh weight was observed in P₂ (6% urea priming) was 79.26 mg followed by P₁ (72.47 mg). On the other hand, P₀ (control) seed had 66.07 mg seedling fresh weight found from T₀ (0% PEG) treatment. In T₁ (5% PEG) treatment seedling fresh weight of P₂ (6% urea priming) and P₀ (control) seeds were recorded 71.08 mg, 64.85 mg and 59.89 mg, respectively. On the other hand minimum seedling fresh weight of P₂ (6% urea priming), P₁ (hydro priming) and P₀ (control) seeds were found in T₄ (PEG 20%) treatment as 43.63 mg, 38.97 mg and 32.78 mg, respectively. Keya (2018) revealed that the plant fresh weight of priming seed in wheat is greater than that of non-primed seed in drought stress conditions where PEG was used as a drought inducer.

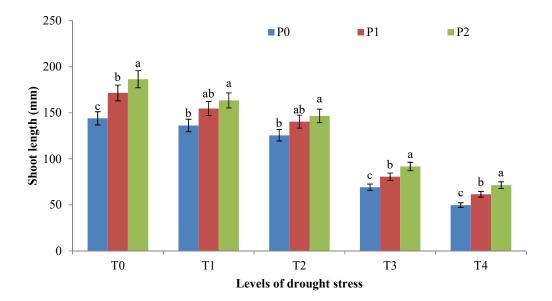


 P_0 = Control (no seed priming), P_1 = Hydro priming (seeds primed with distilled water) and P_2 = 6% urea solution (seeds primed with 6% urea solution) T_0 = 0% PEG, T_1 = 5% PEG, T_2 = 10% PEG, T_3 = 15% PEG and T_4 = 20% PEG

Figure 8. Effect of drought stress on seedling fresh weight (mg) of BARI Gom 30 $(LSD_{(0.01)}= 11.24, 9.85, 9.76, 9.33 \text{ and } 8.30 \text{ at } T_0, T_1, T_2, T_3 \text{ and } T_4, \text{ respectively})$

4.2.3 Effect on shoot length (mm)

Priming significantly influenced shoot length in drought stress condition induced by PEG (Fig. 9). Results found that shoot length from primed and non-primed seeds decreased gradually with increasing rate of drought stress level. But shoot length of P₂ (6% urea priming) and P₁ (hydro priming) were higher compared with P₀ (control) seeds. From the result of the experiment showed that the treatment P₂ (6% urea solution priming) seed had maximum shoot length 186.42 mm followed by P₁ (hydro priming) was 171.52 mm at T₀ (0% PEG) treatment. On the other hand P₀ (control) seed had 144.04 mm shoot length found from T₀ (0% PEG) treatment. In T₁ (5% PEG) treatment shoot length of P₂ (6% urea priming), P₁ (hydro priming) and P₀ (control) seeds were 163.41 mm, 154.62 mm and 136.22 mm, respectively. On the other hand minimum shoot length of P₂ (6% urea priming), P₁ (hydro priming) and P₀ (control) seeds were found in T₄ (PEG 20%) treatment 71.51 mm, 61.49 mm and 49.79 mm, respectively. Kaur *et al.* (2002) reported that nutrient and hydro priming showed three to four fold more growth with respect to root and shoot length in comparison with seedlings obtained from non-primed seeds in drought condition.



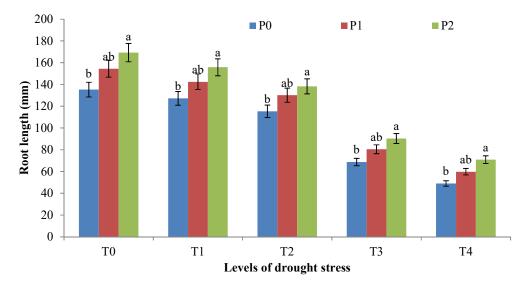
 P_0 = Control (no seed priming), P_1 = Hydro priming (seeds primed with distilled water) and P_2 = 6% urea solution (seeds primed with 6% urea solution)

 $T_0=0\%$ PEG, $T_1=5\%$ PEG, $T_2=10\%$ PEG, $T_3=15\%$ PEG and $T_4=20\%$ PEG

Figure 9. Effect of drought stress on shoot length (mm) of BARI Gom 30 $(LSD_{(0.01)} = 14.40, 19.90, 18.81, 10.68 \text{ and } 7.02 \text{ at } T_0, T_1, T_2, T_3 \text{ and } T_4, \text{ respectively})$

4.2.4 Effect on root length (mm)

Significant variation on root length was found in case of osmo priming and hydro priming (Fig. 10). With increasing rate of drought stress level, root length of primed and non-primed seeds decreased gradually. But root length of P_2 (6% urea priming) and P_1 (hydro priming) were higher compared with P_0 (control) seeds. From the result of the experiment showed that, the treatment P_2 (6% urea priming) seed had maximum root length 169.22 mm followed by P_1 (hydro priming) was 154.46 mm at T_0 (0% PEG) treatment. On the other hand, P_0 (control) seed had 135.21 mm root length observed from T_0 (0% PEG) treatment. In T_1 (5% PEG) treatment root length of P_2 (6% urea priming), P_1 (hydro priming) and P_0 (control) seeds were 155.80 mm, 142.46 mm and 127.26 mm, respectively. On the other hand minimum root length of P_2 (6% urea priming), P_1 (hydro priming) and P_0 (control) seeds were found in T_4 (PEG 20%) treatment were 70.89 mm, 59.84 mm and 49.02 mm, respectively. Salehzade *et al.* (2009) reported that the increased shoot and root length with osmo priming treatments may be due to increasing nuclear replication in root and shoot.



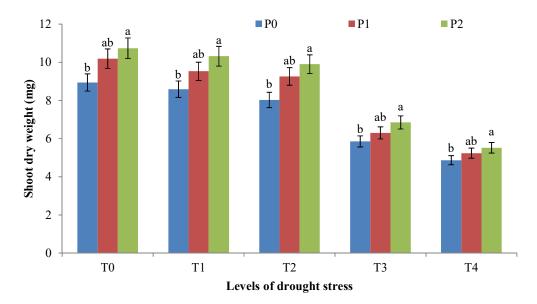
 P_0 = Control (no seed priming), P_1 = Hydro priming (seeds primed with distilled water) and P_2 = 6% urea solution (seeds primed with 6% urea solution)

 $T_0\!\!=0\%$ PEG, $T_1\!\!=5\%$ PEG, $T_2\!\!=10\%$ PEG, $T_3\!\!=15\%$ PEG and $T_4\!\!=20\%$ PEG

Figure 10. Effect of drought stress on root length (mm) of BARI Gom 30 (LSD_(0.01)= 24.28, 20.15, 17.68, 12.50 and 12.47 at T₀, T₁, T₂, T₃ and T₄, respectively)

4.2.5 Effect on shoot dry weight (mg)

Shoot dry weight significantly influenced by seed priming (Fig. 11). Shoot dry weight from primed and non-primed seeds decreased gradually with increasing rate of drought stress level. But shoot dry weight of P₂ (6% urea priming) seeds and P₁ (hydro priming) seeds were higher compared to P₀ (control) seeds. The maximum shoot dry weight was found at P₂ (6% urea priming) treatment which was 10.74 mg followed by P₁ (hydro priming) was 10.19 mg at T₀ (0% PEG) treatment. On the other hand, P₀ (control) seeds had 8.94 mg shoot dry weight which were recorded from T₀ (0% PEG) treatment. In T₁ (5% PEG) treatment shoot dry weight of P₂ (6% urea priming), P₁ (hydro priming) and P₀ (control) seed were 10.32 mg, 9.53 mg and 8.59 mg, respectively. On the other hand minimum shoot dry weight of P₂ (6% urea priming), P₁ (hydro priming) and P₀ (control) seeds were found in T₄ (PEG 20%) treatment which were 5.52 mg, 5.24 mg and 4.87 mg, respectively. Laghari *et al.* (2016) found that shoot and root dry weight has affected by temperature regimes, hydro-priming periods showed highly significant where as their interaction was significant for shoot dry weight.



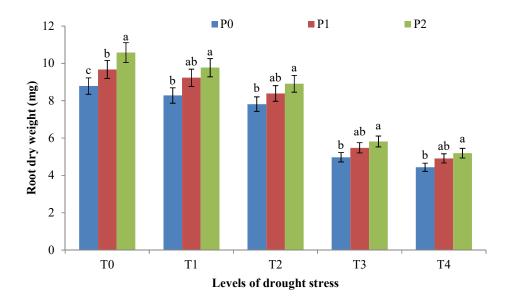
 P_0 = Control (no seed priming), P_1 = Hydro priming (seeds primed with distilled water) and P_2 = 6% urea solution (seeds primed with 6% urea solution) T_0 = 0% PEG, T_1 = 5% PEG, T_2 = 10% PEG, T_3 = 15% PEG and T_4 = 20% PEG

Figure 11. Effect of drought stress on shoot dry weight (mg) of BARI Gom 30 $(LSD_{(0.01)}= 1.29, 1.30, 1.49, 1.00 \text{ and } 0.56 \text{ at } T_0, T_1, T_2, T_3 \text{ and } T_4, \text{ respectively})$

4.2.6 Effect on root dry weight (mg)

Significant variation was exerted in root dry weight due to the effect of different drought stress levels (Fig. 12). Results observed that root dry weight from primed and non-primed seeds decreased gradually with increasing rate of drought stress level. But root dry weight of P_2 (6% urea priming) seeds and P_1 (hydro priming) seeds were higher compared to P_0 (control) seeds. The treatment P_2 (6% urea priming) seeds had maximum root dry weight 10.58 mg followed by P_1 (hydro priming) which was 9.68 mg at T_0 (0% PEG) while P_0 (control) seeds had 8.79 mg root dry weight recorded from T_0 (0% PEG) treatment. In T_1 (5% PEG) treatment root dry weight of P_2 (6% urea priming), P_1 (hydro priming) and P_0 (control) seeds were found in T_4 (PEG 20%) treatment which were 5.19 mg, 4.91 mg and 4.44 mg, respectively. Hu *et al.* (2005) also stated that shoot length and biomass of shoots and root were better when treated with water than unprimed seeds. They also stated that root length, number of root and root dry weights were significantly higher in primed seeds than the non-primed 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.



 P_0 = Control (no seed priming), P_1 = Hydro priming (seeds primed with distilled water) and P_2 = 6% urea solution (seeds primed with 6% urea solution) T_0 = 0% PEG, T_1 = 5% PEG, T_2 = 10% PEG, T_3 = 15% PEG and T_4 = 20% PEG

Figure 12. Effect of drought stress on root dry weight (mg) of BARI Gom 30 (LSD_(0.01)= 0.83, 1.19, 1.04, 0.84 and 0.51 at T₀, T₁, T₂, T₃ and T₄, respectively)

4.2.7 Effect on relative water content (RWC) (%)

Relative water content (RWC) showed significant influence in drought stress condition (Table 2). With increasing rate of drought stress level relative water content of primed and non-primed seeds decreased gradually. But relative water content (RWC) of P₂ (6% urea priming) treatment seeds and P₁ (hydro priming) treatment seeds were higher compared to P₀ (control) treatment seeds. Maximum relative water content 81.59% was found in P₂ (6% urea priming) treatment followed by P₁ (hydro priming) treatment which was 79.65% at T₀ (0% PEG) while P₀ (control) seeds had 78.22% relative water content of P₂ (6% urea priming) treatment. In T₁ (5% PEG) treatment, relative water content of P₂ (6% urea priming) treatment, P₁ (hydro priming) treatment and P₀ (control) seeds were 79.35%, 77.00% and 75.59%, respectively. On the other hand minimum relative water content of P₂ (6% urea priming) treatment, P₁ (hydro priming) and P₀ (control) seeds were found in T₄ (PEG 20%) treatment which were 72.22%,

69.96%, and 68.17%, respectively. Baque *et al.* (2002) reported that osmo and hydro primed increases better water use efficiency thus helps to improves plant growth than non-primed under different stress condition. In addition, according to Faijunnahar *et al.* (2017), optimal osmo priming under drought stress conditions increased the enzymatic activities of seeds, which promote healthy plant growth more than unprimed seeds.

Treatments	Relative water content (%) at					
	To	T 1	T ₂	T 3	T4	
P ₀	78.22 b	75.59 b	71.91 b	69.87 b	68.17 b	
P1	79.65 ab	77.00 ab	73.97 ab	72.54 ab	69.96 ab	
P ₂	81.59 a	79.35 a	75.57 a	73.79 a	72.22 a	
LSD(0.01)	2.56	3.61	2.62	3.46	3.53	
CV%	1.40	2.03	1.55	2.09	2.19	

 Table 2. Effect of drought stress on relative water content (%) of BARI Gom 30 treated with different priming agents

In a column means having similar letter(s) are statistically identical and those having dissimilar letter(s) differ significantly as per 0.01 level of probability

Here, P_0 = Control (no seed priming), P_1 = Hydro priming (seeds primed with distilled water) and P_2 = 6% urea solution (seeds primed with 6% urea solution)

 $T_0{=}\,0\%$ PEG, $T_1{=}\,5\%$ PEG, $T_2{=}\,10\%$ PEG, $T_3{=}\,15\%$ PEG and $T_4{=}\,20\%$ PEG

4.2.8 Effect on water saturation deficit (WSD) (%)

Statistically significant difference was noted on water saturation Deficit (WSD) due to effect of drought stress levels (Table 3). Results found that water saturation deficit (WSD) from primed and non-primed seeds increased gradually with increasing rate of drought stress level. But water saturation deficit of P₂ (6% urea priming) seeds and P₁ (hydro priming) seeds were lower compared to P₀ (without priming) seeds. In T₀ (0% PEG) treatment, water saturation deficit of P₂ (6% urea priming), P₁ (hydro priming) and P₀ (control) seeds were 18.41%, 20.35% and 21.78%, respectively which was minimum of all the drought stress levels. In T₁ (5% PEG) treatment, water saturation deficit of P₂ (6% urea priming) treatment and P₀ (control) seeds were 20.65%, 23.00% and 24.41%, respectively. On the other hand, maximum water saturation deficit of P₂ (6% urea priming), P₁ (hydro priming) and P₀ (control)

seeds were found in T₄ (PEG 20%) treatment which were 27.78%, 30.04% and 31.83%, respectively. According to Baque *et al.* (2014), enzymatic activities were lower in non-prime seed, resulting in weak and lean seedlings. However, due to over priming time, the ageing process was accelerated, resulting in weak and lean seedlings that failed to uptake enough water and provided more water saturation deficit value under stress conditions.

Treatments	Water saturation deficit (%) at					
	T ₀	T 1	T ₂	T ₃	T ₄	
P ₀	21.78 a	24.41 a	28.09 a	30.13 a	31.83 a	
P ₁	20.35 ab	23.00 ab	26.03 ab	27.46 ab	30.04 ab	
P ₂	18.41 b	20.65 b	24.43 b	26.21 b	27.78 b	
LSD(0.01)	2.49	3.08	3.28	3.86	3.84	
CV%	5.37	5.92	5.47	6.03	5.60	

 Table 3. Effect of drought stress on water saturation deficit (%) of BARI Gom 30 treated with different priming agents

In a column means having similar letter(s) are statistically identical and those having dissimilar letter(s) differ significantly as per 0.01 level of probability

Here, P_0 = Control (no seed priming), P_1 = Hydro priming (seeds primed with distilled water) and P_2 = 6% urea solution (seeds primed with 6% urea solution) T_0 = 0% PEG, T_1 = 5% PEG, T_2 = 10% PEG, T_3 = 15% PEG and T_4 = 20% PEG

4.2.9 Effect on water retention capacity (WRC) (%)

Marked influence was exerted on water retention capacity (WRC) of wheat due to drought stress levels (Table 4). Water retention capacity (WRC) of primed and nonprimed seeds decreased gradually with increasing rate of drought stress level. But water retention capacity (WRC) of P₂ (6% urea priming) seeds and P₁ (hydro priming) seeds were higher compared to P₀ (control) seeds. Highest water retention capacity 15.22% was found in P₂ (6% urea priming) seeds followed by P₁ (hydro priming) 13.97% at T₀ (0% PEG) treatment while P₀ (control) seeds had 12.37% water retention capacity at T₀ (0% PEG) treatment. In T₁ (5% PEG) treatment, water retention capacity of P₂ (6% urea priming) treatment, P₁ (hydro priming) treatment and P₀ (control) seeds were 14.49%, 13.26% and 11.86%, respectively. On the other hand, the lowest water retention capacity of P_2 (6% urea priming), P_1 (hydro priming) and P_0 (control) seeds were found in T₄ (PEG 20%) treatment which were 10.84%, 10.34%, and 10.12%, respectively. Primed seed resulted in the maximum relative water content, higher water retention capacity, and exudation rate in drought-affected wheat, according to Baque *et al.* (2002).

Treatments	Water retention capacity (%) at				
	To	T1	T ₂	Тз	T4
P ₀	12.37 b	11.86 b	11.46 b	10.71 b	10.12 b
\mathbf{P}_1	13.97 ab	13.26 ab	12.81 ab	11.49 ab	10.34 b
P ₂	15.22 a	14.49 a	13.60 a	11.99 a	10.84 a
LSD(0.01)	1.77	1.73	1.41	0.84	0.48
CV%	5.58	5.73	4.88	3.23	2.01

 Table 4. Effect of drought stress on water retention capacity of BARI Gom 30 treated with different priming agents

In a column means having similar letter(s) are statistically identical and those having dissimilar letter(s) differ significantly as per 0.01 level of probability

Here, P_0 = Control (no seed priming), P_1 = Hydro priming (seeds primed with distilled water) and P_2 = 6% urea solution (seeds primed with 6% urea solution)

 $T_0\!\!=\!0\%$ PEG, $T_1\!\!=\!5\%$ PEG, $T_2\!\!=\!10\%$ PEG, $T_3\!\!=\!15\%$ PEG and $T_4\!\!=\!20\%$ PEG

4.2.10 Effect on vigor index (VI)

Significant variation was observed on vigor index in wheat seedlings due to different drought stress levels (Table 5). Results observed that vigor index from primed and nonprimed seeds decreased gradually with increasing rate of drought stress level. But vigor index of P₂ (6% urea priming) seeds and P₁ (hydro priming) seeds were higher compared with P₀ (without priming) seeds. Treatment P₂ (6% urea priming) seed had maximum seedling vigor index (333.74) followed by P₁ (hydro priming) treatment (271.31) at T₀ (0% PEG) treatment while P₀ control seedlings had (189.72) vigor index at T₀ (0% PEG) treatment. In T₁ (5% PEG) treatment, seedlings vigor index of P₂ (6% urea priming) treatment, P₁ (hydro priming) treatment and P₀ (control) seeds were 286.07, 232.35 and 157.46, respectively. On the other hand minimum seedlings vigor index of P₂ (6% urea priming), P₁ (hydro priming) and P₀ (control) seeds were recorded from T₄ (PEG 20%) treatment which were 87.86, 60.81 and 38.78, respectively. Janmohammadi *et al.* (2008) reported that hydro priming significantly improved seedling vigor index under both stress and non-stress conditions. Ghiyasi and Tajbakhsh (2013) reported that osmo priming treatment in soybean should be used to overcome seedling vigour index imbibition under drought stress conditions. According to Safiatou (2012), osmo priming with mannitol in Bambara groundnut and sorghum resulted in the highest seedling vigor index under drought stress conditions.

Treatments	Vigor index at					
	T ₀	T ₁	T ₂	T 3	T ₄	
P ₀	189.72 c	157.46 c	124.70 c	58.85 c	38.78 c	
P ₁	271.31 b	232.35 b	196.68 b	89.11 b	60.81 b	
P ₂	333.74 a	286.07 a	233.07 a	123.87 a	87.86 a	
LSD(0.01)	39.14	17.66	12.19	8.83	7.33	
CV%	6.43	3.41	2.87	4.24	5.11	

 Table 5. Effect of drought stress on vigor index of BARI Gom 30 treated with different priming agents

In a column means having similar letter(s) are statistically identical and those having dissimilar letter(s) differ significantly as per 0.01 level of probability

Here, P_0 = Control (no seed priming), P_1 = Hydro priming (seeds primed with distilled water) and P_2 = 6% urea solution (seeds primed with 6% urea solution) T_0 = 0% PEG, T_1 = 5% PEG, T_2 = 10% PEG, T_3 = 15% PEG and T_4 = 20% PEG

CHAPTER V

SUMMARY AND CONCLUSION

The experiment was conducted at the central laboratory of the department of Agronomy, Sher-e-Bangla Agricultural University, Dhaka from August 2019 to July 2020 to study the induction of drought tolerance capability in wheat through urea and hydro priming. The research studies were conducted with two different experiments laid out in Completely Randomized Design (CRD) with four replications.

In the 1st experiment wheat seeds of BARI Gom 30 were pre-soaked in 0%, 2%, 4%, 6%, 8% and 10% urea solutions, respectively and untreated seeds were used as control treatment. Hydro priming seeds also done by pre-soaking seeds in distilled water. Results revealed that all the characteristics related to germination (germination percentage, germination index and germination time), growth parameters (fresh weight of seedlings, shoot length, root length, shoot dry weight, root dry weight and vigor index) and water relation behaviors (relative water content, water retention capacity and water saturation deficit) were significantly influenced by different concentrations of urea followed by hydro priming. All the parameters were given best result in 6% urea solution compared to other treatments. Hydro priming seeds has given better result than control but not more than 6% urea concentration priming seeds. Growth, germination and water relation behaviors of all parameters were increases up to 6% urea priming and decreased gradually with increasing urea solutions.

In the 2nd experiment primed seeds (6% urea priming and hydro priming) and nonprimed seeds were germinated under drought stress condition (Polyethylene Glycol) to calculate the germination behavior. 6% urea priming, hydro priming and control (nonprimed) seeds were germinated under 0%, 5%, 10%, 15% and 20% PEG solution induced drought stress condition. Results showed that under drought stress condition, osmo/nutrient priming given better result than hydro primed seeds in case of all germination, growth and water relations parameters (germination percentage, shoot length, root length, shoot and root dry weight, seedling fresh weight, relative water content, water retention capacity, coefficient of germination and vigor index). Under control condition means without stress or 0% stress condition, all the characters like as germination percentage, seedling fresh weight, shoot length, root length, shoot and root dry weight, relative water content, water retention capacity, vigor index and water saturation deficit showed best results. From the third experiment, priming with 6% urea solution with 9 hours priming for BARI Gom 30 expressed better results over nonprimed and hydro primed seeds at drought stress condition. Results revealed that germination behavior and seedling growth of non- primed and hydro primed seeds decreased drastically as drought stress increased but osmo/nutrient primed seeds with 6% urea solution showed considerable tolerance capability up to drought stress level and then significantly decreased with increasing drought stress.

CONCLUSION

In this study nutrient/osmo priming in wheat variety BARI Gom 30 showed better response. Germination, growth and water relation behaviors of BARI Gom 30 gave the best result when seeds treated with 6% urea concentration compared to hydro primed and non-primed seeds and decreased gradually with increasing the concentrations of urea. So, priming wheat seeds with 6% urea solution for 9 hours is considered as best priming concentration and priming time to induce drought tolerance capability of wheat variety BARI Gom 30 for increasing germination behavior, seedling growth and water relations behavior under a certain level of drought stress condition.

RECOMMENDATION

In this study seeds of BARI Gom 30 treated with 6% urea solution for 9 hours gave the best result compared to hydro primed and non-primed seeds. Osmo/nutrient primed seed enhances germination, seedling growth and water relations behaviors of wheat varieties. Under drought stress conditions, it could be recommended that seeds should be treated with 6% urea solution for 9 hours. Further study should be carried out with different priming agents in different concentrations and different time duration in field or different location will be given better result.

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APPENDICES

Appendix I. Mean square values of priming concentrations on the germination behaviors and seedling growth of BARI Gom-30

		Mean square values of						
Source of variation	Degrees of freedom	Germination percentage	Seedling Fresh weight	Root length	Shoot length	Root dry Weight		
Treatments	6	62.48**	406.75**	310.06**	346.21**	3.00**		
Error	21	4.38	32.51	18.25	15.74	0.29		

**Significant at 1% level of significance

Appendix II. Mean square values of priming concentrations on growth and water
relation behaviors of BARI Gom-30

Source of	Degrees	Mean square values of					
variation	of freedom	Shoot dry weight	Relative water content	Water saturation deficit	Water retention capacity	Vigor index	
Treatments	6	20.43**	82.21**	82.24**	22.04**	3341.61**	
Error	21	0.42	8.37	1.85	0.51	42.33	

**Significant at 1% level of significance

Appendix III. Mean square values of drought levels on germination percentages of BARI Gom-30 seeds treated with different priming agents [Control, Hydro priming and 6% urea conc.]

Sources of variation	Degrees of freedom	Mean square of germination percentages on different drought level at						
	licedom	PEG 0%	PEG 0% PEG 5% PEG 10% PEG 15% PEG 20%					
Treatments	2	677.89**	677.89** 908.46** 948.04** 642.75** 504.51**					
Error	9	16.31	20.22	15.15	13.50	14.35		

** Significant at 1% level of significance

Appendix IV. Mean square values of drought levels on seedling fresh weight of BARI Gom-30 seeds treated with different primed agents [Control, Hydro priming and 6% urea conc.]

Sources of variation	Degrees of	Mean square of seedling fresh weight on different drought level at						
	freedom	PEG 0%	PEG 5%	PEG 10%	PEG 15%	PEG 20%		
Treatments	2	173.89**	125.65**	236.20**	124.40**	118.56**		
Error	9	23.94	18.40	18.07	16.50	13.06		

** Significant at 1% level of significance

Appendix V. Mean square values of drought levels on shoot length of BARI Gom-30 seeds treated with different priming agents [Control, Hydro priming and 6% urea conc.]

Sources of variation	Degrees of	Mean	Mean square of shoot length on different drought level at					
	freedom	PEG 0%	PEG 5%	PEG 10%	PEG 15%	PEG 20%		
Treatments	2	1848.39**	769.93**	465.86**	508.97**	472.50**		
Error	9	39.29	75.02	67.00	21.61	9.35		

** Significant at 1% level of significance

Appendix VI. Mean square values of drought levels on root length of BARI Gom-30 seeds treated with different priming agents [Control, Hydro priming and 6% urea conc.]

Sources of variation	Degrees of	Mean	Mean square of root length on different drought level at					
	freedom	PEG 0%	PEG 5%	PEG 10%	PEG 15%	PEG 20%		
Treatments	2	1166.52**	815.82**	546.59**	469.14**	478.10**		
Error	9	111.64	76.94	59.26	29.59	29.48		

** Significant at 1% level of significance

Appendix VII. Mean square values of drought levels on shoot dry weight of BARI Gom-30 seeds treated with different priming agents [Control, Hydro priming and 6% urea conc.]

Sources of variation	Degrees of	Mean square of shoot dry weight on different drought level at					
	freedom	PEG 0%	PEG 5%	PEG 10%	PEG 15%	PEG 20%	
Treatments	2	3.39**	2.99**	3.60**	1.00*	0.43**	
Error	9	0.32	0.32	0.42	0.19	0.06	

** Significant at 1% level of significance

* Significant at 5% level of significance

Appendix VIII. Mean square values of drought levels on root dry weight of BARI Gom-30 with different priming agents [Control, Hydro priming and 6% urea conc.]

Sources of variation	Degrees of	Mean square of root dry weight on different drought level at				
	freedom	PEG 0%	PEG 5%	PEG 10%	PEG 15%	PEG 20%
Treatments	2	3.23**	2.25**	1.19*	0.73*	0.56**
Error	9	0.13	0.27	0.21	0.14	0.05

** Significant at 1% level of significance

* Significant at 5% level of significance

Appendix IX. Mean square values of drought levels on relative water content of BARI Gom-30 seeds treated with different priming agents [Control, Hydro priming and 6% urea conc.]

Sources of variation	Degrees of	Mean square of relative water content on different drought level at					
	freedom	PEG 0%	PEG 5%	PEG 10%	PEG 15%	PEG 20%	
Treatments	2	11.44**	14.43*	13.47**	16.04**	16.48**	
Error	9	1.24	2.47	1.31	2.27	2.36	

** Significant at 1% level of significance

* Significant at 5% level of significance

Appendix X. Mean square values of drought levels on water saturation deficit of BARI Gom-30 seeds treated with different priming agents [Control, Hydro priming and 6% urea conc.]

Sources of variation	Degrees of	Mean square of water saturation deficit on different drought level at					
	freedom	PEG 0%	PEG 5%	PEG 10%	PEG 15%	PEG 20%	
Treatments	2	11.41**	14.40**	13.50*	16.05*	16.41*	
Error	9	1.18	1.80	2.05	2.83	2.79	

** Significant at 1% level of significance

* Significant at 5% level of significance

Appendix XI. Mean square values of drought levels on water retention capacity of BARI Gom-30 seeds treated with different priming agents [Control, Hydro priming and 6% urea conc.]

Sources of variation	Degrees of freedom	Mean square of water retention capacity on different drought level at						
	needoni	PEG 0%	PEG 5%	PEG 10%	PEG 15%	PEG 20%		
Treatments	2	8.15**	6.95**	4.67**	1.68**	0.54**		
Error	9	0.59	0.57	0.38	0.14	0.04		

** Significant at 1% level of significance

Appendix XII. Mean square values of drought levels on vigor index of BARI Gom-30 seeds treated with different priming agents [Control, Hydro priming and 6% urea conc.]

Sources of variation	Degrees of	Mean square of vigor index on different drought level at				
	freedom	PEG 0%	PEG 5%	PEG 10%	PEG 15%	PEG 20%
Treatments	2	20864.10**	16689.90**	12166.30**	4234.35**	2417.25**
Error	9	290.20	59.10	28.20	14.79	10.19

** Significant at 1% level of significance