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Competence of different priming agents for increasing seed germination, seedling growth and vigor of wheat

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ARTICLE INFORMATION	Abstract
Article History Submitted: 09 Nov 2021 Accepted: 25 Dec 2021 First online: 30 Dec 2021	Wheat in Bangladesh under late sown conditions is often exposed to heat and moisture stresses resulting in poor growth and reduced yield. Seed priming can be a feasible tool to combat those abiotic stresses. The objectives of the present study were to evaluate the efficacy of different seed priming agents for increasing seed emergence, seedling growth, and vigor of wheat. A laboratory experiment was conducted at the Agro Innovation Laboratory.
Academic Editor A K M Mominul Islam akmmominulislam@bau.edu.bd	Department of Agronomy, Bangladesh Agricultural University during the 2nd week of November in 2019 with the wheat variety BARI Gom-33. The experiment comprised the following two factors arranged in a completely randomized design with four replications; Factor A: Priming agent (23) namely; (i) Control (No priming) (ii) Hydropriming (distilled water) (iii) 10000 ppm NaCl (iv) 20000 ppm NaCl (v) 30000 ppm NaCl (vi) 50 ppm
*Corresponding Author Md Parvez Anwar parvezanwar@bau.edu.bd OPEN Caccess	PEG (vii) 100 ppm PEG (viii) 150 ppm PEG (ix) 1 ppm Na ₂ MoO ₄ (x) 2 ppm Na ₂ MoO ₄ (xi) 3 ppm Na ₂ MoO ₄ (xii) 5000 ppm ZnSO ₄ (xiii) 10000 ppm ZnSO ₄ (xiii) 10000 ppm ZnSO ₄ (xvi) 50 ppm CuSO ₄ (xvii) 75 ppm CuSO ₄ (xviii) 10000 ppm KCl (xix) 20000 ppm KCl (xx) 20000 ppm KCl (xx) 20000 ppm KCl (xxi) 10000 ppm CaCl ₂ (xxii) 20000 ppm CaCl ₂ (xxiii) 30000 ppm CaCl ₂ ; Factor B: Priming duration (2); (i) 6 hours (ii) 12 hours. One hundred seeds were placed manually on moist sand maintaining more or less equal distance in each petri dish. Results revealed that seed priming has a positive impact (except CuSO ₄ and ZnSO ₄) on seed germination and seedling growth of wheat. Germination parameters and seedling growth traits were significantly affected by priming agents, and a clear advantage of seed priming over non control was evident. Seed priming for 12 hours performed better than6 hours priming. Among the priming agents tried, CaCl ₂ performed the best in terms of germination percent and index, root and shoot length, seedling length, dry weight and vigor were. It may therefore be concluded that seed priming can increase seed germination rate and improve seedling vigor of wheat to some extent, and pre-sowing seed priming with CaCl ₂ for 6 hrs can be recommended for wheat.
	Keywords: Seed invigoration, seed emergence, germination coefficient, seedling vigor, wheat



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1 Introduction

Wheat (Triticum aestivum L.) is an important crop considering first in terms of area and third in terms of production out of three major cereal crops i.e. maize, rice and wheat in the world (Gahtyari et al., 2017). Wheat grain comprises three groups of major components namely starch, protein, and cell wall polysaccharides (dietary fiber), and a range of minor components beneficial to human health (Shewry et al., 2013). It is consumed as the major staple food worldwide and has been the most abundant source of calories and protein in the human diet alongside supplying nearly 20% of the total dietary protein (Braun et al., 2010). Global wheat production is expected to reach a new record of 780 million tons in 2021, according to a preliminary forecast (FAOSTAT, 2021). Wheat is considered the second most important cereal crop in Bangladesh after rice (Islam, 2021). A unique feature of wheat in Bangladesh is the 100% adoption of modern high yielding varieties (Rahman and Hasan, 2009).

Wheat is now becoming an indispensable food item of the people of Bangladesh. Day by day its demand is raising due to changes in lifestyle and food habits (Barma et al., 2019). The total area under the wheat crop of FY 2019-20 has been estimated as 3,32,274 hectares compared to 3,30,348 hectares of FY 2018-19 (BBS, 2020), 0.58% higher than the previous year. Total production of wheat in FY 2019-20, has been estimated as 10,29,354 metric tons compared to 10,16,811 metric tons in the FY 2018-19, which is 1.23% higher (BBS, 2020). Despite that Bangladesh has been emerged as the fifth biggest importer of wheat in recent years (FAOSTAT, 2019). However, sustainable wheat production in Bangladesh is threatened by several stresses (biotic and abiotic) in addition to the competition with other winter crops (Barma et al., 2019). According to Zampieri et al. (2017), among the abiotic stresses heat stress is most severe to affect crop growth and productivity under the context of global climate change. An estimated 6% fall in wheat production occurs globally for every 1 °C rise in temperature (Asseng et al., 2014). Due to increasing temperature, it was estimated that Bangladesh would be 1 °C warmer by 2020 compared to 1971 (Kamrul and Hore, 2021).

Seed priming, partial hydration of seeds without radicle emergence (Farooq et al., 2007b), is a technique that improves the germination, seedling emergence, growth, and yield attributes of a crop (Pant and Bose, 2016). It is a controlled hydration process that is followed by redrying and triggers many of the physiological processes associated with the early phase of germination and prepares the seed for radicle protrusion which suspends the seeds in the lag phase (Paparella et al., 2015). Acceleration of germination in prime seeds can be due to the increasing activity of the degrading enzymes, such as α -amylase, synthesis

of RNA and DNA, the amount of ATP, and the number of mitochondria (Afzal et al., 2002). Seed priming can break seed dormancy, curtail seedling emergence time, improve seedling vigor, and leads to better germination and growth of plants (Mondal et al., 2011; Srivastava and Bose, 2012). Primed seeds in solutions of macro and micronutrients have been shown to improve germination and seedling vigor of wheat (Joshi et al., 2018; Hussain et al., 2019; Farooq et al., 2020a; Rai-Kalal and Jajoo, 2021), rice (Mamun et al., 2018; Anwar et al., 2021), maize (Arief et al., 2020) and other crops (Prażak et al., 2020; Farooq et al., 2020b).

Generally, various seed priming techniques, which include hydropriming, osmopriming, chemical priming, nutrient priming, hormonal priming, and redox priming, are used to induce pre-germination changes (Paparella et al., 2015; Hussain et al., 2019). Seed pretreatment, either by coating (Wei et al., 2014) or pre-soaking (polyamines) (Chunthaburee et al., 2014), was found to maintain enhanced tolerance when plants were exposed to stress conditions. Primed seeds showed the higher activities of many enzymes for metabolism of proteins (proteases), carbohydrates (α and β amylases), and lipids (isocitrate lyase), involved in the mobilization of stored reserves in seed, and these enzymes play a pivot role in the breakdown of the macromolecules for embryo growth and development which exerts a positive influence on early and better seedling emergence (Varier et al., 2010; Sisodia et al., 2018). Priming is a double technology to enhance rapid and uniform emergence in many crop species, thus achieve high vigor and better yields (Farooq et al., 2007a).

Therefore, seed priming approach should be explored to mitigate high-temperature stress of wheat under late sown condition in Bangladesh. Considering the above facts, the present research work was undertaken to assess the performance of a range of seed priming techniques on seed germination and seedling vigor of wheat and to recognize the best priming technique for increased germination and enhanced seedling vigor of wheat.

2 Materials and Methods

2.1 Experimental site and duration

The laboratory experiment was conducted at the Agro Innovation Laboratory, Department of Agronomy, Bangladesh Agricultural University during the 2nd week of November in 2019.

2.2 Experimental treatments and design

The experiment comprised two factors namely priming agent (23 levels) and priming duration (2 levels) arranged in a Completely Randomized Design (CRD) with 4 replications. Priming agents included Control (No priming), Hydropriming (distilled water), 10000 ppm NaCl, 20000 ppm NaCl, 30000 ppm NaCl, 50 ppm PEG, 100 ppm PEG, 150 ppm PEG, 1 ppm Na₂MoO₄, 2 ppm Na₂MoO₄, 3 ppm Na₂MoO₄, 5000 ppm ZnSO₄, 10000 ppm ZnSO₄, 15000 ppm ZnSO₄, 25 ppm CuSO₄, 50 ppm CuSO₄, 75 ppm CuSO₄, 10000 ppm KCl, 20000 ppm KCl, 30000 ppm KCl, 10000 ppm CaCl₂, 20000 ppm CaCl₂ and 30000 ppm CaCl₂. Priming durations were 6 and 12 hrs.

2.3 Plant material used

BARI Gom-33, developed by Bangladesh Agricultural Research Institute (BARI) collaborated with the International Maize and Wheat Improvement Center (CIMMYT) in 2017, was used as the plant material in the present study. It was a simple cross between Kachu and Solala. Kachu is a 'Kauz'-derived highyielding wheat variety carrying a 2NS segment for blast resistance and Solala is derived from a CIM-MYT's pre-breeding cross, involving a durum wheat derivative that provides a 5–8% yield benefit and is 50-55 ppm zinc (Zn) enriched. It was found with 1.8% disease severity comparing with other varieties. BARI Gom-33 is grown in rabi (winter) season in Bangladesh. The optimum sowing time is between 15 November and 30 November. Generally, 110-115 days are required from sowing to harvest. The yield is 3.95 t ha⁻¹. It is high yielding, moderately drought tolerant, grain white glossy and medium in size. This blast-resistant variety is also resistant to leaf rust disease and tolerant to leaf spot disease (BARI, 2007).

2.4 Priming agents used

All the priming agents used in the experiment were of laboratory grade. Details of the priming agents are presented in Table 1.

2.5 Seed priming

Seeds were soaked in different priming agent solution (previously prepared using distilled water) as per treatments for 6 hours or 12 hours at room temperature ($25 \pm 2 \,^{\circ}$ C). The ratio of seed weight to solution volume was 1:5 (g L⁻¹). Then, seeds were removed from the priming agent and, wiped lightly with blotting paper, and allowed to dry by forced air. Dried seeds were put in brown envelopes with tags in sealed polythene bags and stored in a refrigerator at $5 \pm 1 \,^{\circ}$ C until being placed for germination. While control treatment received no prior seed priming.

2.6 Germination medium and seed placement

Sterilized sand was used as germination media and plastic-made Petri dishes of 90 mm diameter with 15 mm depth were used as containers. The moisture content of the media was maintained at around 80% of field capacity by watering with distilled water as necessary. One hundred seeds were placed manually on moist soil maintaining more or less equal distance in each petri dish. Petri dishes were put on the desk of the laboratory at room temperature 25 ± 2 °C. Every petri dish was monitored regularly to maintain proper moisture levels. No occurrence of insects or diseases was recorded.

2.7 Observations made

Data were collected on germination percentage, mean germination time, germination index, seedling vigor index, germination co-efficient, seedling root and shoot length, root-shoot ratio, root, shoot and seedling dry weight.

2.8 Germination percentage (GP)

The number of germinated seeds was counted on the 7th day. The appearance of plumule over the sand layer was considered germination.

$$GP = \frac{S_G}{S_T} \times 100 \tag{1}$$

where, GP = germination (%), S_G and S_T denote the numbers of seed germinated and total number of seeds set for germination, respectively.

2.9 Mean germination time (MGT)

$$MGT = \frac{\sum D_n}{n}$$
(2)

where *n* is the number of seeds germinated on day *D*, and *D* is the number of the day counted from the beginning of germination.

Germination index (GI)

$$GI = \frac{G_i}{D_i} + \ldots + \frac{G_n}{D_n}$$
(3)

where G_i = number of seeds germinated on *i*th day (D_i) , and G_n = number of seeds germinated on final counting day (D_n) .

2.10 Seedling vigor index (SVI)

After seed placement for germination, on the 7th day, 5 seedlings from each replicate were randomly selected. Root and shoot lengths were measured, and then oven-dried at 70 °C for 72 hrs. to record the root

Sl. no.	Priming agent	Chemical formula	Manufacturer
1	Sodium chloride	NaCl	MERCK, India
2	Potassium chloride	KCl	MERCK, India
3	Calcium chloride	CaCl ₂	MERCK, India
4	Copper sulfate	$CuSO_4.5H_2O$	MERCK, India
5	Zinc sulfate	$ZnSO_4.7H_2O$	MERCK, India
6	Sodium molybdate	Na ₂ MoO ₄ .2H ₂ O	MERCK, India
7	Polyethylene glycol 4000	PEG 4000	LOBAL Chemie, India

Table 1. Description of the priming agents

and shoot dry weight of seedlings. Seedling vigor index (SVI) was calculated as follows:

$$SVI = \frac{L \times GP}{100}$$
(4)

where L = seedling length (root length + shoot length) in cm, and GP = germination percentage.

2.11 Germination co-efficient (GC)

The co-efficient of germination was calculated using the following formula (Copeland 1976).

$$GC = \frac{N100 \left(A_1 + A_2 + \dots + A_n\right)}{A_1 T_1 + A_2 T_2 + \dots + A_n T_n}$$
(5)

Where *A* is the number of seeds germinated, *T* is the time corresponding to A, n = Number of days to the final count.

2.12 Seedling growth

On the 7th day of seed placement for germination, 5 seedlings from each replicate were randomly selected. Root length was measured from the base of the plant up to the end of the longest root and expressed in mm. Shoot length was measured from the base of the plant up to the tip of the longest leaf and expressed in mm. The root-shoot ratio was calculated as the ratio of root length to shoot length. Root and shoot dry weight of sample seedlings were measured after drying the roots in an oven at 70 °C for 72 hrs. Finally, the root and shoot dry weight of each seedling were calculated and expressed in mg.

2.13 Statistical analysis

The recorded data were compiled and tabulated for statistical analysis. Analysis of variance (ANOVA) was done with the help of the computer package MSTAT-C (Statistical software). The mean differences among the treatments were adjudged by Duncan's Multiple Range Test (Gomez and Gomez, 1984) at 5% level of significance.

3 Results

3.1 Germination and seedling vigor

Priming agent brings a significant effect (p<0.01) on seed germination rate and seedling vigor inof BARI Gom-33 (Table 2). In general, seed priming showed a positive effect on seed germination and seedling vigor of wheat. Hydropriming and CuSO₄ priming at any concentration reduced germination rate while CaCl₂ priming enhanced germination rate the most. Compared to control, seed priming with 10000 ppm CaCl₂ took the least mean germination time (1.01 days), and priming with 15000 ppm ZnSO₄ resulted in the highest mean germination time (2.95 days). Priming with 10000 ppm CaCl₂, 10000 ppm KCl, 20000 ppm KCl, and 20000 ppm CaCl₂ produce the best germination index; on the other hand, priming with CaCl₂, 50 ppm PEG, and 100 ppm PEG resulted in the most vigorous seedlings. Duration of priming had a significant effect (p<0.01) on germination rate and seedling vigor in BARI Gom-33. The 6-hour priming duration resulted in higher germination rate and seedling vigor compared to 12-hour priming duration (Table 3). Interaction between priming agent and duration of priming did not produce any significant effect (p>0.01) on germination rate, mean germination, germination index, or seedling vigor either (Table 4).

3.2 Seedling growth

Priming agent exerted a significant effect (p<0.01) on the root length of BARI Gom-33. Seed priming with CaCl₂, 50 ppm PEG and 150 ppm PEG performed the best in terms of root length while ZnSO₄ priming performed the worst. It is interesting to note that ZnSO₄ priming resulted in even shorter root length compared to control (Table 5). The duration of priming and interaction between priming agent and priming duration had no significant effect (p>0.01) on the root length of BARI Gom-33 (Table 6 and Table 7). A significant positive effect of seed priming was found on shoot length. Priming agent CaCl₂, 50 ppm PEG, and 150 ppm PEG performed the best in terms of shoot length, on the other hand ZnSO₄ priming resulted in shorter shoot length compared to control (Table 5).

Priming agent	Final GP (%)	MGT (days)	GI	SVI	GC
Control	84.75 i	3.550 a	30.751	11.92 j	22.88 h
Hydropriming	89.13 fgh	2.675 cd	42.38 hi	14.21 i	24.90 efgh
10000 ppm NaCl	92.38 cde	1.813 fg	67.13 efg	18.06 cde	25.77 def
20000 ppm NaCl	91.25 defgh	2.063 e	66.13 fg	17.43 def	25.65 def
30000 ppm NaCl	88.38 h	1.888 ef	65.13 g	15.78 gh	25.56 defg
50 ppm PEG	96.00 ab	1.450 hi	68.75 cdef	21.26 b	26.91 abcde
100 ppm PEG	94.88 bc	1.612 gh	67.75 defg	20.78 b	26.31 cde
150 ppm PEG	92.88 cde	1.650 fgh	65.75 fg	19.22 c	26.39 bcde
1 ppm Na ₂ MoO ₄	91.88 def	2.563 cd	43.75 h	16.55 fgh	23.41 h
$2 \text{ ppm Na}_2 \text{MoO}_4$	92.50 cde	2.713 bc	41.50 hij	16.73 efg	23.01 h
$3 \text{ ppm Na}_2\text{MoO}_4$	90.25 efgh	2.600 cd	39.50 ijk	15.28 hi	23.10 h
5000 ppm ZnSO ₄	93.00 cde	2.425 d	38.88 ijk	9.271 k	23.67 fgh
10000 ppm ZnSO ₄	91.63 defg	2.737 bc	37.88 jk	8.545 kl	23.41 gh
15000 ppm ZnSO ₄	90.38 efgh	2.950 b	36.88 k	7.4181	23.38 gh
25 ppm CuSO ₄	90.13 efgh	2.475 cd	41.13 hij	11.11 j	23.40 gh
50 ppm CuSO ₄	91.00 defgh	2.600 cd	39.50 ijk	10.68 j	23.79 fgh
75 ppm CuSO ₄	88.75 gh	2.688 cd	38.13 jk	9.379 k	23.06 h
10000 ppm KCl	96.88 ab	1.263 ijk	73.38 ab	18.91 c	27.73 abcd
20000 ppm KCl	96.63 ab	1.313 ij	72.13 abc	18.25 cd	27.35 abcd
30000 ppm KCl	94.00 bcd	1.250 ijk	70.50 bcde	17.11 defg	27.55 abcd
10000 ppm CaCl ₂	98.88 a	1.013 k	74.25 ait	22.77 a	28.83 a
20000 ppm CaCl ₂	96.63 ab	1.112 jk	72.88 ab	21.32 b	28.55 ab
30000 ppm CaCl ₂	96.25 ab	1.188 jk	70.75 abcd	20.74 b	28.45 abc
Sx	0.92	0.08	1.13	0.45	0.69
Sig. level	**	**	**	**	**
CV (%)	2.81	11.24	5.82	8.1	7.65

Table 2. Effect of priming agent on germination and seedling vigor of BARI Gom-33

GP = germination percentage, MGT = mean germination time, GI = germination index, SVI = seedling vigour index, and GC = germination co-efficient

Table 3. Effect of priming duration on germination and seedling vigor of BARI Gom-33

Priming duration	Final GP (%)	MGT (days)	GI	SVI	GC
6 hrs	93.20 a	2.08	55.33	16.12 a	25.37
12 hrs	91.88 b	2.06	54.65	15.42 b	25.33
Sx	0.27	0.02	0.33	0.13	0.2
Sig. level	**	NS	NS	**	NS
CV (%)	2.81	11.24	5.82	8.1	7.65

GP = germination percentage, MGT = mean germination time, GI = germination index, SVI = seedling vigour index, and GC = germination co-efficient

Seed priming duration also had a significant effect (p<0.01) on the shoot length of BARI Gom-33, and 6 hrs priming performed better than 12 hrs priming (Table 6). Priming agent and duration failed to interact significantly for shoot length of BARI Gom-33 (Table 7). Priming agents showed a significant effect on seedling length of BARI Gom-33. Seed priming with 10000 ppm CaCl₂, 20000 ppm CaCl₂ and 50 ppm PEG resulted in the highest seedling length, while ZnSO₄ priming produced the lowest seeding length which

was even lower than that of control (Table 5). The duration of priming made a significant effect (p<0.01) on the seedling length of BARI Gom-33, and priming for 6 hrs resulted in higher seedling length than 12 hrs priming (Table 6). Like root length and shoot length, seedling length of BARI Gom-33 was also found unaffected by the interaction between seed priming agent and priming duration (Table 7).

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Priming agent × prii	ming duration	Final GP (%)	MGT (days)	GI	SVI	GC
Control	6 hrs	84.75	3.55	30.75	11.92	22.88
	12 hrs	84.75	3.55	30.75	11.92	22.88
Priming agent × prim Control Hydropriming 10000 ppm NaCl 20000 ppm NaCl 30000 ppm NaCl 30000 ppm NaCl 50 ppm PEG 100 ppm PEG 150 ppm PEG 150 ppm PEG 1 ppm Na2MoO4 2 ppm Na2MoO4 3 ppm Na2MoO4 5000 ppm ZnSO4 10000 ppm ZnSO4 50 ppm CuSO4 50 ppm CuSO4 50 ppm CuSO4 75 ppm CuSO4 10000 ppm KCl 30000 ppm KCl 20000 ppm CaCl2 20000 ppm CaCl2	6 hrs	89.25	2.85	42.00	14.90	25.13
	12 hrs	89.00	2.50	42.75	13.53	24.68
10000 ppm NaCl	6 hrs	93.75	1.93	68.25	18.38	25.75
11	12 hrs	91.00	1.70	66.00	17.73	25.80
20000 ppm NaCl	6 brs	92.00	2 10	67.25	18.01	25.65
20000 ppin ruci	12 hrs	90.50	2.03	65.00	16.87	25.65
20000	(1	90. 2 E	1.70	(()5	16.20	25.50
30000 ppm NaCI	6 nrs 12 brs	89.25 87.50	1.78	66.25 64.00	16.38	25.50
	12105	07.50	2.00	04.00	15.17	25.05
50 ppm PEG	6 hrs	97.00	1.50	70.25	21.68	27.08
	12 hrs	95.00	1.40	67.25	20.83	26.75
100 ppm PEG	6 hrs	94.75	1.70	68.75	20.89	26.48
	12 hrs	95.00	1.53	66.75	20.66	26.15
150 ppm PEG	6 hrs	93.75	1.70	65.75	19.24	26.53
11	12 hrs	92.00	1.60	65.75	19.19	26.25
1 ppm Na ₂ MoO ₄	6 hrs	93.00	2.58	44.00	17.16	23.58
- FF	12 hrs	90.75	2.55	43.5	15.93	23.25
2 ppm Na ₂ MoO ₄	6 hrs	93.00	2.68	40.75	16.01	23.00
2 ppin Na_2WOO_4	12 hrs	92.00	2.08	40.75	16.51	23.00
2 mm Na MaO	(1	92.00	2.70	12.25	10.01	20.00
$3 \text{ ppm Na}_2 \text{MoO}_4$	6 hrs	91.25	2.50	39.75	15.83	23.15
	12 nrs	89.23	2.70	39.25	14.73	23.05
5000 ppm ZnSO ₄	6 hrs	93.75	2.40	40.00	9.46	23.75
	12 hrs	92.25	2.45	37.75	9.08	23.60
10000 ppm $ZnSO_4$	6 hrs	93.25	2.83	38.75	8.94	23.38
	12 hrs	90.00	2.65	37.00	8.15	23.45
15000 ppm ZnSO ₄	6 hrs	90.5	2.90	36.50	7.64	23.50
11 7	12 hrs	90.25	3.00	37.25	7.19	23.25
25 ppm CuSO	6 hrs	90.75	2.38	40.75	11.33	22.93
	12 hrs	89.5	2.58	41.50	10.88	23.88
	6 hrs	01 50	2.52	28 75	11.02	22 72
50 ppin Cu_4	12 hrs	90.50	2.53	40.25	10.32	23.75
	12105	20.00	2.00	10.25	10.52	20.00
75 ppm $CuSO_4$	6 hrs	89.25	2.58	38.25	9.45	23.08
	12 hrs	88.23	2.80	38.00	9.31	23.05
10000 ppm KCl	6 hrs	97.75	1.28	73.75	19.49	27.78
	12 hrs	96.00	1.25	73.00	18.34	27.68
20000 ppm KCl	6 hrs	97.25	1.40	71.5	18.70	27.30
	12 hrs	96.00	1.23	72.75	17.8	27.40
30000 ppm KCl	6 hrs	95.25	1.30	70.75	17.42	27.48
11	12 hrs	92.75	1.20	70.25	16.79	27.63
10000 ppm CaCla	6 hrs	99.00	1 10	75.00	23.15	29.08
10000 ppin euer2	12 hrs	98.75	0.93	73.50	22.39	28.58
20000	(1	07.25	1.00	72.00	21.0	20.40
20000 ppm CaCl_2	6 nrs 12 brs	97.25	1.20	73.00	21.69	28.48
	12105	90.00	1.05	12.15	20.95	20.05
30000 ppm CaCl ₂	6 hrs	96.25	1.18	71.75	21.08	28.45
	12 hrs	96.25	1.20	69.75	20.40	28.45
S <i>x</i>		1.30	0.12	1.60	0.64	0.97
Sig. level		NS	NS	NS	NS	NS
CV (%)		2.81	11.24	5.82	8.10	7.65

Table 4. Interaction effects of priming agent and duration on germination and seedling vigor of BARI Gom-33

GP = germination percentage, MGT = mean germination time, GI = germination index, SVI = seedling vigour index, and GC = germination co-efficient, NS = Not significant

3.3 Seedling dry matter

Seed priming agent significantly affected (p<0.01) seedling root dry weight of BARI Gom-33 (Table 8). Priming with KCl produced the highest value; on the contrary, no advantages of priming with 75 ppm CuSO₄, 15000 ppm ZnSO₄, 10000 ppm ZnSO₄, and 50 ppm CuSO₄ was observed rather they produced statistically lower root dry weight than that of no priming (Table 8). Priming duration had no significant effect (p>0.01) on seedling root dry weight of BARI Gom-33 (Table 9). Also, no significant effect (p>0.01) was found for the interaction between priming agent and duration on root dry weight of BARI Gom-33 (Table 10).

An overall positive significant effect of seed priming was observed on shoot dry weight of BARI Gom-33. Seed priming with 10000 ppm CaCl₂, 20000 ppm CaCl₂ and, 50 ppm PEG produced the highest and statistically similar seedling shoot dry weight (Table 8). On the contrary, no advantage of 75 ppm CuSO₄, 15000 ppm ZnSO₄, and 50 ppm CuSO₄ priming was observed since they produced statistically lower seedling shoot dry weight than no priming (Table 8). Duration of priming also showed a significant effect (p<0.01) on seedling shoot dry weight, and 6 hrs priming was found better than 12 hrs priming (Table 8). No significant effect was found for the interaction between priming agent and duration on shoot dry weight of BARI Gom-33 (Table 10).

The seedling dry weight of BARI Gom-33 was significantly affected (p<0.01) by seed priming agents. Priming agents like KCl and 10000 ppm CaCl₂ produced the highest seedling dry weight while 75 ppm CuSO₄ and 15000 ppm ZnSO₄ produced the lowest seedling dry weight which was lower than no priming control (Table 8). Priming duration also exerted a significant effect on seedling dry weight, and priming for 6 hrs resulted in higher seedling dry weight than 12 hrs priming (Table 9). No significant effect for the interaction between priming agent and duration on the seedling dry weight of BARI Gom-33 was found (Table 10). Priming agent, priming duration, and their interaction failed to produce any significant effect on the root-shoot ratio of BARI Gom-33 (Tables 8 to 10).

4 Discussion

Seed germination rate, germination pattern, seedling growth, and vigor are very crucial for plant growth and productivity under both normal and adverse conditions like heat and moisture stresses. Crop plants often pass through a period of abiotic stresses during their life cycle under natural environments which adversely affect their growth and productivity (Hussain et al., 2018). As evident from different reports (Wahid et al., 2008; Patanè et al., 2009), seed priming provides plants with greater tolerance when exposed to stress. Therefore, it was, hypothesized that pre-sowing seed treatment could enhance germination, increase germination rate and improve seedling vigor and growth that could be helpful for wheat seedlings to overcome different abiotic stresses under late sown conditions which is now very common under Bangladesh context.

A positive impact of seed priming (except hydropriming and CuSO₄ priming) on seed germination percentage of wheat is evident from this study. In general, CaCl₂ priming performed the best. These findings are in harmony with those of many others who confirmed increased and faster germination along with synchronized emergence of primed seeds in wheat and other crop species (Khan et al., 2020; Kaczmarek et al., 2016; Anwar et al., 2020). Among the osmopriming agents CaCl₂ showed the best activity concerning seed germination, germination speed, and germination index of BRRI dhan40, BRRI dhan41, and BINA dhan7 (Islam et al., 2012).

According to Ajouri et al. (2004), priming induces a range of biochemical changes in the seed that is required for initiating the germination process i.e., breaking of dormancy, hydrolysis or metabolism of inhibitors, imbibition, and enzymes activation. Priming is an effective strategy to impart abiotic stress tolerance besides strengthening the defense line of crop plants. In seed priming, pre-sowing treatments are applied that control the hydration level within the seed and allow pre-germinative metabolic processes (physiochemical) to proceed while preventing radical emergence (Hussain et al., 2015a; Lutts et al., 2016; Zheng et al., 2015).

Plants that emerged from the primed seeds showed vigorous head start and higher stress tolerance mainly because of more effective energy metabolism, OA (Osmotic adjustment), quick cellular defense systems, enlarged embryo, and enhanced enzymatic activation (Jisha et al., 2012). Seed priming offers a smart, innovative, realistic, and effective option for achieving faster and uniform emergence, vigorous stand establishment, and higher productivity in crop plants under normal and stressful conditions (Hussain et al., 2019). It is also evident from some research work that priming with $CuSO_4$ treatments on oat (*Avena sativa* L.) seeds had no significant effects on germination speed (Iqbal, 2020).

This study verified that priming with $CaCl_2$ and PEG promoted seedling length most while $ZnSO_4$ priming inhibited seedling length. It is interesting to note that $ZnSO_4$ priming resulted in a shorter seedling length compared to control. Primed seedlings can produce normal seedlings under stress conditions. Ashraf and Foolad (2005) reported that under stress conditions primed seedlings can grow normally without any disturbance. Priming leaves a stress memory in the seed as the pre-germinative soaking process of seeds in the priming technique ex-

Priming agent	Root length (cm)	Shoot length (cm)	Seedling length (cm)
Control	7.200 fg	6.850 g	14.05 i
Hydropriming	8.125 f	7.787 f	15.91 h
10000 ppm NaCl	10.69 bcd	8.837 de	19.52 de
20000 ppm NaCl	10.70 bcd	8.375 ef	19.08 ef
30000 ppm NaCl	10.23 cde	7.600 f	17.83 fg
50 ppm PEG	11.68 ab	10.48 ab	22.15 ab
100 ppm PEG	11.63 ab	10.30 ab	21.92 abc
150 ppm PEG	10.86 bc	9.825 bc	20.69 cd
1 ppm Na ₂ MoO ₄	9.688 de	8.338 ef	18.02 fg
2 ppm Na ₂ MoO ₄	9.650 de	8.400 ef	18.05 fg
3 ppm Na ₂ MoO ₄	9.250 e	7.688 f	16.94 gh
5000 ppm $ZnSO_4$	5.300 ij	4.675 ij	9.9751
10000 ppm ZnSO ₄	4.838 ij	4.488 jk	9.325 lm
15000 ppm ZnSO ₄	4.412 j	3.787 k	8.200 m
25 ppm CuSO ₄	6.475 gh	5.850 h	12.32 j
50 ppm CuSO ₄	6.400 gh	5.338 hi	11.74 jk
75 ppm $CuSO_4$	5.600 hi	4.988 ij	10.59 kl
10000 ppm KCl	10.21 cde	9.300 cd	19.51 de
20000 ppm KCl	9.663 de	9.212 cd	18.88 ef
30000 ppm KCl	9.475 e	8.712 de	18.19 efg
10000 ppm CaCl ₂	12.07 a	10.96 a	23.04 a
20000 ppm CaCl ₂	11.49 ab	10.57 ab	22.06 ab
30000 ppm CaCl ₂	11.44 ab	10.11 b	21.55 bc
S <i>x</i>	0.34	0.26	0.44
Sig. level	**	**	**
CV (%)	10.68	9.39	7.29

Table 5. Effect of different priming agents on seedling growth of BARI Gom-33

** = Significant at 1% level of probability

Table 6. Effect of priming duration on seedling growth of BARI Gom-33

Priming duration	Root length (cm)	Shoot length (cm)	Seedling length (cm)
6 hrs	9.11	8.08 a	17.19 a
12 hrs	8.89	7.79 b	16.69 b
Sx	0.1	0.08	0.13
Sig. level	NS	**	**
CV (%)	10.68	9.39	7.29

** = Significant at 1% level of probability, NS = Not significant

hibits stress exposure that may improve the stress tolerance capacity in seeds during the post-germinative or seedling establishment period (Hussain et al., 2015b; Chakraborty and Dwivedi, 2021).

Seed priming with $CaCl_2$ performed better in comparison to control. Because Ca_2^+ increases cell-wall integrity and acts as a secondary messenger in signaling pathways of developmental and physiological processes, also Ca_2^+ icon decoded into downstream responses that ultimately lead to defense (Thor, 2019). The results of this study suggested that the osmopriming of wheat seeds with $CaCl_2$ would be more exacting than other priming agents. Vigorous seedling growth, emergence, and yield performance by CaCl₂ priming have also been reported in wheat under late sown conditions (Farooq et al., 2008).

Bose et al. (2018) stated that seed priming with PEG significantly improved the levels of photosynthetic pigments under abiotic stress conditions. Lee and Kim (2000) revealed that priming increased the metabolic activities of seed ultimately gained a substantial shoot length than non-primed seed. Osmopriming with PEG enhanced the ATPase activity in peanuts with substantial improvement in RNA syn-

Priming agent × prim	ning duration	Root length (cm)	Shoot length (cm)	Seedling length (cm)
Control	6 hrs	7.20	6.85	14.05
	12 hrs	7.20	6.85	14.05
Hydropriming	6 hrs	8.38	8.28	16.65
, , , ,	12 hrs	7.88	7.30	15.18
10000 ppm NaCl	6 hrs	10.80	8.78	19.58
11	12 hrs	10.58	8.90	19.48
20000 ppm NaCl	6 hrs	10.93	8.60	19.53
11	12 hrs	10.48	8.15	18.63
30000 ppm NaCl	6 hrs	10.33	7.98	18.30
11	12 hrs	10.13	7.23	17.35
50 ppm PEG	6 hrs	11.73	10.63	22.35
11	12 hrs	11.63	10.33	21.95
100 ppm PEG	6 hrs	11.68	10.40	22.08
11	12 hrs	11.58	10.20	21.78
150 ppm PEG	6 hrs	10.75	9.78	20.53
	12 hrs	10.98	9.88	20.85
1 ppm Na ₂ MoO ₄	6 hrs	9.95	8.50	18.45
- pp	12 hrs	9.43	8.18	17.60
2 ppm Na ₂ MoO ₄	6 hrs	9.63	8.53	18 15
2 ppin 1 40201004	12 hrs	9.68	8.28	17.95
3 ppm Na ₂ MoO ₄	6 hrs	9.45	7 93	17 38
5 ppm 1 40210004	12 hrs	9.05	7.45	16.50
	6 brs	5.28	1.83	10.10
5000 ppin 2050 ₄	12 hrs	5.33	4.53	9.85
10000 ppm ZnSQ.	6 hrs	4.88	4 70	9 58
10000 ppin 2004	12 hrs	4.80	4.28	9.08
15000 ppm ZnSQ.	6 hrs	4 60	3.83	8.43
15000 ppin 21504	12 hrs	4.23	3.75	7.98
25 ppm CuSO.	6 hrs	6 73	5 78	12 50
20 ppm CubO ₄	12 hrs	6.23	5.93	12.15
50 ppm CuSO.	6 hrs	6 50	5 55	12.05
50 ppin CubC ₄	12 hrs	6.30	5.13	11.43
 75 ppm CuSO.	6 hrs	5 73	4.88	10.60
75 ppin Cu504	12 hrs	5.48	5.10	10.58
10000 ppm KCl	6 hrs	10 30	9.63	19 93
10000 ppin Kei	12 hrs	10.13	8.98	19.10
20000 ppm KCl	6 hrs	9.83	9.40	19 23
20000 ppin Kei	12 hrs	9.50	9.03	18.53
30000 ppm KCl	6 brs	9 55	8 73	18 28
50000 ppin KCi	12 hrs	9.40	8.70	18.10
	6 hrs	12.25	11 15	22.40
10000 ppin CaCi ₂	12 hrs	11.90	10.78	22.68
20000 mmm CaCl	6 hrs	11.00	10.70	22.00
20000 ppm CaCl_2	o nrs 12 hrs	11.60	10.70	22.30 21.83
20000	(has	11.50	10.20	21.00
SUUUU ppm CaCl ₂	o nrs 12 hrs	11.53 11.35	10.38 9.85	21.90 21.20
	12 10 3	0.40	0.07	0.(1
Sx Sig level		0.48 NIS	U.37 NS	0.61 NS
CV (%)		10.68	9.39	7.29
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Table 7. Interaction effect of priming agent and duration on seedling growth of BARI Gom-33

NS = Not significant

Priming agent	Root dry matter	Shoot dry matter	Seedling dry matter	Root: Shoot
0 0	(mg)	(mg)	(mg)	
Control	3.675 i	3.525 k	7.200 i	0.927
Hydropriming	4.463 gh	4.175 j	8.637 h	0.941
10000 ppm NaCl	4.925 ef	5.275 f	10.20 fg	0.818
20000 ppm NaCl	4.863 fg	4.838 gh	9.700 g	0.856
30000 ppm NaCl	4.350 h	4.438 ij	8.788 h	0.856
50 ppm PEG	5.100 def	6.313 abc	11.41 c	0.722
100 ppm PEG	5.025 def	6.125 bcd	11.15 cd	0.73
150 ppm PEG	4.800 fg	5.700 e	10.50 ef	0.738
1 ppm Na ₂ MoO ₄	5.625 c	5.162 fg	10.79 def	0.937
$2 \text{ ppm Na}_2 \text{MoO}_4$	5.537 c	5.012 fg	10.55 ef	0.946
$3 \text{ ppm Na}_2 \text{MoO}_4$	5.325 cde	4.550 hi	9.875 g	1.02
5000 ppm ZnSO_4	3.388 ij	3.138 lm	6.525 jk	0.972
10000 ppm ZnSO ₄	3.150 jk	3.000 lmn	6.150 jkl	0.916
15000 ppm ZnSO ₄	2.888 kl	2.750 n	5.637 Îm	0.923
25 ppm CuSO ₄	3.425 ij	3.175 l	6.60 j	0.97
50 ppm CuSO_4	3.200 jk	2.800 mn	6.000 kl	0.986
75 ppm CuSO ₄	2.563 1	2.750 n	5.313 m	0.8
10000 ppm KCl	6.863 a	6.275 bc	13.14 a	0.94
20000 ppm KCl	6.525 ab	6.113 bcd	12.64 ab	0.941
30000 ppm KCl	6.325 b	5.787 de	12.11 b	0.952
10000 ppm CaCl ₂	5.450 cd	6.662 a	12.11 b	0.726
20000 ppm $CaCl_2$	4.988 ef	6.475 ab	11.46 c	0.705
30000 ppm CaCl_2	4.950 ef	6.088 cd	11.04 cde	0.721
S <i>x</i>	0.14	0.12	0.2	0.1
Sig. level	**	**	**	NS
CV (%)	8.66	7.07	5.89	31.16

Table 8. Effect of different priming agents on seedling dry matter of BARI Gom-33

** = Significant at 1% level of probability, NS = Not significant

Table 9. Effect of different	priming durations on	seedling dry matter of	of BARI Gom-33

Priming duration	Root dry matter (mg)	Shoot dry matter (mg)	Seedling dry matter (mg)	Root: Shoot
6 hrs	4.74	4.88 a	9.62 a	0.87
12 hrs	4.60	4.70 b	9.30 b	0.87
S <i>x̄</i>	0.04	0.04	0.06	0.03
Sig. level	NS	**	**	NS
CV (%)	8.66	7.07	5.89	31.16

** = Significant at 1% level of probability, NS = Not significant

theses and activity of acid phosphatase in the cotyledon and embryonic axis (Nawaz et al., 2013). In Arabidopsis seeds, osmopriming (-0.75 MPa PEG 6000) led to the accumulation of α tubulin and β tubulin (*Tubulin subunits*) (Kubala et al., 2015). Numerous research demonstrated that osmoprimed seeds are effective to improve germination, emergence, and seedling establishment of several plants, especially under stress conditions like ryegrass, sorghum, tomato, and wheat, Barley (Sun et al., 2010; Salehzade et al., 2009; Zhang et al., 2015; Amini, 2013; Amooaghaie and Nikzad, 2013). Baque et al. (2016) reported that the highest shoot length of wheat was secured when the seed was primed with PEG. Osmopriming of seeds with PEG has great potential to improve the stand establishment and performance of crop plants under stressful environments (Zhang et al., 2015; Salah et al., 2015; Aydinoğlu et al., 2019). Seeds treated with PEG6000 (-0.6 MPa) showed improved germination and healthy seedling growth, im-

Priming agent \times pr	riming duration	Root dry matter (mg)	Shoot dry matter (mg)	Seedling dry matter (mg)	Root: Shoot
Control	6 hrs	3.68	3.53	7.20	0.92
	12 hrs	3.68	3.53	7.20	0.92
Hydropriming	6 hrs	4.53	4.33	8.85	0.93
	12 hrs	4.40	4.03	8.43	0.95
10000 ppm NaCl	6 hrs	5.00	5.40	10.40	0.84
	12 hrs	4.85	5.15	10.00	0.80
20000 ppm NaCl	6 hrs	4.90	4.90	9.80	0.83
	12 hrs	4.83	4.78	9.60	0.89
30000 ppm NaCl	6 hrs	4.53	4.58	9.10	0.84
	12 hrs	4.18	4.30	8.48	0.87
50 ppm PEG	6 hrs	5.20	6.33	11.53	0.74
	12 hrs	5.00	6.30	11.30	0.71
100 ppm PEG	6 hrs	5.13	6.20	11.33	0.74
	12 hrs	4.93	6.05	10.98	0.72
150 ppm PEG	6 hrs	4.75	5.85	10.60	0.73
	12 hrs	4.85	5.55	10.40	0.75
1 ppm Na ₂ MoO ₄	6 hrs 12 hrs	5.70	5.30	11.00 10.58	0.94
2 ppm Na ₂ MoO ₄	6 hrs 12 hrs	5.58	5.15	10.73	0.94
3 ppm Na ₂ MoO ₄	6 hrs 12 hrs	5.48	4.63	10.10	1.01
5000 ppm ZnSO ₄	6 hrs	3.40	3.25	6.65	0.92
10000 ppm ZnSO ₄	6 hrs	3.15	3.05	6.20	0.91
15000 ppm ZnSO ₄	6 hrs 12 hrs	2.95	2.78	5.73	0.99
25 ppm CuSO ₄	6 hrs 12 hrs	3.63	3.20	6.83 6.38	1.00
50 ppm CuSO ₄	6 hrs 12 hrs	3.35 3.05	2.90 2.70	6.25 5.75	0.99 0.99
75 ppm CuSO ₄	6 hrs	2.70	2.88	5.58	0.78
	12 hrs	2.43	2.63	5.05	0.82
10000 ppm KCl	6 hrs	6.88	6.20	13.08	0.95
	12 hrs	6.85	6.35	13.20	0.93
20000 ppm KCl	6 hrs	6.55	6.23	12.78	0.93
	12 hrs	6.50	6.00	12.50	0.95
30000 ppm KCl	6 hrs	6.45	5.93	12.38	0.95
	12 hrs	6.20	5.65	11.85	0.95
10000 ppm CaCl ₂	6 hrs	5.45	6.80	12.25	0.71
	12 hrs	5.45	6.53	11.98	0.74
20000 ppm CaCl ₂	6 hrs	5.08	6.55	11.63	0.71
	12 hrs	4.90	6.40	11.30	0.70
30000 ppm CaCl ₂	6 hrs	5.05	6.23	11.28	0.72
	12 hrs	4.85	5.95	10.80	0.72
S <i>î</i>		0.20	0.17	0.28	0.14
Sig. level		NS	NS	NS	NS
CV (%)		8.66	7.07	5.89	31 16

Table 10. Interaction effect of different priming agents and duration on seedling dry matter of BARI Gom-33

NS = Not significant

proved root length, and antioxidant defense mechanism (Mouradi et al., 2016). The beneficial effects of PEG osmopriming were evident on root growth as reported by Patanè et al. (2009).

Priming with KCl produced the highest root dry weight. On the contrary, no advantage of $CuSO_4$ or $ZnSO_4$ priming was observed since they produced a statistically lower root dry weight than control. It is because of KCl is a strong stressor that affected dry matter of wheat (Natasha, 2021). A positive influence of seed priming on seedling growth (length and dry matter) was also reported by (Anwar et al., 2021), and KCl and CaCl₂ performed the best.

Seed priming with CaCl₂ and PEG produced the highest and statistically similar seedling shoot dry weight. On the contrary, CuSO₄ and ZnSO₄ priming were observed inhibiting the same since they produced lower seedling shoot dry weight than no priming. CaCl₂ showed better results in incrementing the shoot length of the Yard-long bean (Karim et al., 2020). The highest plumule length of BARI Gom-27 was obtained from seeds pre-treated with 10% PEG solution (Baque et al., 2016). Significant positive correlation between root and shoot length, root with root/shoot length ratio, and shoot with root/shoot length ratio were reported by Baloch et al. (2012) at 0 and 15% PEG stress levels. Seed priming with PEG (30%) significantly increased the shoot and root length as well as seedling fresh and dry biomass as revealed by Salah et al. (2015).

Priming agents KCl and CaCl₂ produced the highest seedling dry weight and CuSO₄ and ZnSO₄ produced the lowest seedling dry weight. Seed priming with KCl or CaCl₂ also improved the seedling growth, stand establishment as well yield performance in direct-seeded rice (Farooq et al., 2006). The K^+ is essential for activation of enzymes, for turgor and membrane potential balance, and in osmotic regulation in cells (Cherel, 2004). Priming improved the K⁺ balance that activates alpha-amylase, a basis for seed invigoration. A similar result was also obtained by Hasan et al. (2016) who reported that rice seeds primed with 5% CaCl₂ and 3% KCl for 24 hours produced the highest shoot dry mass of rice seedlings. Jisha et al. (2012) reported that the overall growth of plants was enhanced due to the seed-priming treatments. Maiti et al. (2013) observed that seed priming increases the seedling vigor of several vegetable crops. Seed germination, radicle/plumule emergence are inhibited by Cu_2^+ at high concentrations and Zn_2^+ at a different dose (Ahmed et al., 2021). Previous results demonstrated that seeds primed in 100 ppm CuSO₄ showed a reduction in seedlings shoot length, root length, seedling fresh biomass as compared to CuSO₄ at 200 ppm concentration (Iqbal, 2020).

CaCl₂ and PEG increased seed germination and early seedling growth in two wheat cultivars in both control and drought conditions (Asaduzzaman et al.,

2021). Chen et al. (2021) revealed that seed priming enhanced germination, seedling growth, and water relation behavior of wheat genotypes. All the characters showed the best results when wheat seeds were treated with 10% PEG solution. Osmopriming of rice seeds with KCl and CaCl₂ improved the emergence, emergence energy, and seedling emergence index (Farooq et al., 2006). Like K^+ , Ca_2^+ also plays very important roles in cell elongation and division, maintains cell wall integrity, regulates the uptake of nutrients across the membrane, and improves uptake of water in plants, and alleviates the adverse effect of Na⁺ during plant growth (Patade et al., 2009; Gobinathan et al., 2009). Germination speed is related to seedling vigor and it could be a significant determinant of good field performance (da Cruz and Milach, 2004). Priming of barley seeds with CaCl₂ improved drought tolerance attributed to enhanced transpiration rate without negative effects on the leaf turgor status and better stomatal aperture (Kaczmarek et al., 2016).

Intriguingly, the duration of priming had a significant effect on germination rate and seedling vigor in BARI Gom-33. The 6-hour priming duration showed a higher germination rate and seedling vigor compared to the 12-hour priming duration irrespective of concentration of any priming agent. As reported by Ibrahim (2019), priming agent and duration are very important factors that determine the germination success and seedling establishment.

5 Conclusion

In conclusion, present findings confirm the potentiality of seed priming to enhance the seed germination and seedling vigor of wheat, and pre-sowing seed priming with $CaCl_2$ for 6 hours was found the best for BARI Gom-33. Further detailed studies are required to explore the potentiality of seed priming to combat abiotic stresses like heat and moisture stresses in wheat under Bangladesh conditions.

Conflict of Interest

The authors declare that there is no conflict of interests regarding the publication of this paper.

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