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EFFECT OF SEED PRIMING ON GERMINATION, SEEDLING EMERGENCE AND DEVELOPMENT OF SPRING RICE *VAR*. HARDINATH-1

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ABSTRACT

Poor germination percentage and non-uniform seedling emergence and establishment are major causes of low rice yield in water-deficit arid ecosystems. A seed priming study with a low concentration of MoP, DAP, urea, vitamin C, ZnSO4, sucrose, NaCl, and hydro priming was conducted to assess the seed germination, seedling emergence, establishment, and seedling growth of the spring rice variety Hardinath-1. The experiment was conducted using a completely randomized design with three replications. Two test sets of experiments were carried out using blotting paper rolls and separate sand trays. Germination was observed at intervals of 24 hours up to 7 days, and germination and growth parameters were monitored. The results showed that germination (88.67%) and seedling emergence (88.67%) were high in vitamin C at 7 days after sowing (DAS). Germination energy (81%), germination speed (91.78%), and vigor index (91.78) were higher with seed priming of MoP. The lowest germination (56.44%) was observed with the priming of urea. At 30 DAS, the longest shoot length (19.93 cm) and longest root length (11.79 cm) were observed with the priming of ZnSO4. The highest seedling fresh weight (0.75 g) and dry weight (0.16 g), the highest number of leaves (3.57), and the longest leaf length (11.97 cm) were found with the priming of MoP, NaCl, and ZnSO4, respectively. However, the lowest seedling growth parameters were observed in the control priming. The overall results revealed that seed priming with MoP is an effective tool to enhance the germination percentage and seedling establishment of rice.

Keywords: seed priming; emergence; germination; Hardinath-1; seedlings

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INTRODUCTION

Rice is Nepal's major cereal crop in production and cultivated area (MoALD, 2021). Rice is grown in an area of 1.45 million ha, and production is 5.55 million Mt in Nepal (MoALD, 2021). Rice comes first among the cereals in Nepal, occupying about 41.22% of the total area under cereals crops and contributing about 49.9% of total consumable cereal production (MoALD, 2016). Rice production contributes 21% and 7% of AGDP and GDP, respectively of Nepal (MoF, 2020) and hence it has significant impacts on national economic growth, food security, and overall human welfare (Ghimire et al., 2021). Rice can be grown in two seasons in Nepal Spring and Monsoon.

However, the spring rice is not much popular in many parts of Nepal. It is mainly due to shortage of good variety seeds, poor seed germination and seedling establishment, and lack of technical knowledge among farmers that ultimately reduces the yield. Marginalized families with low income cannot afford to invest in irrigation and better seedling care for spring rice. It needs low-cost and low-risk technology for them. Agronomic practices such as seed priming, would overcome those issues and increase rice yields in low-cost investment.

Successful germination and seedling establishment are critical for optimum plant population maintenance and expansion, as well as recovery from arrangements (Noor-un-Nisa et al., 2013). Seed priming may be a key factor to rapid germination and emergence for a crop to be established successfully (Hussian et al., 2015). Seed priming is a pre-sowing approach that induce a physiological condition to promotes effective seed germination (Lutts, 2016). In essence, seed priming is a physiological process in which seeds are pre-soaked before planting, allowing for partial imbibition and regulating the seed's temperature and moisture content to get the seed closer to the germination stage (Choudhary, 2008; Nascimento, 2004).

Previous research stated that seed priming might enhance germination and establishment in numerous crops including maize, wheat, rice, sunflower and canola (Razaji et al., 2012; Basra et al., 2005). Farooq et al. (2007) stated that seed priming positively impacts germination, including boosting the rate, percentage, uniformity, and speed of germination as well as plant development, blooming, and yield (Du and Tuong, 2002). Water absorption by seeds is a critical step in seeds' germination and development because seed priming is dependent on seed imbibition behavior (Bewley and Black, 1994).

The present study aims to investigate how seed priming affects rice germination, emergence, and seedling growth in light of the problems associated with rice cultivation. Specific goals include achieving the successful and specific establishment of spring rice seedlings, identifying key factors such as germination percentage, energy, germination speed, and vigor index, and determining the most effective seed priming treatment.

MATERIALS AND METHODS

Experimental details

The experiment was set up in the Girija Prasad Koirala College of Agriculture and Research Center (GPCAR) laboratory in Gothgaun, Morang, Nepal, from February 2022 to March 2022. (Fig. 1). The experimental site's latitude, longitude, and elevation were 26°40′51″, 87°21′11″, and 143 meters above sea level. It is located in the tropical part of Nepal. The lab's typical temperatures and relative humidity (RH) for the experiment were 25 to 30 0C and 65 to 80%, respectively. The experiment was conducted using a Completely Randomized Design (CRD) with three replications of each treatment. Each blotting paper contained 100 seeds in a 10×10 square pattern, and after 7 DAS, 5×10 seedlings were transplanted in trays in a rectangular pattern. Each replication contains precisely 100 seeds, and spacing was maintained at 1.5-2 cm (seed to seed distance) and 3-3.5 cm (seedlings to seedlings distance). Therefore, the experiment contains a total of 27 setups. Treatments were prepared by mixing 4 gm of each treatment in 200 ml of water and setting seeds in the solution (Table 1). Approximately 100 seeds were taken from each of the nine replications for the germination test. Furthermore, all seeds were primed in treatment solutions for 18h and allowed to air dry for 3h before sowing. Seeds were sown in blotting paper and checked for germination every 24h for up to 7 days. After 7 DAS, 50 seedlings from each replication of nine treatments are randomly selected and transplanted in trays. At the interval of 10 days, the ten seedlings were selected randomly, uprooted, analyzed, and destroyed. Trays used in the research were made of plastic with a depth of 5 cm, respectively which were equal in size with equal volume, and the soil was filled in each tray. Primed seedlings were transplanted at 2 cm deep and irrigated daily throughout the research.

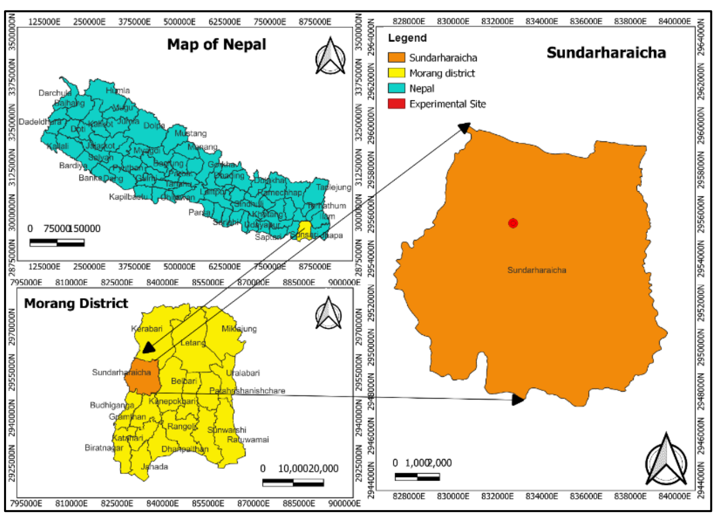

Figure 1. Experimental site of the research.

Treatment details

Following treatments were utilized in the experiment (Table 1):

Table 1. List of treatment used in the research.

S.N.	Treatment	Concentration of solutions	Treatment symbols
1	Muriate of Potash (MOP)	2%	T1
2	Di-ammonium Phosphate (DAP)	2%	T2
3	Urea	2%	T3
4	Zinc Sulphate ($ZnSO_4$)	2%	T4
5	Ascorbic acid or Vitamin C	2%	T5
6	Sucrose	2%	T6
7	Sodium Chloride (NaCl)	2%	T7
8	Distilled water (dH_2O)	-	T8
9	Control	-	T9

Data collection

Germination percentage (GP)

Following the start of the experiment, the number of seeds germinated was recorded every day for the first seven days. Seeds with an identifiable radicle were judged to have germinated, while seeds with an identifiable cotyledon were taken to have emerged. The following formula was used to calculate the GP:

$$Germination\ percentage\ (\%) = \frac{No. of\ seeds\ germinated}{Total\ no. of\ seeds\ sown} \times 100$$

Germination speed (GS)

Generally, the total number of seeds germinating during a specific period is called the GS. The total number of seeds that germinated each day was included in the computation of seed GS and was calculated using the method below:

$$Germination\ speed\ (\%) = \frac{No. of\ seeds\ germinated\ in\ 72\ h}{No. of\ seed\ germinated\ in\ 168\ h} \times 100$$

Germination energy (GE)

GE is defined as the percentage by which the number of seeds germinates within a definite period in a given sample. GE was calculated using the following formula:

$$Germination\ energy\ (\%) = Percentage\ of\ seed\ germinated\ in\ 72\ h$$

Vigor index (VI)

VI provides an illustration of a seed's capacity to control its degree of activity and performance throughout germination and emergence. The following formula may be used to get the seed VI:

$$Vigor\ Index\ (VI) = \frac{X1}{N1} + \frac{X2}{N2} + \cdots\cdots\cdots\cdots + \frac{Xn}{Nn}$$

Where,

X1 = No. of seedlings at 1st count

X2 = No. of seedlings at 2nd count

Xn = No. of seedlings at final count

N1 = No. of days at 1st count

N2 = No. of days at 2nd count

Nn = No. of days at final count

Emergence percentage (EP)

The emergence percentage is the total amount of seedlings that emerged in a seed lot expressed in percentage. It can be calculated by using the following formula:

$$Emergence\ percentage\ (\%) = \frac{No.of\ seedlings\ emerged}{Total\ no.of\ seed\ set\ for\ test} \times 100$$

Growth parameters

Using a scale, the root length (RL) and shoot length (SL) of seeds from all treatments and replications were measured from the beginning of the primary root to the top of the plant at 10, 20, and 30 days after sowing (DAS). To determine the weight of seedlings at 10, 20, and 30 DAS, ten fresh plants were uprooted and weighed on an electronic balance for their fresh weight (FW). Subsequently, the same ten plants were dried at 165°C in a hot air oven to obtain their dry weight (DW). Additionally, at 30 DAS, ten plants from each treatment were assessed for both leaf number (LN) and leaf length (LL).

Statistical analysis

Microsoft Excel was used to input, tabulate, and analyze the acquired data (2019 version). Gen Stat (18th Edition) was used to evaluate the collected data for the various parameters statistically. Duncan's Multiple Range Test (DMRT) separated the means at a 5% significance level.

RESULTS AND DISCUSSION

Performances of treatments on germination parameters

Analysis of variance shows that GP was highly significant at a 1% significance level, as shown in (Table 2). At 3 DAS, the GP was high in MOP and low in urea priming. At 5 DAS, the GP was high in DAP and low in urea priming. At 7 DAS, the vitamin C group had a higher GP than the control group, which was followed by MoP, DAP, ZnSO4, hydroponic priming with distilled water, and then NaCl and sucrose priming. They were significantly different from each other. Further, analysis of variance shows that GS, GE, and VI were highly significant at a 1% level of significance. GE was recorded more in MoP and less in urea priming. Seed primed with other treatments differed significantly. GS was recorded more in seeds treated with MoP solution (91.78%) and low in seeds treated with urea (48.51%), followed by speed of DAP, hydro priming, and vitamin C (Fig. 2). VI was more in MoP-treated seeds (91.78) and less in seed treated by 2% urea solution (60.34), followed by DAP, vitamin C and hydro priming (distilled water), which were significantly similar.

Table 2. Mean performances of treatments on germination percentage, germination speed, germination energy, and vigor index in spring rice variety

Treatments	Germination percentage (GP)			Germination speed (GS)	Germination energy (GE)	Vigor index (VI)
	3 DAS	5 DAS	7 DAS			
2% MOP	81.00^{a}	84.00^{a}	88.33^{a}	91.78^{a}	81.00^{a}	91.78^{a}
2% DAP	67.67^{b}	85.33^{a}	88.00^{a}	76.80^{b}	67.67^{b}	86.83^{ab}
2% Urea	35.67^{f}	61.00^{c}	73.33^{b}	48.51^{e}	35.67^{f}	60.34^{e}
2%Vitamin C	64.67^{bc}	83.33^{a}	88.67^{a}	72.97^{bc}	64.67^{bc}	84.83^{b}
2% $ZnSO_4$	54.00^{de}	80.00^{ab}	86.67$_{a}$	62.38^{d}	54.00^{de}	76.52^{d}
2% Sucrose	57.00^{cd}	73.00^{b}	82.33^{a}	69.14^{bcd}	57.00^{cd}	75.03^{d}
2% NaCl	55.33^{de}	81.33^{ab}	83.33^{a}	66.41^{cd}	55.33^{de}	78.53^{cd}
Hydro priming	64.67^{bc}	81.00^{ab}	85.00^{a}	76.10$_{bc}$	64.67^{bc}	82.97^{bc}
Control	48.00^{e}	62.67^{c}	67.33^{c}	71.58^{bcd}	48.00^{e}	63.29^{e}
LSD	7.957	8.093	5.784	9.164	7.957	5.937
Mean±SE	58.67±3.78	76.85±3.85	82.56±2.75	70.63±4.36	58.67±3.79	77.79±2.83
CV%	7.9	6.1	4.1	7.6	7.9	4.4
F-test	**	**	**	**	**	**

**Significant at 1% level of significance, LSD: Least significant difference, SE: Standard error, CV: Coefficient of variance

Performances of treatments on growth parameters

Analysis of variance shows that the emergence percentage was highly significant at a 1% level of significance, as shown in (Table 3). The EP of vitamin C priming was higher (88.67%) than the other treatments. Furthermore, it was significantly lower in control (67.33%) at 7 DAS, followed by the emergence percentages of MoP, DAP, ZnSO4, hydropriming, NaCl, and sucrose, which were all significantly lower. Likewise, leaf length was highly significant at 1% level of significance, and leaf number was non-significant. At 30 DAS, LL was observed more in ZnSO4 and less in urea, followed by MoP, sucrose, vitamin C and hydro priming (distilled water), which were statistically similar to each other (Fig. 3). At 30 DAS, more leaves were observed in NaCl priming, and fewer leaves were observed in control. All treatments were significantly at par.

Table 3. Mean performances of treatments on emergence percentage, leaf number, and leaf length in spring rice variety at 30 DAS.

Treatments	Emergence %	Leaf number at 30DAS	Leaf length at 30DAS
2% MOP	88.33[a]	3.53[a]	11.37[ab]
2% DAP	88.00[a]	3.33[a]	10.00[cd]
2% Urea	73.33[b]	3.43[a]	9.30[d]
2% Vitamin C	88.67[a]	3.53[a]	11.34[ab]
2% ZnSO4	86.67[a]	3.53[a]	11.97[a]
2% Sucrose	82.33[a]	3.33[a]	11.35[ab]
2% NaCl	83.33[a]	3.57[a]	10.25[bcd]
Hydro priming	85.00[a]	3.53[a]	11.23[abc]
Control	67.33[c]	3.26[a]	10.81[abc]
LSD	5.784	0.51	1.152
Mean±SE	82.56± 2.75	3.44±0.24	10.85±0.54
CV%	4.1	8.6	6.2
F-test	**	NS	**

**Significant at 1% level of significance, NS: Non-significant, LSD: Least significant difference, SE: Standard error, CV: Coefficient of variance

Root length was non-significant at 10 DAS and highly significant at 0.1% and 1% level of significance at 20 and 30 DAS, respectively (Table 4). At 10 DAS, the highest RL was found in the control and the lowest RL was found in the MoP priming. At 20 DAS, the highest RL was observed in $ZnSO_4$ (9.93 cm) followed by hydro priming (9.42 cm), NaCl (9.39 cm), control (8.77 cm), vitamin C (8.74 cm), sucrose (8.14 cm), urea (8.01 cm), MoP (7.32 cm), and DAP (7.14 cm). At 30 DAS, the highest RL (11.79 cm) was observed in $ZnSO_4$ whereas in control the RL was (11.49 cm) (Fig. 3). Further, shoot length was significantly at par at a 5% significance level. In 10 DAS, the most extended SL was observed in control seedlings and the shortest in $ZnSO_4$ seedlings. The highest SL was found in DAP priming and the lowest SL was found in sucrose priming at 20 DAS. At 30 DAS, the highest SL was observed in $ZnSO_4$ and the lowest SL was found in control which was non-significant with each other.

Table 4. Mean performances of treatments on root length and shoot length in spring rice variety.

Treatments	Root length (cm)			Shoot length (cm)		
	10 DAS	20 DAS	30 DAS	10 DAS	20 DAS	30 DAS
2% MOP	6.12[a]	7.32[d]	10.65[c]	10.17[a]	14.25[a]	19.48[a]
2% DAP	6.14[a]	7.14[d]	10.59[c]	10.12[a]	14.28[a]	18.53[a]
2% Urea	6.61[a]	8.01[cd]	11.04[bc]	10.07[a]	13.79[a]	18.34[a]
2% Vitamin C	7.14[a]	8.74[bc]	11.46[ab]	10.29[a]	14.05[a]	19.49[a]
2% ZnSO4	6.93[a]	9.93[a]	11.79[a]	10.00[a]	13.93[a]	19.93[a]
2% Sucrose	6.83[a]	8.14[cd]	11.17[abc]	10.01[a]	13.30[a]	18.68[a]
2% NaCl	7.05[a]	9.39[ab]	11.65[ab]	10.31[a]	13.86[a]	18.60[a]
Hydro priming	7.19[a]	9.42[ab]	11.70[ab]	10.13[a]	14.02[a]	19.57[a]
Control	7.20[a]	8.77[bc]	11.49[ab]	10.73[a]	13.56[a]	17.98[a]
LSD	0.976	0.845	0.650	0.724	0.898	1.850
Mean±SE	6.80±0.64	8.54±0.46	11.28±0.31	10.20±0.34	13.89±0.43	18.96±0.88
CV%	8.4	6.7	3.4	4.1	3.8	5.7
F-test	NS	***	**	NS	NS	NS

**Significant at 1% level of significance, ***Significant at 0.1% level of significance, NS: Non-significant, LSD: Least significant difference, SE: Standard error, CV: Coefficient of variance

Fresh weight was non-significant, as shown in (Table 5). At 10 DAS, the highest FW was found in $ZnSO_4$, whereas the lowest FW was observed in hydro priming. At 20 DAS, the highest FW was measured in DAP and the lowest in NaCl. At 30 DAS, the highest FW was obtained with MoP priming and the lowest FW was recorded. Moreover, the analysis of variance shows that DW was non-significant in 10 and 30 DAS and significantly different at a 5% level of significance at 20 DAS. At 20 DAS, the highest DW was observed in Vitamin C priming and the lowest DW was observed in control. At 30 DAS, the highest DW was recorded with MoP priming, whereas the lowest was in hydro priming (Fig. 3).

Seed priming is a widely used technique in agriculture that involves the pre-sowing treatment of seeds with various compounds to enhance their germination, seedling vigor, and crop performance (Anwar et al., 2020). Seeds treated with MoP (KCl, the main component of MoP) outperformed the others, and the yield of rice seeds treated with MoP would surely increase. Bam (2006) reported that seeds immersed in KCl or KH2PO4 solutions showed higher dehydrogenase activity than seeds steeped in water, indicating that they were more vivacious and viable. This finding agrees with our conclusion that seed priming with MoP solution improved seedling characteristics. Koirala (2019) demonstrated that seed priming with a 2% MoP solution was an effective tactic for boosting the germination, emergence, and seedling characteristics of the spring rice variety Hardinath-1. Our study also found that 2% MoP primed seeds performed well. Farooq et al. (2007) evaluated various priming methods and found that traditional soaking, hydroponic priming, osmo hardening with KCl or CaCl2, vitamin priming, and seed hardening were all effective. Choudhary et al. (2021) reported that seed priming with Mg(NO3)2 and ZnSO4 improved growth attributes under field conditions, consistent with our results. Tuiwong et al. (2022) confirmed that priming seeds with N and Zn solutions before planting could improve seedling growth performance. Singhal and Bose (2021) found that seed priming with a combination of Mg(NO3)2 and ZnSO4 salt was superior to single salt and non-priming treatments for improving wheat crop qualities under drought stress. Bukhari (2021) supported our findings that Zinc Sulphate outperformed the control group regarding germination, emergence, and growth.

Table 5. Mean performances of treatments on fresh weight and dry weight in spring rice variety.

Treatments	Fresh weight (g)			Dry weight (g)		
	10 DAS	20 DAS	30 DAS	10 DAS	20 DAS	30 DAS
2% MOP	0.46^{ab}	0.55^{a}	0.75^{a}	0.14^{a}	0.14^{b}	0.16^{a}
2% DAP	0.42^{bc}	0.57^{a}	0.71^{ab}	0.13^{a}	0.14^{b}	0.16^{a}
2% Urea	0.44^{abc}	0.55^{a}	0.67^{ab}	0.14^{a}	0.15^{b}	0.16^{ab}
2% Vitamin C	0.43^{abc}	0.53^{a}	0.65^{b}	0.14^{a}	0.15^{a}	0.15^{ab}
2% ZnSO4	0.46^{a}	0.55^{a}	0.69^{ab}	0.13^{a}	0.15^{ab}	0.16^{ab}
2% Sucrose	0.45^{ab}	0.54^{a}	0.66^{ab}	0.14^{a}	0.14^{ab}	0.16^{ab}
2% NaCl	0.42^{abc}	0.51^{a}	0.67^{ab}	0.14^{a}	0.14^{a}	0.15^{b}
Hydro priming	0.41^{c}	0.56^{a}	0.63^{b}	0.13^{a}	0.15^{b}	0.16^{ab}
Control	0.44^{abc}	0.56^{a}	0.64^{b}	0.14^{a}	0.14^{ab}	0.15^{b}
LSD	0.08	0.49	0.08	0.01	0.01	0.01
Mean±SE	0.68 ±0.04	0.55±0.23	0.68 ±0.04	0.14 ±0.01	0.14 ±0.01	0.16±0.01
CV%	7.1	5.19	7.1	2.0	2.1	2.6
F-test	NS	NS	NS	NS	*	NS

*Significant at 5% level of significance, NS: Non-significant, LSD: Least significant difference, SE: Standard error, CV: Coefficient of variance

Additionally, Khan et al. (2014) demonstrated that treatment with certain pesticides, including phorate, carbosulfan, and chlorpyriphos, led to significant increases in rice plant growth 15-30 days after planting. These findings were supported by our own study, which showed that seed priming with DAP performed well in enhancing seedling growth. Another effective seed priming approach is the use of vitamin C treatment, as demonstrated by Ghimire et al. (2021). This study found that seeds treated with vitamin C exhibited better performance than those subjected to alternating soaking and drying or other treatments. The use of vitamin C may enhance seedling vigor by reducing oxidative stress and increasing the activity of enzymes responsible for various metabolic processes. In conclusion, seed priming is an important technique for enhancing crop yield and improving plant growth. The use of various priming methods, including treatment with pesticides, vitamins, and other compounds, can help to improve seedling vigor and ultimately lead to better crop performance. These findings may be applied to a range of different crops and can be used to develop effective seed priming strategies for agriculture.

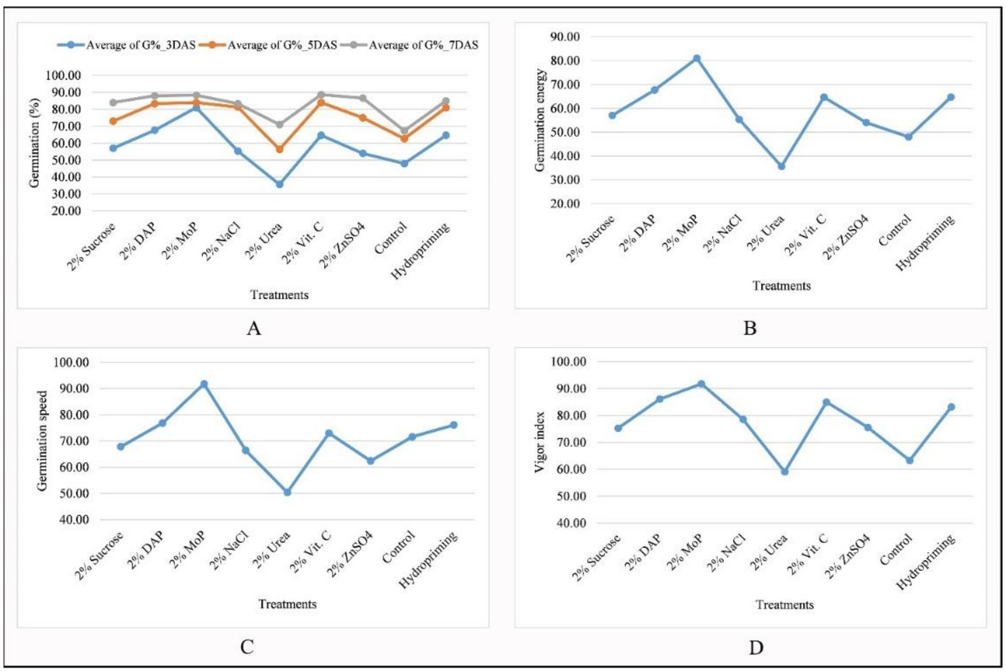

Figure 2. Effects of different treatments on germination parameters; (A) Germination percentage, (B) Germination energy, (C) Germination speed, and (D) Vigor index.

Figure 3. Effect of different treatments on growth parameters; (A) Emergence percentage, (B) Leaf length & number, (C) Root length, (D) Shoot length, (E) Fresh weight, and (F) Dry weight.

CONCLUSION

This study showed significant variations among the different treatments concerning different seed and seedling growth parameters. At 7 DAS, vitamin C had the highest germination percentage; it also had the highest germination energy, speed, and vigor index; MoP priming had the highest fresh and dry weight, and vitamin C had the highest emergence percentage. At 30 DAS, ZnSO4 priming produced the longest shoot, root and leaf lengths, while NaCl priming produced the most leaves. 2% urea solution and control resulted in the lowest germination and seedling parameters.

Increased seedling vigor at an early stage of seedling growth would help to increase final crop yield by ensuring a better stand. The suitable treatment could create a favorable environment for the growth and development of rice seedlings. More studies may be required to suggest the best possible technique of seed priming in rice by other priming treatments, too, evaluating all the growth parameters and final yield. As a result, seed priming with 2% MoP solution for 18h is effective for the spring rice variety Hardinath-1, followed by 2% Vitamin C, ZnSO4, and hydro priming with distilled water.

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