



Effect of different chemical priming agents on physiological and morphological characteristics of rice (*Oryza sativa* L.)

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ABSTRACT

The research was conducted following a Completely Randomized Design (CRD) to investigate the effects of seed priming using various chemical treatments on the germination and growth parameters of two rice cultivars, BRRI Dhan-66 and IR 80991-B-330-U-1. Ten different priming agents, including H_3BO_3 , $CaCO_3$, $CuSO_4$, DAP, $FeCl_2$, MoP, PEG (5 %), PEG (10 %), Urea, and $ZnSO_4$, were applied to treat the seeds, each treatment being replicated three times. A control group underwent hydro-priming. The seeds were soaked in the treatments for 24 h. After the priming treatments, the seeds were subjected to a redrying process at a temperature of $26 \pm 2^\circ C$ until they regained their original weight before being transplanted onto blotting paper. Germination parameters such as germination percentage, germination speed, germination energy, and vigor index were recorded for seven consecutive days. Growth parameters including root length, shoot length, fresh seedling weight, and dry seedling weight were measured at 10, 20, and 30 days after sowing. The results indicate significant variations among the treatments for germination parameters ($p \leq 0.001$). Similarly, significant variations were observed in growth parameters, including shoot length, fresh weight, and dry weight ($p \leq 0.001$). Among the rice varieties, BRRI Dhan-66 exhibited better results for germination percentage (81.58 %), germination speed (62.78 %), germination energy (52.06 %), vigor index (1312), fresh weight (0.807g), and dry weight of seedlings (0.053g). In contrast, the $FeCl_2$ treatment showed the best results, inducing respective increases of 25.19 %, 93.35 %, 94.95 %, and 29.07 % for germination percentage, speed, energy, and vigor index compared to the control, respectively. For growth parameters, the DAP and $CuSO_4$ treatments demonstrated better results. Our findings highlight that improved germination of primed rice seedlings is associated with germination energy, speed, vigor index, and the fresh weight of the seedlings. Furthermore, Pearson's correlation coefficient revealed there is significant positive correlation between germination percentage, speed, energy and vigor index but the strongest correlation exists between germination speed and germination energy ($R = 0.94^{***}$) followed by germination percentage and vigor index ($R = 0.92^{***}$). Based on our findings, we propose that seed priming significantly enhances rice seedlings' germination

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and growth parameters. BRRI Dhan-66, along with seed treatment using FeCl_2 , can be effectively employed to achieve improved germination and growth in rice cultivation.

1. Introduction

Seed germination is a critical stage in a plant's life cycle, transforming a dormant seed into a healthy seedling. This process involves water absorption, degradation of macromolecules, genetic material repair, embryo and endosperm expansion, and seed coat rupture, culminating in a radicle's emergence. Successful germination significantly impacts early seedling growth and final yield [1]. Seed priming, proposed by Ref. [2], emerges as a practical solution to mitigate drought stress in rice cultivation. This technique enhances rapid and uniform emergence, seedling vigor, and yield in adverse conditions [3]. While various factors can hinder seed germination, priming techniques offer simple interventions to improve it [4]. In rice, issues like chilling, moisture stress, and weed interference led to low germination, weak seedlings, and yield loss [5].

Rice (*Oryza sativa* L.), a fundamental cereal crop, transcends mere sustenance to become an emblem of global nourishment. With over half of the world's population relying on its grains, rice serves as an indispensable staple for sustenance and survival [6]. In the context of Nepal, this cereal holds an even more profound significance, intertwining with the nation's agricultural sector and contributing vitally to its economic landscape. The expanse of Nepal's cultivated land, covering 42.5% (168,047 ha), is dedicated to rice cultivation, yielding 51.6% of the country's total food grain output [MoAD, 2017]. The gravity of rice's role within Nepal's economy is undeniable, rippling across sectors and communities, and deeply ingrained in the fabric of everyday life [7].

Poor and irregular germination is the main issue facing rice producers, which reduces rice yield and causes a large financial loss [8]. Abiotic stress has a key role in reducing rice crop germination rates and yield losses overall. Drought, salt, and very high temperatures are a few more important issues [9]. The primary rice-growing technique used worldwide is transplanting, in which rice is grown from a nursery seedling and then planted in a field that has been flooded. The seeds utilized are nursery-raising pre-germinated varieties, which exhibit poor and delayed germination. Uneven crop stand and an uncontrolled plant population are two important yield-limiting problems for this strategy [10]. Reactive oxygen species (ROS) are produced during abiotic stress, which causes physiological and biochemical alterations in seedlings. As a result, the membranes are harmed, cells leak, and the photosynthetic components are destroyed [9,11]. Such issues have arisen while boosting plant and seedling resistance to germination and abiotic stress using various techniques, such as plant breeding and the creation of transgenics. Among these, seed priming is a simple, low-risk, and affordable technique for enhancing plant and seedling growth and development under challenging environmental circumstances [9].

The regulated hydration method known as "seed priming" offers a practical and essential approach to increasing the crop's emergence, seedling vigor, and stress tolerance [11,12]. It aids in proper seed germination under adverse climatic circumstances. It is a cheap and low-risk option. Rice's tolerance capacity and osmotic stress resistance are reported to be improved by primed seeds' physiological and biochemical alterations [13]. Primed seeds have high emergence capacity, emerge more quickly, and produce a better, more consistent, and robust crop stand. They continue to operate in less-than-ideal field circumstances. Compared to unprimed seeds, primed seeds sprouted, resulting in early blooming and a larger crop yield [14]. By preventing lipid peroxidation during germination, priming strategies also aid in boosting the degree of antioxidant defense enzyme activity. Salts, polyamines, hormones, suitable solutes, and plant extracts are only a few priming agents that have been employed and are advantageous [15]. Pre-enlargement of the embryo and other biochemical changes, including enzyme activation, rapid creation of emerging metabolites, and structural and genetic repair, are the main benefits of seed priming [16]. Conclusively, seed priming helps to improve the agronomic qualities of rice without degrading the quality of harvested paddy [17].

This research study aims to systematically investigate and compare the effects of various priming agents on two distinct rice cultivars, BRRI Dhan-66 and IR 80991-B-330-U-1. Moreover, the central hypothesis of this study is that various seed priming agents influence the germination and growth parameters of rice seedlings. Further, the specific objectives include evaluating the impact of these priming agents on germination percentage, germination speed, germination energy, seedling vigor, and growth parameters. By investigating how different priming agents influence these critical parameters, this research seeks to contribute to the understanding of sustainable approaches to enhance rice cultivation amidst challenging environmental conditions. In doing so, it not only addresses the immediate concerns of crop yield but also aligns with the broader goals of securing food supplies and fostering economic stability.

2. Materials and Methodology

2.1. Experimental site

The experimental design employed in this study was a Completely Block Design (CBD) with two factors: Two varieties and eleven different priming treatments, each being replicated three times. The research was conducted at the G. P. Koirala College of Agriculture and Research Centre laboratory, situated in Sundarharaicha, Morang, Nepal, with geographical coordinates of 26.4° North and 87.21° East latitude. Detailed meteorological data throughout the research period are depicted in Fig. 1.

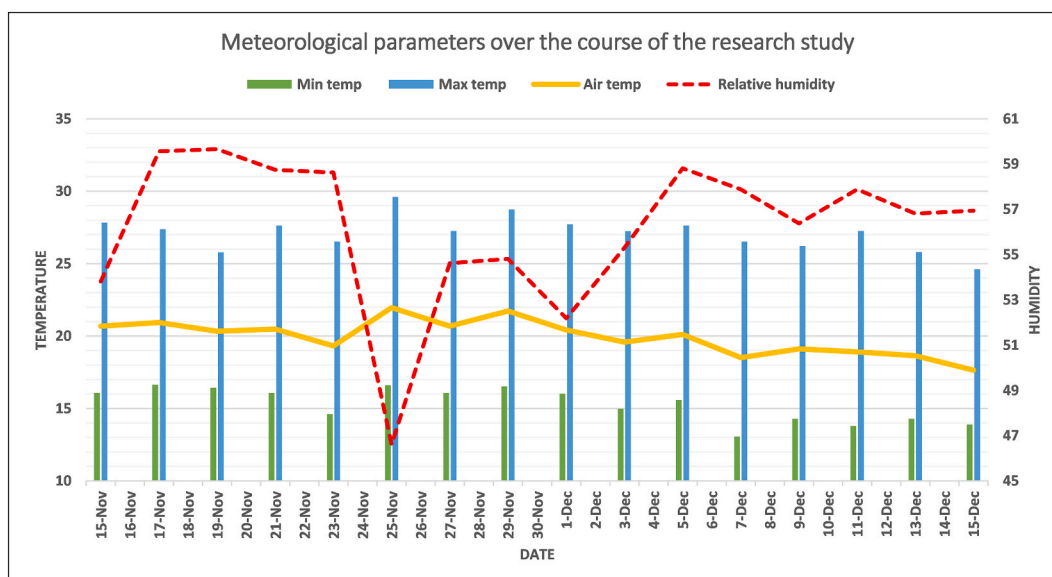


Fig. 1. Meteorological parameters over the course of the research study.

2.2. Plant materials

For the study, two varieties of rice, namely BRRI Dhan-66 and IR 80991-B-330-U-1, were selected. These seeds were procured from the Nepal Agriculture Research Council (NARC) in Tarahara, Sunsari, Nepal.

2.3. Seed priming treatments

The research followed the Completely Randomized Design (CRD) for conducting the experiment. This involved incorporating ten distinct priming agents, alongside a control group that underwent hydro-priming (HP). Each of these treatments was replicated three times to ensure the reliability of the findings. The specific priming agents used in the study and their corresponding doses are outlined in Table 1. To create the priming solutions, the recommended dose for each treatment was mixed with 500 ml of water. The duration of 24 h for priming was selected, as it has been widely employed in previous research and demonstrated effectiveness in enhancing germination and seedling growth in rice [10]. The doses of the priming agents were determined based on prior studies that have investigated their impacts on seed germination and seedling traits. These studies have been properly cited and are listed in Table 1. After undergoing the priming treatments, the seeds underwent a redrying process at a temperature of 26 ± 2 °C until they regained their original weight, following the approach described by Ref. [18]. This redrying process typically lasted around 2–3 h. Subsequently, the seeds were placed on blotting paper for further experimentation. For the control group, the seeds were soaked in distilled water for the same duration. The germination test involved placing 100 seeds from each treatment onto blotting paper, which was then regularly moistened using a water sprayer. The blotting paper containing the seeds was kept in a germination room with temperature regulation set within the range of 20–30 °C. The count of emerged seedlings was recorded daily for seven days. On the eighth day, 25 seedlings from each treatment were transplanted into a seedling tray filled with a mixture of soil and vermicompost in a 5:1 ratio. This was done to monitor and evaluate their subsequent growth performance.

Table 1
Several priming agents utilized in the experimental research.

S. N.	Priming agents	Doses	Reference
1	Control (Hydro priming)	N/A	[19]
2	Polyethylene glycol (PEG)	10 %	[20]
3	Polyethylene glycol (PEG)	5 %	[21]
4	Diammonium phosphate (DAP)	2 %	[22]
5	Boric acid (H_3BO_3)	0.1 %	[23]
6	Urea	2 %	[22]
7	Cupric sulfate ($CuSO_4$)	2 %	[24]
8	Muriate of potash	2 %	[22]
9	Zinc sulfate ($ZnSO_4$)	2 %	[25]
10	Calcium carbonate ($CaCO_3$)	2 %	[26]
11	Ferrous chloride ($FeCl_2$)	2 %	[27]

2.4. Data collection and observation

Various germination and growth parameters were observed during the experiment. The number of seed germinations was recorded seven consecutive days following sowing to assess germination parameters. Additionally, 25 seedlings from each replication were randomly selected and transplanted into trays. Five seedlings were randomly selected, uprooted, analyzed, and destructively assessed every ten days to study growth parameters.

2.4.1. Germination parameters

The germination parameters evaluated in this experiment included germination percentage, speed, energy, and vigor index. Germination percentage is the proportion of viable seeds that successfully sprout and develop into plants under optimal growing conditions. Germination speed is the number of seeds germinating during a specific period. Germination energy is the percentage of seeds that germinate within a definite period in a given sample. The vigor index is the total of the properties of the seed that determine its potential level of activity and performance during germination and emergence. The formulas used to calculate these germination parameters are as follows: Eq. (1) for germination percentage [28], Eq. (2) for germination speed [29], Eq. (3) for germination energy [22], and Eq. (4) for vigor index [30].

$$\text{Germination Percentage (G\%)} = \frac{\text{Number of seed germinated}}{\text{Total number of seed sown}} \times 100 \quad (1)$$

$$\text{Germination Speed (GS)} = \frac{\text{Number of seed germinated in 72 hours}}{\text{Number of seed germinated in 168 hours}} \times 100 \quad (2)$$

$$\text{Germination Energy (GE)} = \% \text{ of seed germinated in 72 hours} \quad (3)$$

$$\text{Vigor Index (VI)} = \text{Seedling length} \times \text{Germination Percentage (G\%)} \quad (4)$$

2.4.2. Growth parameters

To assess the growth parameters of the rice seedlings, root and shoot length were measured using a measuring scale. Five seedlings were randomly selected, uprooted, and measured on days 10, 20, and 30 after sowing. Additionally, the fresh weight of the seedlings was measured by weighing five fresh seedlings using an electronic weighing machine. Finally, the dry weight of the seedlings was calculated by weighing five dry seedlings dried by the air-dry method.

2.5. Statistical analysis

The collected data were entered into MS-Excel (2019 version) and subsequently analyzed using R-studio software (Version 4.2.2). Verification for normal distribution was conducted on the data. Analysis was performed within a completely randomized design framework, utilizing analysis of variance. To compare the means of individual parametric data sets, the Duncan Multiple Range Test (DMRT) was applied as a statistical method with a significance level set at $p \leq 0.05$ [31]. Furthermore, the interaction effect between varieties and treatments was explored using R-studio software. Pearson's correlation coefficient was also calculated using various R-studio packages to examine relationships among different panicle traits [3]. Additionally, Principal Component Analysis (PCA) was carried out on various traits to uncover underlying sources of variability [4]. The gathered data, outcomes of statistical analysis, and significant findings were methodically organized and presented in tabular format. This presentation was achieved using appropriate formatting tools available in MS Word.

3. Results

3.1. Effect of priming agents on germination parameters

Table 2 presents the impact of various priming agents on germination parameters. Notably, seeds primed with FeCl_2 exhibited the highest germination percentage, germination speed percentage, and germination energy percentage. Conversely, the control group, without seed priming, showed the lowest values for these parameters. Vigor index values aligned with these trends, with FeCl_2 -primed seeds displaying the highest, and control seeds showing the lowest vigor index.

Interaction effects between seed priming agents and varieties were assessed. While the germination percentage displayed a non-significant interaction, germination speed percentage and germination energy percentage exhibited significant interactions. For variety BRRI Dhan-66, FeCl_2 priming yielded the highest germination percentage (92.67), while CaCO_3 priming resulted in the highest germination speed percentage (82.78). Likewise, FeCl_2 priming led to the highest germination percentage and germination speed percentage for variety IR 80991-B. The highest germination energy percentage was recorded for BRRI Dhan-66 seeds primed with FeCl_2 , as well as for IR 80991-B seeds primed with FeCl_2 . Control seeds for variety BRRI Dhan-66 showed the lowest values for germination percentage, germination speed percentage, and germination energy percentage. For variety IR 80991-B, the lowest germination percentage was associated with PEG (5 %) priming (55.33), while both germination speed percentage and germination energy percentage were lowest for the control. Consistent with trends in germination parameters, vigor index results were highest for

Table 2

Effect of different priming agents and their interaction on different germination parameters of rice.

Variety		Germination (%)	Germination speed (%)	Germination energy (%)	Vigor index
BRR1 Dhan-66		81.58	62.78	52.06	1312
IR 80991-B		65.52	51.06	33.94	1082
Grand Mean		73.55	56.92	43.00	1197
SEM±		1.578	1.838	1.572	28.6
LSD _{0.05}		3.180	3.705	3.169	57.7
F-test		***	***	***	***
Treatments					
H ₃ BO ₃ (0.1 %)		69.33 ^{bcd}	61.95 ^{bc}	43.33 ^c	1159 ^{cdef}
CaCO ₃ (2 %)		75.67 ^{bc}	69.51 ^{ab}	53.67 ^b	1233 ^{bcd}
CuSO ₄ (2 %)		70.00 ^{bcd}	59.7 ^c	41.67 ^{cd}	1077 ^{ef}
DAP (2 %)		77.33 ^b	69.53 ^{ab}	54.67 ^b	1316 ^{ab}
FeCl ₂ (2 %)		87.33 ^a	75.11 ^a	66.00 ^a	1431 ^a
MoP (2 %)		75.00 ^{bc}	54.51 ^{cd}	42.33 ^{cd}	1197 ^{bcd}
PEG (10 %)		75.33 ^{bc}	58.66 ^{cd}	45.00 ^c	1250 ^{bc}
PEG (5 %)		69.00 ^{bcd}	59.79 ^c	40.33 ^{cd}	1154 ^{cdef}
Urea (2 %)		68.00 ^{cd}	50.08 ^d	34.33 ^d	1094 ^{def}
ZnSO ₄ (2 %)		76.67 ^b	62.28 ^c	48.33 ^{bc}	1239 ^{bcd}
Control (HP)		65.33 ^d	4.99 ^e	3.33 ^e	1015 ^f
SEM±		3.701	4.311	3.687	67.1
LSD _{0.05}		7.459	8.689	7.431	135.3
F-test		***	***	***	***
Interaction among varieties and treatments					
BRR1 Dhan-66	H ₃ BO ₃ (0.1 %)	77.33 ^{bcd}	66.40 ^{defg}	51.33 ^{de}	1326 ^{abc}
	CaCO ₃ (2 %)	84.67 ^{abc}	82.78 ^a	70.00 ^{ab}	1368 ^{ab}
	CuSO ₄ (2 %)	74.00 ^{cdefg}	55.5 ^{efg}	41.33 ^{ef}	1127 ^{cdef}
	DAP (2 %)	84.67 ^{abc}	81.1 ^{ab}	68.67 ^{abc}	1434 ^a
	FeCl ₂ (2 %)	92.67 ^a	81.13 ^{abc}	75.33 ^a	1455 ^a
	MoP (2 %)	86.67 ^{ab}	67.62 ^{bdef}	58.67 ^{cd}	1400 ^{ab}
	PEG (10 %)	86.67 ^{ab}	65.46 ^{defg}	56.67 ^d	1407 ^{ab}
	PEG (5 %)	82.67 ^{abcd}	53.52 ^{fgh}	44.00 ^{ef}	1364 ^{ab}
	Urea (2 %)	72.67 ^{cdefgh}	59.99 ^{defg}	43.33 ^{ef}	1107 ^{def}
	ZnSO ₄ (2 %)	83.33 ^{abcd}	70.47 ^{abcd}	58.67 ^{bcd}	1328 ^{abc}
	Control (HP)	72.00 ^{defgh}	6.55 ^j	4.67 ⁱ	1115 ^{cdef}
IR 80991-B	H ₃ BO ₃ (0.1 %)	61.33 ^{hij}	57.50 ^{defg}	35.33 ^{fgh}	992 ^{def}
	CaCO ₃ (2 %)	66.67 ^{ghij}	56.23 ^{defg}	37.33 ^{fg}	1098 ^{def}
	CuSO ₄ (2 %)	66.00 ^{ghij}	63.8 ^{defg}	42.00 ^{ef}	1027 ^{def}
	DAP (2 %)	70.00 ^{ghi}	57.94 ^{defg}	40.67 ^{ef}	1198 ^{bcd}
	FeCl ₂ (2 %)	82.00 ^{abcde}	69.09 ^{bcd}	56.67 ^d	1407 ^{ab}
	MoP (2 %)	63.33 ^{ghij}	41.40 ^{hi}	26.00 ^{gh}	993 ^{def}
	PEG (10 %)	64.00 ^{ghij}	51.87 ^{ghi}	33.33 ^{fgh}	1094 ^{def}
	PEG (5 %)	55.33 ^j	66.07 ^{defg}	36.67 ^{fgh}	944 ^{ef}
	Urea (2 %)	63.33 ^{ghij}	40.17 ⁱ	25.33 ^h	1082 ^{def}
	ZnSO ₄ (2 %)	70.00 ^{efghi}	54.09 ^{fgh}	38.00 ^f	1151 ^{cde}
	Control (HP)	58.67 ^{ij}	3.42 ^j	2.00 ⁱ	915 ^f
CV (%)		8.7	13.1	14.9	9.7
SEM±		5.234	6.097	5.215	94.9
LSD _{0.05}		10.548	12.287	10.509	191.3
F-test		NS	***	***	*

Treatments means with distinct alphabetical letters signifies significant differences determined by the Duncan Multiple Range Test (DMRT) at $P \leq 0.05$. *Significant at 5 % level of significance, ***Significant at 0.1 % level of significance, ^{NS}Non-significant, LSD: Least significant difference, SEM: Standard error of the mean, CV: Coefficient of difference, DAS: Days after sowing. H₃BO₃: Boric Acid, CaCO₃: Calcium Carbonate, CuSO₄: Cupric Sulfate, DAP: Diammonium Phosphate, FeCl₂: Ferrous Chloride, MOP: Muriate of Potash, PEG: Polyethylene Glycol, ZnSO₄: Zinc Sulfate, HP: Hydro-priming.

FeCl₂-primed seeds and lowest for control seeds, in both BRR1 Dhan-66 and IR 80991-B varieties.

3.2. Effect of priming agents on growth parameters

Priming effects on shoot length were evident in rice seedlings of both varieties. At 10 days after sowing (DAS), priming enhanced shoot length in BRR1 Dhan-66, while at 20 and 30 DAS, IR 80991-B showed greater shoot length (Table 3). A similar trend was observed for root length, where IR 80991-B consistently exhibited greater values at 10, 20, and 30 DAS compared to BRR1 Dhan-66. Primarily, 10 % PEG treatment led to notable root length increase in all seedlings, surpassing other priming agents' effects.

The interaction effect between seed priming agent and variety was not significant for root and shoot length (Table 3). Specifically, in BRR1 Dhan-66, shoot length increase was notable with DAP treatment up to 30 DAS, while FeCl₂ priming exhibited a gradual rise. For IR 80991-B, PEG (5 %) treatment led to heightened shoot length, and a similar trend was seen with MOP treatment up to 30 DAS.

Table 3
Effect of different priming agents and their interaction on shoot and root length of rice.

Variety	Shoot length (cm)			Root length (cm)				
	10 DAS	20 DAS	30 DAS	10 DAS	20 DAS	30 DAS		
BRR1 Dhan-66	2.955	13.59	15.55	3.011	5.994	7.08		
IR 80991-B	2.105	13.64	17.08	3.320	6.205	7.24		
Grand Mean	2.530	13.62	16.32	3.165	6.100	7.16		
SEM±	0.127	0.399	0.380	0.170	0.197	0.241		
LSD _{0.05}	0.256	0.804	0.766	0.343	0.397	0.486		
F-test	***	NS	***	NS	NS	NS		
Treatments								
H ₃ BO ₃ (0.1 %)	2.437 ^{bcd}	14.09 ^a	16.55 ^{abc}	3.477 ^a	6.237 ^a	7.300 ^a		
CaCO ₃ (2 %)	2.900 ^{abc}	13.99 ^a	15.86 ^{abc}	3.333 ^a	5.897 ^a	7.107 ^a		
CuSO ₄ (2 %)	2.062 ^{de}	12.51 ^a	15.09 ^c	2.695 ^a	6.367 ^a	7.470 ^a		
DAP (2 %)	2.647 ^{abcd}	14.27 ^a	17.77 ^a	3.422 ^a	5.810 ^a	7.077 ^a		
FeCl ₂ (2 %)	3.227 ^a	13.65 ^a	15.46 ^{bc}	3.370 ^a	6.280 ^a	7.283 ^a		
MoP (2 %)	2.963 ^{ab}	13.04 ^a	15.27 ^c	3.460 ^{ab}	6.107 ^a	7.077 ^a		
PEG (10 %)	2.890 ^{abc}	13.16 ^a	16.88 ^{abc}	3.353 ^a	6.107 ^a	7.570 ^a		
PEG (5 %)	2.423 ^{bcd}	14.20 ^a	17.45 ^{ab}	3.103 ^a	6.135 ^a	7.157 ^a		
Urea (2 %)	2.233 ^{cd}	13.43 ^a	17.01 ^{abc}	2.658 ^a	6.373 ^a	6.963 ^a		
ZnSO ₄ (2 %)	2.450 ^{bcd}	14.30 ^a	16.12 ^{abc}	2.847 ^a	5.820 ^a	7.037 ^a		
Control (HP)	1.597 ^e	13.17 ^a	16.02 ^{abc}	3.100 ^a	5.963 ^a	6.713 ^a		
SEM±	0.298	0.935	0.892	0.399	0.462	0.566		
LSD _{0.05}	0.601	1.885	1.797	0.804	0.932	1.140		
F-test	***	NS	*	NS	NS	NS		
Interaction among varieties and treatments								
BRR1 Dhan-66	H ₃ BO ₃ (0.1 %)	3.080 ^{abcd}	15.28 ^a	16.13 ^{abcd}	3.060 ^{ab}	6.707 ^a	7.300 ^a	
	CaCO ₃ (2 %)	3.413 ^{abc}	13.81 ^a	14.82 ^{bcd}	3.540 ^{ab}	6.013 ^a	6.893 ^a	
	CuSO ₄ (2 %)	2.700 ^{bcd}	12.36 ^a	14.18 ^{cd}	2.680 ^{ab}	6.493 ^a	7.300 ^a	
	DAP (2 %)	2.933 ^{abcde}	14.11 ^a	18.47 ^a	2.840 ^{ab}	5.833 ^a	6.593 ^a	
	FeCl ₂ (2 %)	3.807 ^a	12.95 ^a	13.84 ^d	3.353 ^{ab}	6.007 ^a	7.120 ^a	
	MoP (2 %)	3.507 ^{ab}	13.74 ^a	14.94 ^{bcd}	3.447 ^{ab}	6.067 ^a	6.780 ^a	
	PEG (10 %)	3.140 ^{abcd}	12.71 ^a	15.61 ^{abcd}	3.453 ^{ab}	5.800 ^a	7.900 ^a	
	PEG (5 %)	2.833 ^{bcd}	14.27 ^a	16.41 ^{abcd}	2.767 ^{ab}	5.943 ^a	7.280 ^a	
	Urea (2 %)	2.667 ^{bcd}	12.33 ^a	15.94 ^{abcd}	2.347 ^b	5.820 ^a	6.533 ^a	
	ZnSO ₄ (2 %)	2.593 ^{bcd}	13.97 ^a	14.81 ^{bcd}	2.787 ^{ab}	5.907 ^a	7.753 ^a	
	Control (HP)	1.827 ^{fghi}	14.01 ^a	15.96 ^{abcd}	2.847 ^{ab}	5.347 ^a	6.447 ^a	
	IR 80991-B	H ₃ BO ₃ (0.1 %)	1.793 ^{ghi}	12.91 ^a	16.97 ^{abc}	3.893 ^a	5.767 ^a	7.30 ^a
		CaCO ₃ (2 %)	2.387 ^{defgh}	14.18 ^a	16.90 ^{abc}	3.127 ^{ab}	5.780 ^a	7.320 ^a
CuSO ₄ (2 %)		1.423 ^{hi}	12.65 ^a	16.01 ^{abcd}	2.710 ^{ab}	6.240 ^a	7.640 ^a	
DAP (2 %)		2.360 ^{defgh}	14.44 ^a	17.06 ^{abc}	4.003 ^a	5.787 ^a	7.560 ^a	
FeCl ₂ (2 %)		2.647 ^{bcd}	14.35 ^a	17.08 ^{abc}	3.387 ^{ab}	6.553 ^a	7.447 ^a	
MoP (2 %)		2.420 ^{cdefg}	12.34 ^a	15.59 ^{abcd}	3.473 ^{ab}	6.147 ^a	7.373 ^a	
PEG (10 %)		2.640 ^{bcd}	13.61 ^a	18.15 ^a	3.253 ^{ab}	6.413 ^a	7.240 ^a	
PEG (5 %)		2.013 ^{efghi}	14.13 ^a	18.49 ^a	3.440 ^{ab}	6.327 ^a	7.033 ^a	
Urea (2 %)		1.800 ^{ghi}	14.53 ^a	18.07 ^a	2.970 ^{ab}	6.927 ^a	7.393 ^a	
ZnSO ₄ (2 %)		2.307 ^{defghi}	14.63 ^a	17.44 ^{ab}	2.907 ^{ab}	5.733 ^a	6.320 ^a	
Control (HP)		1.367 ⁱ	12.32 ^a	16.09 ^{abcd}	3.353 ^{ab}	6.580 ^a	6.980 ^a	
CV (%)		20.4	11.9	9.5	21.8	13.1	13.7	
SEM±		0.422	1.323	1.261	0.564	0.654	0.800	
LSD _{0.05}	0.850	2.666	2.542	1.137	1.318	1.612		
F-Test	NS	NS	NS	NS	NS	NS		

Treatments means with distinct alphabetical letters signifies significant differences determined by the Duncan Multiple Range Test (DMRT) at P ≤ 0.05. *Significant at 5 % level of significance, ***Significant at 0.1 % level of significance, ^{NS}Non-significant, LSD: Least significant difference, SEM: Standard error of the mean, CV: Coefficient of difference, DAS: Days after sowing. H₃BO₃: Boric Acid, CaCO₃: Calcium Carbonate, CuSO₄: Cupric Sulfate, DAP: Diammonium Phosphate, FeCl₂: Ferrous Chloride, MOP: Muriate of Potash, PEG: Polyethylene Glycol, ZnSO₄: Zinc Sulfate, Hydro-priming.

Regarding root length, BRR1 Dhan-66 responded positively to PEG (10 %) treatment, while the control group exhibited slow growth up to 30 DAS. Similarly, IR 80991-B's root length increased notably with CuSO₄ priming, and a gradual increase was seen with ZnSO₄ treatment up to 30 DAS.

The impact of different seed priming treatments on fresh weight and dry weight of the seedlings is evident from Table 4. MOP-primed seeds displayed the highest fresh seedling weight at 10 DAS, while control and other treatments exhibited comparatively lower weights. At 20 DAS, control seeds showed the highest fresh weight, and PEG (10 %) priming yielded the highest fresh weight at 30 DAS. The dry weight of seedlings at 10 DAS was notably higher with CuSO₄ priming, while urea (2 %) treatment led to the highest dry weight at 20 DAS. Urea priming resulted in the highest dry weight at 30 DAS.

The interaction effect between seed priming agents and variety was found to be significant for both fresh weight and dry weight (Table 4). BRR1 Dhan-66 seedlings exhibited the highest fresh weight when MOP-primed, and the lowest with ZnSO₄ priming. At 20

Table 4
Effect of different priming agents and their interaction on fresh and dry weight of rice.

Variety	Fresh weight of seedling (g)			Dry weight of seedling (g)			
	10 DAS	20 DAS	30 DAS	10 DAS	20 DAS	30 DAS	
BRR1 Dhan-66	0.394	0.545	0.807	0.022	0.030	0.053	
IR 80991-B	0.342	0.448	0.533	0.018	0.026	0.039	
Grand Mean	0.368	0.496	0.670	0.020	0.028	0.046	
SEM±	0.0134	0.0155	0.037	0.00081	0.0014	0.0026	
LSD _{0.05}	0.0270	0.0313	0.076	0.00163	0.0028	0.0053	
F-test	***	***	***	***	**	***	
Treatments							
H ₃ BO ₃ (0.1 %)	0.344 ^{ab}	0.503 ^{ab}	0.660 ^b	0.020 ^{bc}	0.029 ^{bcd}	0.046 ^{bcd}	
CaCO ₃ (2 %)	0.404 ^a	0.435 ^b	0.698 ^{ab}	0.020 ^{bc}	0.023 ^d	0.049 ^{bcd}	
CuSO ₄ (2 %)	0.345 ^{ab}	0.491 ^{ab}	0.598 ^b	0.022 ^b	0.034 ^b	0.043 ^{bcd}	
DAP (2 %)	0.401 ^a	0.485 ^{ab}	0.711 ^{ab}	0.021 ^{bc}	0.026 ^{cd}	0.051 ^{abc}	
FeCl ₂ (2 %)	0.396 ^a	0.503 ^{ab}	0.655 ^b	0.017 ^c	0.022 ^d	0.038 ^{cde}	
MoP (2 %)	0.415 ^a	0.499 ^{ab}	0.549 ^b	0.017 ^c	0.025 ^{cd}	0.033 ^e	
PEG (10 %)	0.363 ^a	0.510 ^{ab}	0.863 ^a	0.017 ^c	0.031 ^{bc}	0.057 ^{ab}	
PEG (5 %)	0.362 ^a	0.487 ^{ab}	0.580 ^b	0.019 ^{bc}	0.025 ^{cd}	0.041 ^{cde}	
Urea (2 %)	0.289 ^b	0.486 ^{ab}	0.595 ^b	0.029 ^a	0.041 ^a	0.063 ^a	
ZnSO ₄ (2 %)	0.383 ^a	0.515 ^{ab}	0.599 ^b	0.016 ^c	0.024 ^{cd}	0.037 ^{de}	
Control (HP)	0.345 ^{ab}	0.547 ^a	0.856 ^a	0.020 ^{bc}	0.026 ^{cd}	0.043 ^{bcd}	
SEM±	0.0314	0.0364	0.088	0.0019	0.0033	0.0061	
LSD _{0.05}	0.0634	0.0734	0.179	0.0038	0.0066	0.0123	
F-test	**	NS	**	***	***	***	
Interaction among varieties and treatments							
BRR1 Dhan-66	H ₃ BO ₃ (0.1 %)	0.345 ^{bcd}	0.556 ^{bc}	0.794 ^{bc}	0.021 ^{bcd}	0.032 ^{bcd}	0.051 ^{bcd}
	CaCO ₃ (2 %)	0.447 ^{ab}	0.473 ^{bcd}	0.785 ^{bcd}	0.022 ^{bcd}	0.024 ^{cdef}	0.057 ^{bcd}
	CuSO ₄ (2 %)	0.392 ^{abcd}	0.504 ^{bcd}	0.672 ^{bcd}	0.022 ^{bcd}	0.035 ^{bc}	0.042 ^{defgh}
	DAP (2 %)	0.446 ^{ab}	0.579 ^b	0.885 ^b	0.025 ^{bc}	0.028 ^{cdef}	0.069 ^{ab}
	FeCl ₂ (2 %)	0.381 ^{abcd}	0.486 ^{bcd}	0.737 ^{bcd}	0.017 ^{efg}	0.021 ^f	0.041 ^{defgh}
	MoP (2 %)	0.470 ^a	0.580 ^b	0.653 ^{bcd}	0.018 ^{defg}	0.029 ^{cdef}	0.038 ^{efgh}
	PEG (10 %)	0.395 ^{abcd}	0.592 ^b	1.246 ^a	0.021 ^{bcd}	0.041 ^{ab}	0.080 ^a
	PEG (5 %)	0.352 ^{bcd}	0.488 ^{bcd}	0.604 ^{bcd}	0.022 ^{bcd}	0.026 ^{cdef}	0.048 ^{cdefgh}
	Urea (2 %)	0.394 ^{abcd}	0.488 ^{bcd}	0.654 ^{bcd}	0.024 ^{bcd}	0.032 ^{bcd}	0.061 ^{bcd}
	ZnSO ₄ (2 %)	0.342 ^{bcd}	0.549 ^{bcd}	0.631 ^{bcd}	0.019 ^{cdefg}	0.027 ^{cdefg}	0.044 ^{defgh}
	Control (HP)	0.369 ^{abcd}	0.700 ^a	1.214 ^a	0.026 ^b	0.034 ^{bcd}	0.052 ^{bcd}
IR 80991-B	H ₃ BO ₃ (0.1 %)	0.343 ^{bcd}	0.449 ^{cdef}	0.527 ^{cde}	0.019 ^{cdefg}	0.026 ^{cdef}	0.041 ^{defgh}
	CaCO ₃ (2 %)	0.361 ^{bcd}	0.397 ^{ef}	0.611 ^{bcd}	0.018 ^{efg}	0.022 ^{ef}	0.041 ^{defgh}
	CuSO ₄ (2 %)	0.299 ^d	0.477 ^{bcd}	0.524 ^{cde}	0.022 ^{bcd}	0.033 ^{bcd}	0.044 ^{defgh}
	DAP (2 %)	0.356 ^{bcd}	0.392 ^f	0.536 ^{cde}	0.017 ^{efg}	0.024 ^{def}	0.034 ^{fgh}
	FeCl ₂ (2 %)	0.411 ^{abc}	0.521 ^{bcd}	0.573 ^{cde}	0.016 ^{efg}	0.022 ^{ef}	0.036 ^{fgh}
	MoP (2 %)	0.361 ^{bcd}	0.417 ^{ef}	0.445 ^e	0.015 ^{fg}	0.022 ^{ef}	0.029 ^h
	PEG (10 %)	0.332 ^{cd}	0.428 ^{def}	0.481 ^{de}	0.021 ^f	0.021 ^f	0.034 ^{fgh}
	PEG (5 %)	0.372 ^{abcd}	0.486 ^{cdef}	0.556 ^{cde}	0.017 ^{efg}	0.024 ^{cdef}	0.033 ^{fgh}
	Urea (2 %)	0.185 ^e	0.484 ^{cdef}	0.536 ^{cde}	0.034 ^a	0.050 ^a	0.066 ^{abc}
	ZnSO ₄ (2 %)	0.424 ^{abc}	0.481 ^{cdef}	0.568 ^{cde}	0.014 ^g	0.022 ^{ef}	0.031 ^{gh}
	Control (HP)	0.322 ^{cd}	0.394 ^f	0.498 ^{cde}	0.014 ^g	0.019 ^f	0.034 ^{fgh}
CV (%)		14.8	12.7	23.0	16.3	19.9	22.9
SEM±		0.044	0.0515	0.1257	0.0026	0.0046	0.0086
LSD _{0.05}		0.089	0.1038	0.2532	0.0054	0.0093	0.0174
F-Test		**	***	***	***	***	**

Treatments means with distinct alphabetical letters signifies significant differences determined by the Duncan Multiple Range Test (DMRT) at P ≤ 0.05. **Significant at 1 % level of significance, ***Significant at 0.1 % level of significance, ^{NS}Non-significant, LSD: Least significant difference, SEM: Standard error of the mean, CV: Coefficient of difference, DAS: Days after sowing. H₃BO₃: Boric Acid, CaCO₃: Calcium Carbonate, CuSO₄: Cupric Sulfate, DAP: Diammonium Phosphate, FeCl₂: Ferrous Chloride, MOP: Muriate of Potash, PEG: Polyethylene Glycol, ZnSO₄: Zinc Sulfate, Hydro-priming.

DAS, control seeds showed the highest fresh weight for this variety, contrasting with the lowest for CaCO₃ priming. PEG (10 %) priming yielded the highest fresh weight at 30 DAS, while PEG (5 %) yielded the lowest. Regarding dry weight, BRR1 Dhan-66 seedlings displayed the highest when kept under control, and the lowest with FeCl₂ priming. At 20 DAS, PEG (10 %) priming resulted in the highest dry weight, in contrast with the lowest observed with FeCl₂ priming. DAP priming yielded the highest dry weight at 30 DAS, while MOP priming yielded the lowest. For variety IR80991-B, ZnSO₄ priming led to the highest fresh weight at 10 DAS, while urea treatment resulted in the lowest. FeCl₂ priming yielded the highest fresh weight at 20 DAS, while DAP priming led to the lowest. CaCO₃ priming yielded the highest fresh weight at 30 DAS, while MOP yielded the lowest. Similarly, for dry weight, urea treatment resulted in the highest at 10 DAS, and ZnSO₄, PEG (10 %), and control yielded the lowest. Urea treatment also led to the highest dry weight at 20 DAS, while control yielded the lowest. At 30 DAS, CuSO₄ priming resulted in the highest dry weight, while MOP priming led to the lowest.

3.3. Correlation and PCA analysis

Correlation heat map analysis was performed to evaluate the relationships among all measured traits to explore the effects of different priming agents on germination and growth parameters (Fig. 2). The analysis revealed that germination percentage, germination speed, germination energy, and vigor index showed a positive correlation with all traits except root and shoot length. Furthermore, root length also exhibited a positive correlation with shoot length and dry weight, while shoot length was negatively correlated with both fresh and dry weight. According to Pearson's correlation coefficient, the results indicate that germination percentage was significantly and positively correlated with germination speed ($R = 0.48^{***}$), germination energy ($R = 0.73^{***}$), vigor index ($R = 0.92^{***}$), fresh weight ($R = 0.34^{**}$), and dry weight ($R = 0.32^{**}$), whereas it was significantly negatively correlated with shoot length ($R = -0.36^{**}$). Similarly, germination speed was significantly and positively correlated with germination energy ($R = 0.94^{***}$) and vigor index ($R = 0.51^{***}$). Likewise, germination energy exhibited a significant positive correlation with vigor index ($R = 0.73^{***}$). The vigor index was also significantly positively correlated with fresh weight ($R = 0.31^*$) and dry weight ($R = 0.32^{**}$). Furthermore, a significant positive correlation was found between fresh weight and dry weight ($R = 0.51^{***}$).

Principal Component Analysis (PCA) was conducted to investigate variations in germination and growth traits (Table 5). The results revealed that three principal components (PCs) had eigenvalues greater than one (Fig. 3). PC1 exhibited the highest variation, with germination energy (0.490) having the highest loading, followed by germination percentage (0.486), vigor index (0.477), and germination speed (0.403). PC2 had the most substantial loading in fresh weight (0.648), followed by dry weight (0.532). Similarly, PC3 had the highest loading in shoot length (0.699), followed by root length (0.604). The biplot depicting PCA1 and PCA2 displayed distinct patterns among the varieties and treatments for germination and growth traits (Fig. 4). The first two PCA axes accounted for 61.1 % of the total variation. Based on the PCA biplot, five interactions were clustered in the first quadrant (BRR1 Dhan-66 \times PEG (10 %), H_3BO_3 , PEG (5 %), DAP, and $CaCO_3$), effectively separated from others based on fresh weight, dry weight, germination percentage, and vigor index. Likewise, four interactions were grouped in the second quadrant (IR 80991-B and BRR1 Dhan-66 \times Control and Urea). Eight interactions were also grouped in the third quadrant based on shoot length (IR 80991-B \times PEG (10 %), H_3BO_3 , $CuSO_4$, PEG (5 %), $ZnSO_4$, MoP, DAP, and $CaCO_3$). Lastly, five interactions were clustered in the fourth quadrant, effectively differentiated from others based on germination energy, germination speed, and root length (IR 80991-B \times $FeCl_2$ and BRR1 Dhan-66 \times $CuSO_4$, $ZnSO_4$, MoP, and $FeCl_2$). Considering the contribution of each measured variable, it is evident that germination energy (red) followed by germination percentage (light red), germination speed, vigor index, and fresh weight (orange) had the most significant impact on the applied treatments (Fig. 5).

4. Discussions

4.1. Effect of priming agent on germination parameter

Germination of seeds is a pivotal stage that significantly influences the final grain yield of rice, and it is subject to regulation by a multitude of abiotic and biotic factors. Among these factors, drought stress has been identified to profoundly impede germination rate, seedling growth, and starch metabolism in rice cultivars. Intriguingly, these stress conditions lead to heightened antioxidant enzyme activity and lipid peroxidation, indicating the plant's defense response [32]. To address these challenges, seed priming has emerged as a promising technique for achieving rapid and uniform seed germination. Priming treatments exert their effects by eliciting various

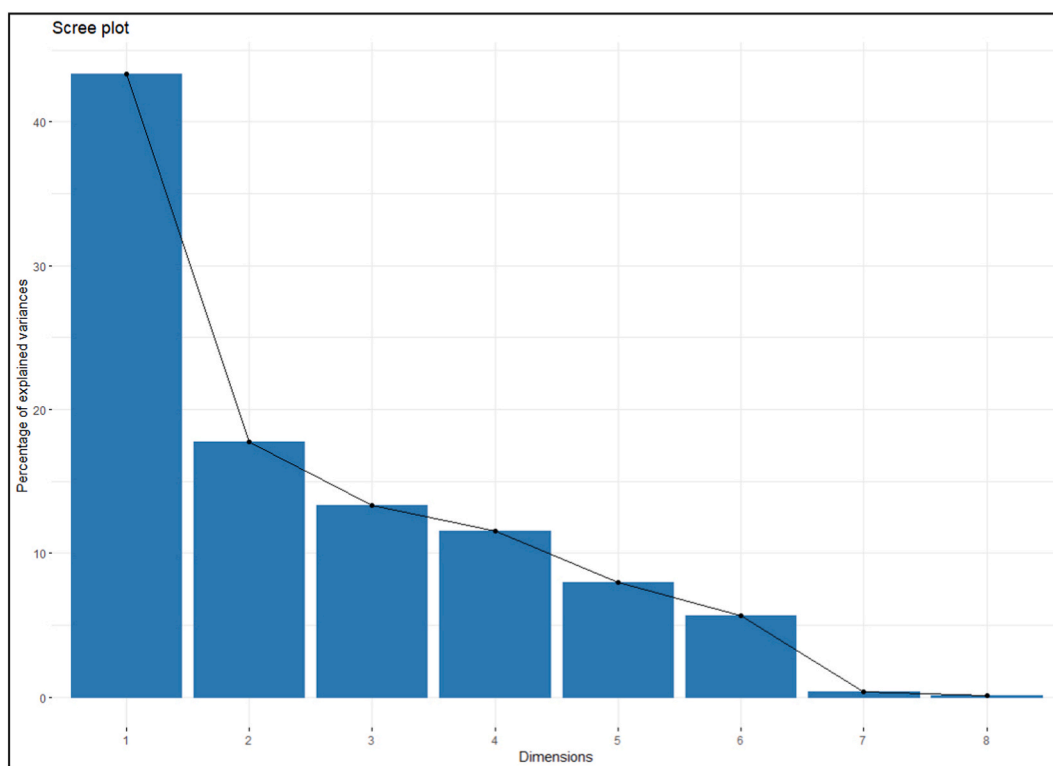


Fig. 2. Correlation heatmap illustrating the relationship between different germination and growth parameters in rice using the Pearson correlation coefficient method. *Significant at 5 % level of significance, **Significant at 1 % level of significance, ***Significant at 0.1 % level of significance; Boxes without stars denote non-significant associations; GP: Germination percentage, GS: Germination speed, GE: Germination energy, VI: Vigor index, RL: Root length, SL: Shoot length, FW: Fresh weight of seedling, and DW: Dry weight of seedling.

Table 5

Principal component analysis (PCA) of germination and growth traits in investigated rice varieties.

	PC1	PC2	PC3	PC4	PC5	PC6	PC7	PC8
Germination percentage	0.486	0.068	-0.182	0.040	-0.419	0.163	-6.90e-01	-0.213
Germination speed	0.403	-0.391	0.072	-0.213	0.476	-0.223	5.47e-02	-0.594
Germination energy	0.490	-0.273	-0.026	-0.137	0.223	-0.111	-8.31e-02	0.772
Vigor index	0.477	-0.000	0.101	-0.045	-0.514	0.172	6.79e-01	-0.061
Root length	0.065	-0.246	0.604	0.738	-0.048	-0.113	-9.48e-02	0.010
Shoot length	-0.155	-0.057	0.699	-0.621	-0.229	0.002	-2.08e-01	0.036
Fresh weight	0.210	0.648	0.106	0.001	-0.005	-0.723	1.64e-02	-0.001
Dry weight	0.243	0.532	0.289	0.041	0.477	0.586	-1.74e-05	-0.002
Standard deviation	1.861	1.192	1.031	0.959	0.799	0.672	0.166	0.080
Proportion of Variance	0.433	0.177	0.133	0.115	0.079	0.056	0.003	0.000
Cumulative Proportion	0.433	0.611	0.744	0.859	0.939	0.995	0.999	1.000

**Fig. 3.** Scree plot of eigen values after principal component analysis (PCA) representing the variance explained by each principal component (PC).

biochemical changes that prepare seeds for germination, ultimately leading to improved germination performance and related parameters [33]. One crucial effect of priming is the augmentation of α -amylase activity, an enzyme responsible for breaking down stored carbohydrates. This heightened enzymatic activity enhances the availability of energy-rich compounds required for germination. In our research study, the application of FeCl_2 (2 %) as well as DAP (2 %) as priming agents yielded remarkable results, with the highest germination percentage (87.33 %), germination speed (75.11 %), germination energy (66.00 %), and vigor index (1431) observed. This result inclined with the result of [34], who found iron the best priming media under salt stress conditions. The superiority of these specific priming agents can be mechanistically attributed to their influence on key biochemical pathways. Iron (Fe^{2+}), present in FeCl_2 , plays a multifaceted role in cellular processes. It facilitates various chemical reactions, including those involved in photosynthesis, respiration, and nitrogen fixation, thereby enhancing overall metabolic activity [35]. The presence of Fe^{2+} likely accelerates metabolic pathways essential for germination, contributing to the observed improvements in germination parameters. Moreover, the priming process is known to activate enzymes such as α -amylase and protease, which are pivotal for breaking down storage materials and facilitating the mobilization of nutrients during germination. The participation of Fe^{2+} in these enzymatic activities could further amplify their effects, leading to the robust germination outcomes observed. It's noteworthy that the choice of rice variety also plays a significant role in germination performance. For instance, variety BRRI Dhan-66 exhibited substantially higher germination percentage, speed, energy, and vigor index compared to the IR 80991-B variety. This discrepancy could stem from inherent genetic

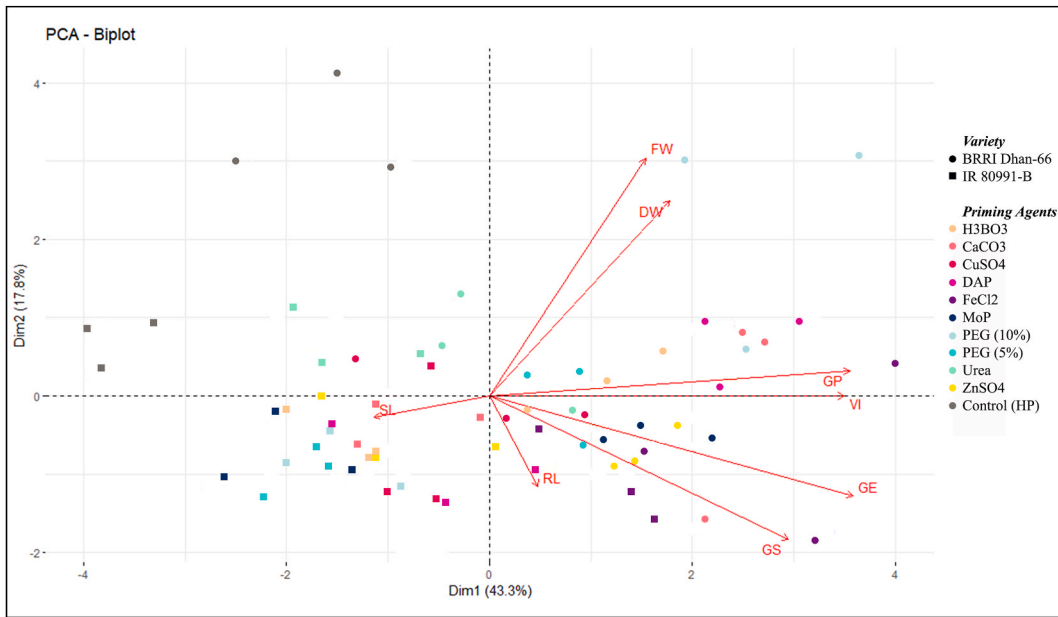


Fig. 4. Principal component analysis (PCA) of germination and growth traits within the studied rice varieties, illustrating the variance and trait patterns among them via a graphical method called biplot. GP: Germination percentage, GS: Germination speed, GE: Germination energy, VI: Vigor index, RL: Root length, SL: Shoot length, FW: Fresh weight of seedling, and DW: Dry weight of seedling. H₃BO₃: Boric Acid, CaCO₃: Calcium Carbonate, CuSO₄: Cupric Sulfate, DAP: Diammonium Phosphate, FeCl₂: Ferrous Chloride, MOP: Muriate of Potash, PEG: Polyethylene Glycol, ZnSO₄: Zinc Sulfate, Hydro-priming.

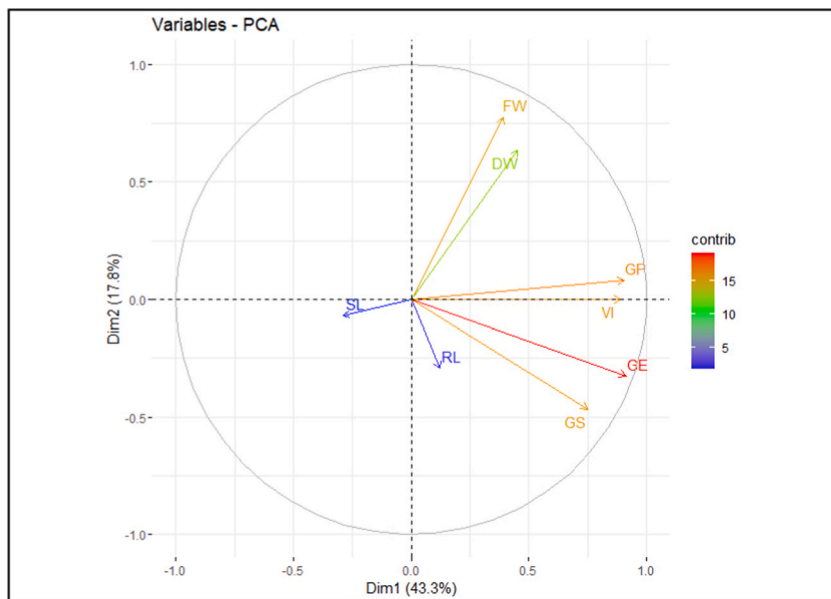


Fig. 5. Principal component analysis (PCA), loading plot illustrating the contribution of each measured variables. GP: Germination percentage, GS: Germination speed, GE: Germination energy, VI: Vigor index, RL: Root length, SL: Shoot length, FW: Fresh weight of seedling, and DW: Dry weight of seedling.

differences between the varieties, influencing their responsiveness to priming agents and stress conditions. In summary, the enhanced germination parameters observed following priming with FeCl₂ and DAP can be elucidated through the involvement of iron-mediated metabolic processes, enzyme activation, and nutrient mobilization.

4.2. Effect of priming agent on growth parameters

The establishment of robust plants is a critical determinant of final rice yield, and this process is intrinsically linked to the early development of seedlings. Key indicators of early seedling establishment include shoot and root length, as well as fresh and dry weight. Variability in root and shoot length among rice varieties has been documented, highlighting the genetic basis of these traits under different priming conditions [36]. Importantly, elevated stress levels have been shown to negatively impact parameters such as shoot and root dry and fresh weights, as well as shoot and root length [37]. Given these effects, the strategic choice of rice variety combined with appropriate priming media emerges as a potential avenue for enhancing rice growth parameters. In our study, the intricate relationship between developmental stages and priming media on growth parameters became evident. Specifically, the elongation of shoot length was most pronounced in BRRI Dhan-66 at 10 days after sowing (DAS), whereas at 20 and 30 DAS, IR80991-B exhibited the longest shoot length. Meanwhile, the longest root length was consistently demonstrated by the IR 80991-B variety across all 10, 20, and 30 DAS stages, aligning with the findings of previous research [8]. This nuanced variation in shoot and root length emphasizes the importance of growth stage and variety in determining the response to priming agents. Notably, our findings demonstrated that shoot and root length respond differentially to priming media across distinct DAS intervals, highlighting the complexity of these interactions. Delving into the mechanistic underpinnings of these phenomena, our results revealed that certain priming agents exerted notable effects on shoot and root length at specific time points. For instance, FeCl_2 (2 %) and MOP (2 %) promoted longer shoot length at 10 DAS, whereas ZnSO_4 (2 %) and DAP (2 %) facilitated enhanced shoot length at 20 DAS, and DAP (2 %) and PEG (5 %) supported increased shoot length at 30 DAS. These effects could be attributed to the influence of priming agents on cellular expansion and elongation processes, potentially through modulating hormone signaling pathways and nutrient uptake. Similar patterns were evident in root length growth, with different priming agents impacting root elongation at distinct stages. Notably, H_3BO_3 (0.1 %) and MOP (2 %) induced longer root length at 10 DAS, Urea (2 %) and CuSO_4 (2 %) at 20 DAS, and PEG (10 %) and CuSO_4 at 30 DAS. These outcomes likely stem from alterations in root development processes influenced by priming-induced physiological changes, including hormone balance, nutrient availability, and water uptake dynamics. Our findings are comparable to Ref. [22], which has found a significant difference in shoot and root length at 10, 15, and 20 DAS with different priming media. Furthermore, the observed variations in fresh and dry weight between BRRI Dhan-66 and IR 80991-B varieties underscore the significance of genetic diversity in growth performance. Intriguingly, our findings highlighted specific priming agents that led to heightened fresh weight, such as MOP (2 %) at 10 DAS and PEG (10 %) at 30 DAS. Similarly, dry weight exhibited notable increases with Urea (2 %) across all DAS intervals, potentially linked to optimized nitrogen utilization facilitated by priming. These results resonate with prior research linking dry matter accumulation with effective nutrient management strategies, particularly nitrogen optimization [38]. Importantly, the correlation between increased dry weight and higher urea concentrations echoes the findings of [39], confirming the influence of nitrogen-related processes on seedling growth. In summary, the intricate interplay between priming agents, growth stages, and rice varieties elucidates the multifaceted nature of seedling growth regulation. Mechanisms involving hormone signaling, nutrient uptake, and physiological responses underpin the observed changes in shoot and root length, as well as fresh and dry weight.

4.3. Interactive effect of priming agent on germination and growth parameters

Our research underscores the significant impact of distinct priming media on germination and growth parameters in two rice varieties, BRRI Dhan-6 and IRR80991-B. This variability in outcomes can be attributed to differential endospermic amylase activity induced by various priming agents, resulting in alterations in insoluble sugar content [8]. The orchestration of these intricate processes ultimately leads to changes in the pace and quality of germination and seedling growth. A pivotal mechanism underlying these effects is the initiation of pre-germinative metabolism triggered by priming agents. This metabolic reconfiguration serves to expedite germination, synchronize germination timing, improve germination rates, and facilitate robust seedling establishment in rice [10,40]. By delving into these mechanisms, our findings provide insight into the nuanced relationship between priming agents and rice performance. Analyzing the performance of BRRI Dhan-66 and IRR80991-B varieties, our results indicate that FeCl_2 (2 %) priming agent consistently yields favorable results in terms of germination percentage, germination energy percentage, and vigor index for BRRI Dhan-66, with the exception of germination speed, which peaks with CaCO_3 (2 %) priming. These nuanced responses underscore the importance of specific priming agents in influencing different aspects of germination across rice varieties, as illustrated in Figs. 6–9. This alignment with [8] underscores the close interplay between germination parameters, priming media, and the underlying genetic characteristics of the cultivars. Although our investigation did not reveal significant differences in root and shoot length under priming agents for both varieties, a distinct pattern emerged in terms of fresh and dry weight across all days after sowing (DAS) for both varieties. This notable significance in fresh and dry weight is suggestive of the pronounced influence of priming agents on nutrient uptake, allocation, and utilization, all of which are pivotal contributors to seedling growth. This phenomenon resonates with the broader understanding that priming agents can stimulate physiological processes that lead to improved nutrient mobilization and allocation within seeds, ultimately promoting seedling development. Our findings also resonate with [41], which observed that variations in the concentration of different priming media can exert a positive influence on germination and growth parameters. The convergence of our results with this prior research supports the notion that priming-induced changes in metabolic pathways and nutrient dynamics play a central role in shaping the germination and growth responses observed. To conclude, the complex interplay among priming agents, endospermic amylase activity, pre-germinative metabolism, and nutrient dynamics collaboratively contributes to the array of germination and growth results observed in various rice varieties.

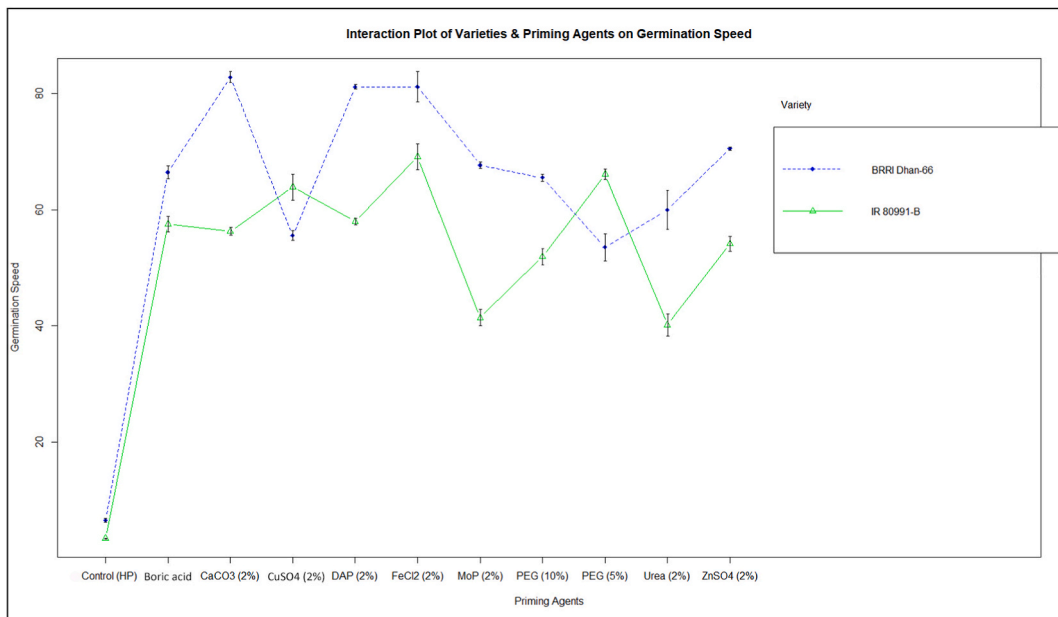


Fig. 6. Interaction plot of varieties and priming agents on germination speed of rice. CaCO₃: Calcium Carbonate, CuSO₄: Cupric Sulfate, DAP: Diammonium Phosphate, FeCl₂: Ferrous Chloride, MOP: Muriate of Potash, PEG: Polyethylene Glycol, ZnSO₄: Zinc Sulfate, Hydro-priming.

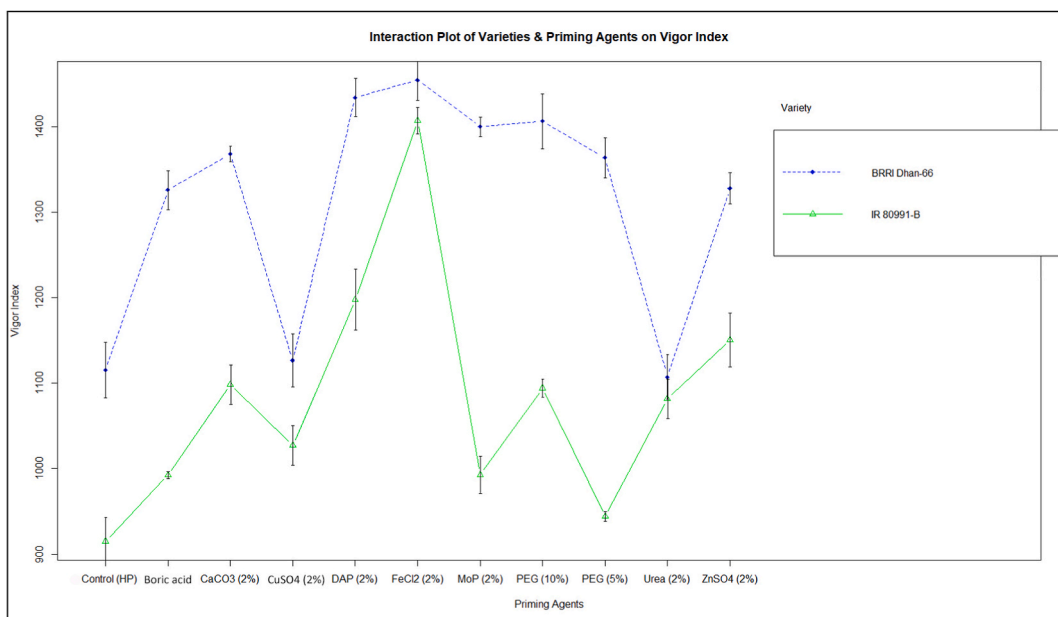


Fig. 7. Interaction plot of varieties and priming agents on vigor index of rice. CaCO₃: Calcium Carbonate, CuSO₄: Cupric Sulfate, DAP: Diammonium Phosphate, FeCl₂: Ferrous Chloride, MOP: Muriate of Potash, PEG: Polyethylene Glycol, ZnSO₄: Zinc Sulfate, Hydro-priming.

4.4. Correlation and PCA analysis

The relationships among germination and growth parameters exhibit a consistent pattern. Seed germination percentage and germination speed demonstrate a positive association with germination energy, vigor index, as well as the fresh and dry weight of seedlings. Conversely, there appears to be a negative influence from root and shoot growth (Fig. 2). This harmonious interaction underscores the fundamental importance of these parameters in facilitating robust seed germination and subsequent seedling development. The positive correlations signify that as germination energy, vigor index, and seedling weight increase, there is a corresponding rise in both the rate and percentage of seed germination. This also leads to an accelerated pace of germination. This

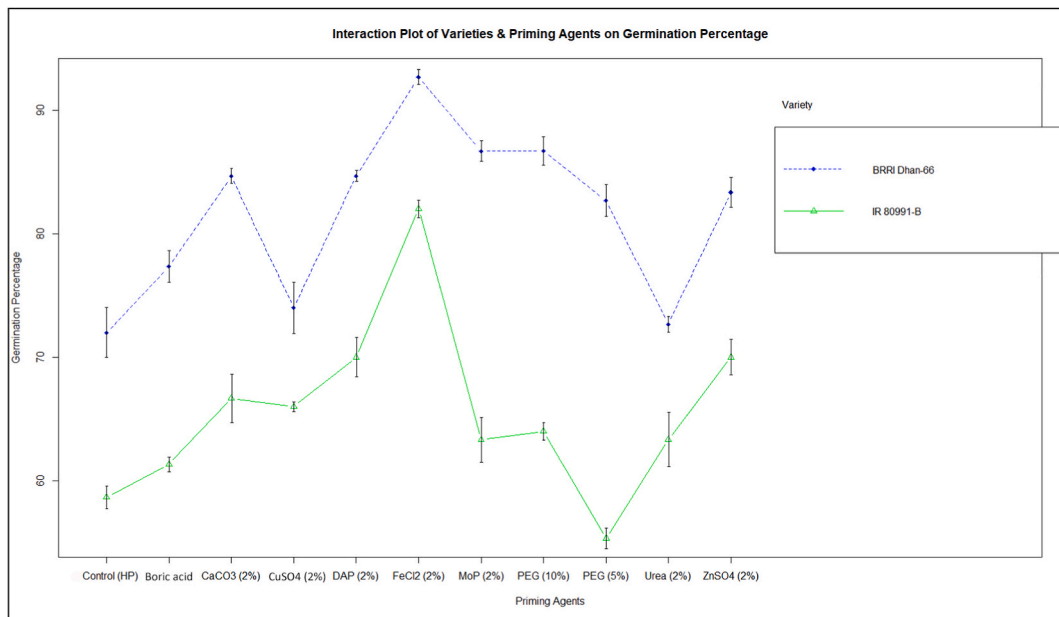


Fig. 8. Interaction plot of varieties and priming agents on germination percentage of rice. CaCO₃: Calcium Carbonate, CuSO₄: Cupric Sulfate, DAP: Diammonium Phosphate, FeCl₂: Ferrous Chloride, MOP: Muriate of Potash, PEG: Polyethylene Glycol, ZnSO₄: Zinc Sulfate, Hydro-priming.

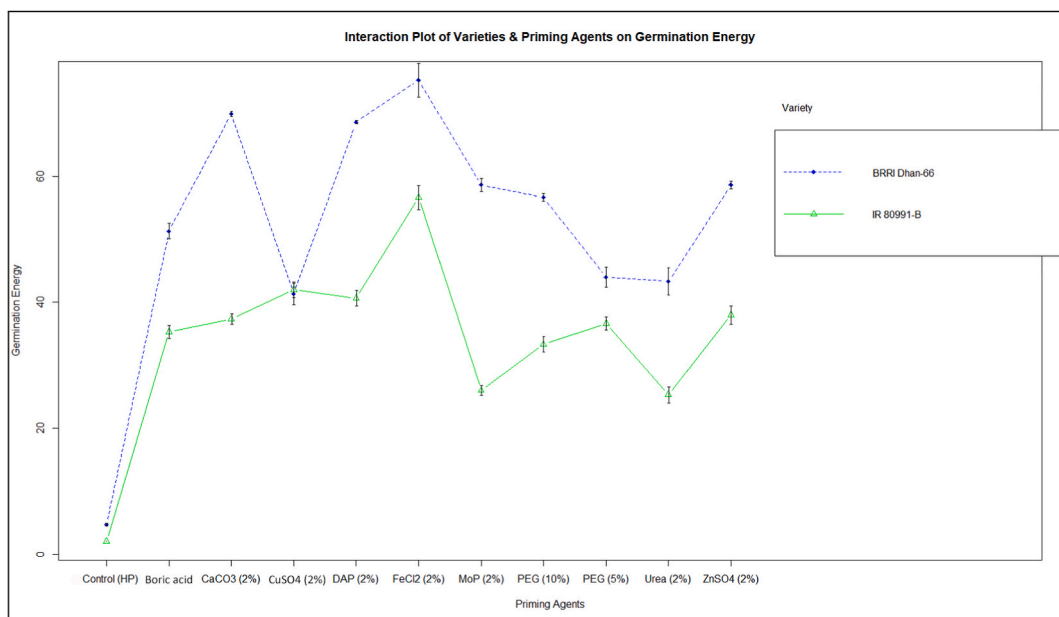


Fig. 9. Interaction plot of varieties and priming agents on germination energy of rice. CaCO₃: Calcium Carbonate, CuSO₄: Cupric Sulfate, DAP: Diammonium Phosphate, FeCl₂: Ferrous Chloride, MOP: Muriate of Potash, PEG: Polyethylene Glycol, ZnSO₄: Zinc Sulfate, Hydro-priming.

observation aligns with a mechanistic understanding where germination energy signifies the overall metabolic vitality of the seeds, the vigor index encapsulates the general well-being and vigor of seedlings, and seedling weight reflects physiological robustness. Collectively, these aspects harmonize to establish an environment that promotes efficient germination and subsequent growth. Our findings are consistent with those of a prior study [3], wherein a positive correlation was observed between germination rate and various traits of rice. Moreover, our findings align with those documented in an alternate study [1]. Nevertheless, they diverge from the results presented in a distinct study [42].

Principal Component Analysis (PCA) revealed a distinct pattern, emphasizing the pivotal role of specific parameters in driving dataset dynamics (Fig. 5). Among these, germination energy, germination percentage, germination speed, vigor index, and the fresh

weight of seedlings emerged as the most influential contributors. Notably, their elevated significance in the initial principal components implies that modifications or fluctuations in these attributes will exert a more pronounced impact on the overall variability of the dataset in contrast to other factors. Our findings exhibit similarity with the outcomes presented in studies [1,4], further reinforcing the coherence and reliability of our results.

5. Conclusion and Future directions

The study at hand convincingly establishes that seed priming, achieved through various treatments, has a significant potential to noticeably improve both the germination and growth aspects of rice seedlings. Each of the applied priming agents had a positive effect on the measured variables, confirming the effectiveness of seed priming as a strong strategy to enhance the performance of rice crops. A noteworthy finding is the remarkable responsiveness of the BRRI Dhan-66 rice cultivar to seed priming methods, suggesting its potential as a high-yielding variety that can further benefit from priming techniques. Additionally, the use of ferrous chloride (FeCl₂) treatment stands out for its particularly positive impact on germination parameters. Collectively, these findings strongly recommend the wider adoption of seed priming as a practical and environmentally mindful approach to boost rice crop productivity. The concrete advantages revealed by this study prompt further consideration for seamlessly incorporating this technique into modern agricultural practices. To optimize seed priming techniques, further research could explore varying concentrations and durations of treatments to identify the most effective regimes. Additionally, expanding the scope of investigation to mature plant traits, including yield potential and resistance to pests and diseases, could provide a more holistic understanding of the long-term impacts of priming treatments. Furthermore, potential research directions might delve into unraveling the intricate molecular and biochemical mechanisms underlying the observed effects of seed priming treatments on rice seedlings and also exploring the potential synergistic effects of these combined treatments. These inquiries offer the promise of a deeper understanding of the intricate processes guiding these enhancements, ultimately paving the way for more informed and targeted applications of seed priming within the realm of rice cultivation.

Data availability statement

Data will be made available on request.

Additional information

No additional information is available for this paper.

CRediT authorship contribution statement

Shubh Pravat Singh Yadav: Conceptualization, Data curation, Formal analysis, Investigation, Methodology, Software, Visualization, Writing – original draft, Writing – review & editing. **Riya Adhikari:** Data curation, Investigation, Methodology, Writing – original draft. **Prava Paudel:** Data curation, Investigation, Methodology, Writing – original draft. **Bibek Shah:** Data curation, Investigation, Methodology, Writing – original draft. **Shobha Pokhrel:** Data curation, Investigation, Methodology, Writing – original draft. **Suraj Puri:** Data curation, Investigation, Methodology. **Robin Adhikari:** Data curation, Investigation, Methodology. **Sangita Bhujel:** Conceptualization, Project administration, Resources, Supervision, Validation, Writing – review & editing.

Declaration of competing interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

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