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Effect of Seed Priming with PGPR on Germination, Mean Emergence Time and Vigour of Paddy under Osmotic Stress

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Abstract

The experiment was carried out during *kharif* (2022) at SHUATS, Naini, Prayagraj with 16 treatments comprised of PGPRs (PAR-6, PES-2, PR-7, PR-12 and PR-15) and PEG to assess the effect of seed priming with PGPRs on certain seed germination attributes of paddy under osmotic stress. All the treatments were replicated thrice and statistical design CRD was followed. Seeds of paddy variety MTU-1172 were treated with different concentrations of PGPRs followed by drying in the dark for 2–3 hours at room temperature. Seeds were sown in petri-dishes lined with blotting paper and different seed germination parameters were recorded. Findings of the experiment revealed that the seed priming with PES-2 showed maximum germination percentage (80%), germination index (7.33), seedling dry biomass (0.11) and vigour index-II (8.8), while as, seed priming with PAR-6 showed maximum value of MET (5.17 days). These findings may be attributed to the fact that pre-sowing of seeds triggered resistance under osmotic stress. Seedling vigour index-I were found maximum of 13.20, 14.77, 16.55 with treatment PR-7 plus PEG (-2bar), respectively at 7th, 14th and 21st days of measurement. Results also indicated that the PGPRs have similar effect like KNO₃, SiO₂ and SA, which might have to increase the seedling characters and vigour index-II in paddy rice. It may be concluded that the isolate PR-7 emerged as a superior PGPR to reduce the PEG-induced osmotic stress of -2 bars. Thus, farmers can use recommended eco-friendly, cost-effective and easily available PGPR for enhancing the productivity of paddy.

Keywords: Germination, osmotic stress, paddy, PGPR, seed priming

1. Introduction

Rice (*Oryza sativa* L.) is a major staple food for most people living in Asia and developing countries including India. Owing to its phylogenetic origin as a semi-aquatic plant, rice is highly dependent on a sufficient amount of water for growing, particularly during initial phase of plant growth (Duenas et al., 2024). Due to the increasing temperature and erratic precipitation, drought has been one of the most important limiting factors for crop productivity and, eventually, for food security worldwide (Fahad et al., 2017). Seed germination is highly sensitive to osmotic stress, as moisture stress disrupts seed hydration, delays metabolic activities, and impairs radical and plumule emergence, leading to poor germination rates and weak seedlings (Saha et al., 2022). Addressing these

challenges requires innovative solutions like seed priming with plant growth-promoting rhizobacteria (PGPR), which enhance metabolic activity and stress resilience, improving germination and early seedling vigor under adverse conditions. PGPR are a group of bacteria that can enhance plant growth and productivity (Mohanty et al., 2021). Priming of seeds is an easy, affordable and safe technique for enhancing emergence, seedling growth, yields and drought stress tolerance of crops (Rauf et al., 2016).

Inoculation of PGPR in the rhizosphere of crops has been seen as one of the most suitable strategies to promote plant growth under osmotic stress conditions (Chieb et al., 2023). PGPRs are known for their beneficial effects on plant growth, development, nutrient uptake and yield even under osmotic



stress conditions (Nwachukwu et al., 2023). Biopriming, a seed treatment combining priming with PGPRs, enhances germination speed, crop establishment, and yield. It allows bacteria to colonize seeds and acclimate to local conditions, improving nutrient uptake, stress tolerance, and harvest quality, making it a sustainable agricultural practice (Mahmood et al., 2016). These inoculants in single or co-inoculation form, enhance nodulation, nodule weight, nitrogen fixation, plant biomass, dry matter and grain yield (Backer et al., 2018). One effective strategy to deal with drought stresses is seed priming. To improve germination, seedling vigor, and stress tolerance, seeds are partially hydrated as part of the priming process prior to planting (Kaur et al., 2023). In addition to reducing the time needed for seedling emergence and accelerating germination, priming also helps in improving crop quality and ensures uniform germination (Anjum et al., 2023). During the priming process, seeds are exposed to outside elements including chemicals, heat, and moisture, which alters their physiological and biochemical condition (Rhaman et al., 2020). The seeds can now withstand environmental challenges like drought, salt, and temperature variations better after priming (Aswathi et al., 2022). Several agricultural crops have benefited from the use of seed priming methods including PGPRs, which has been shown to increase drought tolerance, increase the emergence of roots and shoots, and produce healthier plants. (Vurukonda et al., 2016).

Priming of paddy seeds with different PGPRs has shown to stimulate faster emergence rates, faster seedling establishment and vigorous seedling growth (Devika et al., 2021). Priming also minimizes abiotic stress effect at the germination phase and finally leads to higher seedling emergence as well as vigorous seedling establishment. These biological activities of priming are useful for farmers because they decrease the emergence time, cost of repeated seeding, fertilization and additional irrigation (Acharya et al., 2020). PEG molecules can generate equitable water stress instead of producing immediate physiological harm because they are neutral, non-ionized, and almost impermeable to the membranes of cells (Sobahan et al., 2022). Therefore, keeping the above facts in mind, the study was planned to ascertain the role of different strains of PGPRs in improving the seed germination potential of rice under PEG-induced osmotic stress condition.

2. Materials and Methods

The present experiment was carried out during 2022 at the Post Graduate Laboratory of the Department of Biological Sciences, Sam Higginbottom University of Agriculture, Technology and Sciences, Prayagraj (UP) which is located at 25° 24′ 42" N latitude, 81° 50′ 56" E longitude and 98m altitude above the mean sea level, during kharif (July-August, 2022) in CRD with 3 replications comprising 16 treatment combinations. Seeds of the rice variety Ksheera (MTU-1172) used for this study were obtained from the Department of

Genetics and Plant Breeding, SHUATS, Prayagraj (UP). Seeds were surface sterilized with 0.5% NaClO solution and washed several times with tape water followed by washing with sterilized distilled water. For inoculum preparation, bacterium cultures were grown in yeast extract broth medium at 30°C for 48 hours. Five bacterial strains PAR-6, PES-2, PR-7, PR-12 and PR-15 having high growth levels as compared to others were selected for this laboratory study. Rice seeds were separately soaked in solutions of PGPRs for 5-10 minutes shaking for coating and kept in the dark for 2-3 hours at room temperature 25±2°C. The Hoagland's nutrient broth solution (Gamborg and Wetter, 1975) was prepared and the seeds were placed in a plastic plate covered with blotting paper in-vitro (Glass Chamber), The plate was filled with nutrient medium to the point where seed is immersed in it. After completing 7 days stress treatment was initiated through PEG-4000 in Mild (-2 bar) 11.80 ml 100 ml⁻¹ D-H₂O, Moderate (-5 bar) 19.70 ml 100 ml⁻¹ D-H₃O and Severe (-8 bar) 25.40 ml 100 ml⁻¹ D-H₃O in each plate in different concentration. Blotting papers were kept moist and the contamination of growth media was monitored critically. Observations on germination percentage, mean emergence time and seedling characters were recorded. The data so obtained were used for statistical analysis using SPSS.

3. Results and Discussion

3.1. Seed priming enhanced germination attributes of rice under osmotic stress

Seed priming enhanced germination and vigour attributes of rice under osmotic stress. Seed priming with PGPR PAR-6, PES-2, PR-7 and PR-12 had significant (p≤0.05) effects on germination percentage (GP), germination index (GI), seedling dry biomass (SDB), vigour index-II and mean emergence time (MET) of paddy variety MTU-1172. This study found that the application of PGPR as priming substantially improved the germination percentage, and germination index of MTU-1172 rice under drought/osmotic stress. Improved germination attributes of rice might be related to increased cell division and elongations, water imbibitions by seeds, repair and synthesis of DNA and RNA, increased activities of reserve mobilizing enzymes such as acid phosphatase, dehydrogenase, α -amylase and β -amylase in primed seeds. These findings are in conformity with the findings of Xu et al., 2020.

However, priming of PGPR has no significant effect on GP i.e., PAR-6 and PR-15 of MTU-1172 variety of rice. Whereas, PGPR, PES-2, PR-7 and PR-12 increased germination of the MTU-1172 variety of rice by 100% as compared with control (40%). PES-2, PR-7, PAR-6 and PR-12 increased GI of MTU-1172 of rice by 109%, 76%, 9% and 7% in comparison to control (3.50) but priming of PGPR, PR-15 has no significant effect on GI in rice (Table 1). This study investigated the influence of Plant Growth Promoting Rhizobacteria (PGPR) on seed priming to improve germination and seedling growth of rice seedlings under drought/osmotic stress. It is well known

Table 1: Effect of seed priming with PGPR on germination percentage, germination index, mean emergence time, seedling dry biomass and vigour index-II of paddy

Treat- ments	Germi- nation (%)	Germi- nation Index	Mean emergence Time	Seed- ling dry biomass	Vigour index II
			(days)	(mg)	
Control	60.00	3.50	5.00	0.05	3.00
PAR-6	40.00	3.83	5.17	0.08	3.20
PES-2	80.00	7.33	4.96	0.11	8.80
PR-7	80.00	6.17	5.00	0.09	7.20
PR-12	80.00	5.83	5.12	0.09	7.20
PR-15	40.00	3.50	4.90	0.07	2.80
SEm±	0.58	0.15	0.05	0.01	0.15
CD (<i>p</i> =0.05)	3.08	0.80	0.27	0.03	0.48

that rice production in arid and semi-arid ecosystems of the world is being ravaged by drought under a changing climate. Drought affects germination, seedling emergence and yield of rice. Similar results in paddy have been reported by Yadav et al., 2019.

3.2. Seed priming enhanced seedling biomass and vigor of rice under osmotic stress

Data on MET revealed that it increased by 3.4% and 2.4% in PAR-6 and PR-12 respectively as compared to control (5.00 days) in rice variety MTU-1172 and there was no significant effect of PGPR's PES-2, PR-7 and PR-15 on MET. Similarly, another parameter SDB depicted that PGPR, PES-2 (120%), PR-7 (80%), PR-12 (80%), PAR-6 (60%) and PR-15 (40%) showed increased performance for SDB in rice compared to the control (0.05mg). The highest vigour index-II was recorded in PES-2 (193%) followed by PR-7 & PR-12 (140%) and PAR-6 (7%) primed MTU-1172 rice seedlings (Table 1). These results are due to the fact that pre-sowing priming, invigorating, or triggering resistance against stresses such as drought, salinity, cold or disease also observed that seedling biomass and vigor were enhanced due to PGPR. These findings are in conformity with the findings of (Xu et al., 2020) in rice.

3.3. Seed priming improved seedling growth, vigour and root performance of rice under osmotic stress

Seed priming and its interaction with osmotic stress PR-7+PEG-2 bar and PES-2+PEG-2 bar had significant effects on Seedling length (SL) and vigour index-I in MTU-1172 variety of rice. At mild stress (-2 bars), priming with PR-7, PES-2, PR-12, PAR-6 and PR-15 increased seedling length of MTU-1172 rice

Table 2: Effect of Seed priming with PGPR on seedling characters, vigour index-I, nodal roots and seminal roots under osmotic stress of paddy

Code	Treatments	Seedling length (cm)			Vigor index			
		Pre PEG treatment	PEG treatment		Pre PEG treatment Post PEG treatme		reatment	
		7 th DAS	14 th DAS	21st DAS	7 th DAS	14 th DAS	21st DAS	
$T_{_{0}}$	Control	5.43	7.86	9.90	325.80	471.60	594.00	
$T_{_{1}}$	PAR-6+PEG-2 bar	8.90	10.77	12.90	356.00	430.80	516.00	
$T_{_{2}}$	PAR-6+PEG-5 bar	7.63	9.96	12.20	305.20	398.40	488.00	
T_3	PAR-6+PEG-8 bar	7.73	10.07	12.20	309.20	402.80	488.00	
$T_{_{4}}$	PES-2+PEG-2 bar	12.17	14.10	16.26	973.60	1128.00	1300.80	
$T_{_{5}}$	PES-2+PEG-5 bar	8.57	10.73	12.87	685.60	858.40	1029.60	
$T_{_{6}}$	PES-2+PEG-8 bar	10.73	12.47	14.58	858.40	997.60	1166.40	
T ₇	PR-7+PEG-2 bar	13.20	14.77	16.55	1056.00	1181.60	1324.00	
T ₈	PR-7+PEG-5 bar	6.30	8.73	10.88	504.00	698.40	870.40	
T_{9}	PR-7+PEG-8 bar	6.60	8.37	10.52	528.00	669.60	841.60	
T ₁₀	PR-12+PEG-2 bar	9.87	11.47	13.63	789.60	917.60	1090.40	
T ₁₁	PR-12+PEG-5 bar	10.13	12.83	14.96	810.40	1026.40	1196.80	
T ₁₂	PR-12+PEG-8 bar	5.37	7.30	9.01	429.60	584.00	720.80	
T ₁₃	PR-15+PEG-2 bar	7.67	10.16	12.32	306.80	406.40	492.80	
T ₁₄	PR-15+PEG-5 bar	1.57	3.67	6.12	62.80	146.80	244.80	
T ₁₅	PR-15+PEG-8 bar	4.43	6.20	8.31	177.20	248.00	332.40	
SEm±		0.33	0.26	0.29	0.04	0.05	0.06	
CD (<i>p</i> =0.05)		0.94	0.76	0.84	0.12	0.15	0.17	

Code	Treatments	Number of nodal root			Number of seminal root		
		Pre PEG treatment	Post PEG treatment		Pre PEG treatment	Post PEG treatment	
		7 th DAS	14 th DAS	21st DAS	7 th DAS	14 th DAS	21st DAS
T _o	Control	1.00	2.00	2.67	2.67	5.00	14.00
T ₁	PAR-6+PEG-2 bar	1.00	2.00	2.00	6.00	10.00	12.00
T ₂	PAR-6+PEG-5 bar	1.00	1.67	2.00	3.00	3.33	4.00
T ₃	PAR-6+PEG-8 bar	0.00	1.00	1.00	0.00	2.33	2.67
T ₄	PES-2+PEG-2 bar	2.33	0.33	0.33	3.33	0.00	0.00
T ₅	PES-2+PEG-5 bar	0.67	5.00	4.67	4.00	14.00	15.33
T ₆	PES-2+PEG-8 bar	1.33	2.00	2.33	3.67	9.00	10.00
T ₇	PR-7+PEG-2 bar	1.00	3.33	3.33	4.67	9.67	10.00
T ₈	PR-7+PEG-5 bar	1.00	2.33	2.33	1.67	4.67	5.33
T ₉	PR-7+PEG-8 bar	2.00	3.00	3.33	2.33	7.67	8.33
T ₁₀	PR-12+PEG-2 bar	3.00	3.33	3.33	5.00	12.67	13.33
T ₁₁	PR-12+PEG-5 bar	1.00	2.00	2.33	1.67	5.67	6.67
T ₁₂	PR-12+PEG-8 bar	1.33	3.33	4.33	1.00	10.33	11.00
T ₁₃	PR-15+PEG-2 bar	0.67	1.67	2.00	2.00	6.67	8.00
T ₁₄	PR-15+PEG-5 bar	0.67	1.00	1.00	0.67	4.00	4.33
T ₁₅	PR-15+PEG-8 bar	1.33	1.00	1.00	0.00	4.33	4.67
SEm±		0.55	0.73	0.74	3.12	2.63	2.48
CD (p=	=0.05)	1.57	2.10	2.14	8.99	7.60	7.17

by 67%, 64%, 38%, 30% and 24% as compared with control (9.90 cm). At moderate stress (-5 bars), priming with PR-12, PES-2, PAR-6 and PR-7 increased seedling length of MTU-1172 rice by 51%, 30%, 23% and 10% as compared with control. Whereas, at severe stress (-8 bars), priming with PES-2, PAR-6 and PR-7 increased seedling length of MTU-1172 rice by 47%, 23% and 6% as compared with control and there was no significant effect of PR-12 and PR-15 on seedling length at severe stress (Table 2). Ali et al., 2021 revealed that priming of seeds stimulates pre-germination processes in rice that improve faster germination and seedling emergence. Seedling characteristics such as increased seedling length, vigor and nodal and seminal roots were enhanced due to water imbibitions, cell division and hydrolytic enzymes in rice were stimulated by priming, which triggered faster germination as well as the establishment of seedlings under stressful conditions.

In vigour, at mild stress (-2 bars), priming with PR-7, PES-2 and PR-12 increased vigour of MTU-1172 rice by 122%, 119% and 84% as compared with control. While at moderate/medium stress, priming with PR-12, PES-2 and PR-7 increased vigour of MTU-1172 rice by 101%, 73% and 47% as compared with control. However, at severe stress, PES-2, PR-7 and PR-12 increased vigour by 96%, 42% and 21% as compared to control and there was no significant difference between mild, moderate and severe stress on vigour of rice variety MTU-1172 with PAR-6 and PR-15 (Table 2).

Data on nodal roots opined that at mild, moderate/medium and severe stress effect of priming with PES-2, PR-7 and PR-12 increased nodal roots of MTU-1172 rice by 75%, 25% and 62% as compared to control. Whereas, there was no significant effect of mild, moderate and severe stress of priming on PAR-6 and PR-15 on nodal roots development in rice. In contrast, none of the PGPRs had a positive direct effect on seminal roots development in rice variety MTU 1172 (Table 2). Previous results by Yuan et al., 2010 found that rice priming with water and polyethylene glycol under drought had considerably increased emergence percent, emergence index and decreased emergence time. Zhang et al., 2015 reported that seeds primed with polyethylene glycol grown under drought showed improved emergence percentage, emergence index and vigour index. Khaliq et al., 2015 found that rice priming with selenium substantially increased emergence, emergence index and decreased mean emergence time. All these findings supported the fact that PGPR's PAR-6, PES-2, PAR-7 and PR -12 were responsible for better performance of rice under osmotic stress.

4. Conclusion

Seed priming with different PGPR influenced the seed germination characteristics in rice under PEG-induced osmotic stress (-2.0 bar) conditions. Priming with PAR-6, PES-2, PR-7 and PR-12 was found to be more effective in improving germination, and seedling growth. Therefore, being an easy and affordable technique, seed biopriming with PGPRs may be recommended to be adopted by the farmers for sowing rice in dry regions of India for improving germination, seedling establishment and growth under stress conditions.

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