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Optimisation of Seed Priming Techniques in Paddy

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Authors' contributions

This work was carried out in collaboration among all authors. All authors read and approved the final manuscript.

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ABSTRACT

Aim: To determine the ideal concentration of each priming agent among several selected concentrations which could produce robust paddy seedlings under laboratory conditions.

Place of Study: The present investigation was carried out at College farm, Agricultural College, Bapatla.

Methodology: Certain seedling vigour traits such as germination percentage, shoot length, root length, seedling length, seedling dry weight, seedling vigour index-I & II were recorded and analysed using crompletely randomized block design (CRD). Best concentration is the one with higher vigour indices.

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Result: The results revealed that, 1% Nacl, 40 ppm ascorbic acid, 150 ppm gibberellic acid, 2 % neem leaf extracts, 0.5 & 0.1 % P & Zn were optimum concentrations under each priming technique. When compared among all the techniques, priming with gibberellic acid @ 150 ppm performed better in terms of all the vigour traits.

Keywords: Hormonal priming; paddy; seed invigoration; seed priming techniques.

1. INTRODUCTION

Climate change, decline in water availability, non-availability of farm labour, escalated cost of cultivation together are forcing farmers to take up direct seeded rice cultivation (DSR) over normal transplanting. However, direct seeded rice is not free from problems. When the seeds are sown, they are subjected to several biotic (weeds) and abiotic stresses (moisture stress) especially under dry direct seeded condition. Germination gets effected and seedlings performance would be poor resulting in uneven crop stand and yield loss. Identification of a technique which can solve these problems is necessary. Dingkuhn et al. [1] proposed that rice varieties suitable for direct seeded systems must posess ESV, enhanced foliar growth to combat weeds during the vegetative stage, moderate tillering (10-12 tillers) and enhanced assimilate export from leaves to stems during the late vegetative and reproductive phases. Because it is directly related to crop growth and yield in direct seeded condition, seedling vigour is a prominent breeding target in rice [2]. Seed priming is one such method through which vigour can be induced in the seedlings and help them to perform better under normal as well as stressful situations.

Seed priming is a pre sowing approach for influencing the seedling development by stimulating pre germination metabolic activities prior to the emergence of radicle and improvement in the germination rate and performance of plant. Seed priming is a controlled hydration process in which seeds are dipped in water or any solution for a specific time period to allow the seed to complete its metabolic activities before sowing and then re dried to original weight [3].

Matsushima and Sakagami [4] from Kagoshima, Japan realized that hydro priming (tap water for 12 hours) the rice seedlings (Koshihikari) resulted in significantly higher shoot length, shoot elongation rate, shoot dry weight and root dry weight compared to control at different soil moisture regimes. An experiment conducted by Farooq et al. [5] at Faisalabad, Pakistan, found higher alpha amylase activity and total soluble sugars with hydropriming over no priming for two varieties (Seher-2006 and Shafaq-006) of bread wheat. Subedi et al. [6] revealed that hydro priming and Nacl priming (1.8 % and 3.6 %) for 12 hours improved germination energy, germination index, germination speed, radicle and plumule length in a rice cultivar, Sookha dhan-2 at Lamjung, Nepal over no priming and both the treatments were found to be at par with each other with no discernible effect on root shoot ratio. The experiment conducted by Mamun et al. [7] at the Seed Laboratory of Bangladesh Agricultural Development Corporation (BADC), Gazipur during June to July 2014 on Nerica, BRRI dhan 51, BRRI dhan 41, and BRRI dhan 49 rice varieties revealed that highest germination percentage, speed of germination, shoot length, root length, seedling dry weight, and vigour index were possible with vitamin priming i.e., 10 ppm ascorbic acid. Dhillon et al. [8] observed that hormone priming (50 ppm GA_3) for 12 and 24 hrs increased germination percentage, root length, shoot length, seedling dry weight, and SVI-I and II in rice *cv*. PR 126 rice at PAU, Ludhiana. According to Kale et al. [9], the highest germination %, seedling length (cm), seedling fresh weight (g), seedling dry weight (g) and vigour index were observed in sorghum primed with NLE (3 %) for 8 hrs over no priming with two varieties M35-1 (Maldandi) and SPV 1411 (Parbhani Moti) in Uttar Pradesh. Abbas et al. [10] found that seed priming with 0.1 and 0.5% Zn solution for 24 hours reduced the time for 50% germination, mean germination time and improved germination percentage, germination index, plumule length, radicle length and vigour index over hydro and no priming in an experiment conducted in Pakistan on fine grain aromatic rice cultivars, Super basmati and Shaheen basmati.

Despite of numerous reports supporting the role of seed priming in improving the vigour and yield of various crops in the lab and in the field, there are contradictory reports that differ strikingly. Anwar et al. [11] from Bangladesh found that priming with NaCl (10,000, 20,000 and 30,000 ppm) for 24 hours had no advantage over no priming for germination rate, seedling vigour & growth indices in 5 different winter rice varieties. Viera et al. [12] found that soaking rice seed in GA_3 increased mesocotyl elongation but had little effect on coleoptile elongation. This disparity arises as a result of several factors that contribute significantly to the success of seed priming which includes, priming agent, concentration of the agent, soaking duration, crop species and variety. Keeping these facts in view, the present investigation was carried out to standardise the optimum concentration of each priming agent for the paddy seeds of BPT 5204 variety.

2. MATERIALS AND METHODS

The experiment was carried out at Post Graduate (P.G) Laboratory, Department of Agronomy, Agricultural College, Bapatla during 2021. Breeder seed of BPT 5204 variety seeds were procured from Agricultural Research Station, Bapatla. Rice seeds were subjected to six different concentrations of five priming agents for standardization including hydropriming as control (0). Completely Randomized Design (CRD) was adopted for the standardization with four replications. The optimum concentration of each priming agent thus obtained was again analyzed for comparison in a Completely Randomized design with 7 treatments (no priming, hydropriming & 5 priming agents) each replicated 4 times. Details of different priming agents and respective concentrations selected for priming are as follows.

For halo-priming with Nacl, one gram of Nacl was dissolved in distilled water and made up to 100 ml, to get required concentration of 1% Nacl solution. Similarly 1.5, 2, 2.5, 3 % solutions were prepared with 1.5, 2, 2.5, 3 g of Nacl, respectively. The “0” in the priming treatment was considered as control (soaking in normal water i.e., hydropriming). In case of vitamin priming with ascorbic acid, 0.01, 0.02, 0.03, 0.04 and 0.05 g of ascorbic acid was weighed separately and the volume was made up to 1L with distilled water to get required concentration of 10, 20, 30, 40 and 50 ppm of ascorbic acid solution, respectively. For hormonal priming with gibberellic acid, 0.05 g of GA_3 was weighed separately and dissolved in few drops of ethyl alcohol and the volume was made up to 1 L to obtain required concentration of 50 ppm of GA_3 solution. 0.1, 0.15, 0.2, 0.3 g of GA_3 were individually weighed and used to prepare 100, 150, 200, 300 ppm of GA_3 solution, respectively in a similar way. The fresh leaves of neem tree were collected and dried under shade to prepare for botanical extract priming with neem leaf extracts. The shade dried leaves were powdered using mortar and pestle or electric grinder. One gram of neem leaf powder was dissolved in distilled water and the extract was filtered by using muslin cloth to remove unwanted material and leaf debris and the volume was made up to 100 ml to get 1% of extract solution. Similarly, 2, 4, 6, 8 and 10% of neem leaf extracts were prepared with 2, 4, 6, 8 and 10g of leaf powder, respectively. Whereas in case of nutri-priming with P (KH_2PO_4) & Zn (Zn-EDTA) solution, 0.44, 2.19, and 4.4 g of KH_2PO_4 were weighed separately and the volume was made to 100 ml to get 0.1, 0.5 and 1% of P solutions respectively. 0.83 and 4.15 g of Zn-EDTA was dissolved in 100 ml of distilled water to get 0.1 and 0.5 % of Zn solution respectively.

The seeds were soaked for 12 hrs [13, 14, 15] in different concentrations of priming agents on seed weight to solution volume (W/V) basis of 1:5 (g ml^{-1}) [3]. After soaking was completed, the seeds were thoroughly washed and dried back to original moisture content. The dried seeds were kept separately in cloth bags under ambient conditions for further evaluation. Four hundred seeds in four replicates of 100 seeds each were arranged in between moist paper towel and were kept in germinator at 25℃ ± 1℃ temperature. On the day of final count (14th day), the vigour of the seedlings was evaluated with the following attributes as per Between Papers method [16].

2.1 Germination Percentage (%)

On the day of final count, the number of normal seedlings were counted and percentage of germination was computed.

2.2 Shoot and Root Length (cm)

After final germination count, 10 normal seedlings were kept separately for length measurement. The shoot and root portion of the seedlings were separated with the help of a sharp razor blade and length measurement was taken with the help of centimetre scale.

2.3 Seedling Dry Weight (mg)

The shoot and root portion of the normal seedling after length measurement were dried in an oven at 85℃ temperature for 48 hours and then dry weight was measured in electronic balance and expressed in gram.

2.4 Root: shoot ratio

The root length to shoot length ratio of the 10 selected seedlings was computed and their mean was expressed as root / shoot ratio.

2.5 Seed Vigour Index (SVI)

Seedling vigour index (SVI) can explain seedling performance comprehensively. Hence, SVI-I and II were chosen as important indicators for standardizing the concentration of priming agents. Vigour index of seeds for all the treatments were calculated as per the formula developed by Abdul - Baki and Anderson [17].

Seed vigour index (I)= Mean seedling length (cm) × Germination (%)

Seed vigour index (II)=Mean seedling dry weight (mg) × Germination (%)

3. RESULTS AND DISCUSSION

The data on the effect of different concentrations of Nacl on seedling vigour traits of paddy was presented in Table 2. Significant difference was observed among different priming concentrations w.r.t all the vigour traits except root:shoot ratio. Among different halopriming concentrations, vigour traits such as root length, shoot length, seedling length, seedling dry weight, germination percentage, SVI-1 and SVI-II were found higher in the seedlings primed with 1.5% Nacl which was at par with 1 % Nacl. A positive influence of halo priming on seedling vigour was observed up to a concentration of 1.5%. With an increase in the concentration (2 & 3%), negative influence was recorded (decline in SVI-I and SVI-II. In view of the resource optimization, the concentration of 1 % Nacl was considered as optimum concentration instead of 1.5 % Nacl as both of them performed equally in terms of vigour indices.

On observing the data presented in Table 3, there was significant difference among different priming concentrations w.r.t all the vigour traits except root:shoot ratio. Among various concentrations of ascorbic acid, seedlings primed with 50 ppm were found to have higher vigour traits which was at par with 40 ppm ascorbic acid. Since, both the concentrations i.e., 40 and 50 ppm of ascorbic acid invigorated the seedlings similarly, lower concentration (40 ppm) was selected as best one over higher concentration (50 ppm).

The effect of different concentrations of hormonal priming on seedling vigour traits of paddy was presented in Table 4. Similar to halo, vitamin and harmonal priming, significant difference was observed among various concentrations w.r.t all the vigour traits except root:shoot ratio. Vigour traits (root length, shoot length, seedling length, seedling dry weight, germination percentage, seedling vigour index-I and seedling vigour index-II) were found superior in the seedlings primed with gibberellic acid @ 300 ppm which was at par with 200 and 150 ppm. Despite the fact that all three doses (300, 200 and 150 ppm) had a similar effect on seedling vigour, the lower dosage of 150 ppm was the optimum concentration considering resource optimization.

Data on effect of different concentrations of neem leaf extracts presented in Table 5 revealed that, for all the priming concentrations, vigour traits such as root length, shoot length, seedling length, root:shoot ratio and germination percentage were found to be non significant. Whereas seedling dry weight and seedling vigour indices were significant. All the botanical priming concentrations (2, 4, 6, 8 and 10 %) were found at par with each other and superior over control. However a concentration of 2% was selected as optimum one.

Table 1. Seed priming agents and their respective concentrations selected for standardization under laboratory conditions

S.NO	Seed priming agent	Concentrations					
1	Halo-priming with Nacl (%)	0	1	1.5	2	2.5	3
2	Vitamin priming with Ascorbic acid (ppm)	0	10	20	30	40	50

3	Hormonal priming with Gibberellic acid (ppm)	0	50	100	150	200	300
4	Botanical extract priming with Neem leaf extracts (%)	0	2	4	6	8	10
5	Nutri-priming with P (KH_2PO_4) & Zn (Zn-EDTA) solution (%)	0	0.1 & 0.1	0.5 & 0.1	1.0 & 0.1	0.5 & 0.5	1 & 0.5

Table 2. Effect of halo priming (Nacl) at different concentrations on seedling vigour traits of paddy under laboratory conditions

Nacl concentration (%)	Root length (cm)	Shoot length (cm)	Seedling length (cm)	Seedling dry weight (mg)	Root: Shoot ratio	Germination percentage (%)	Seedling Vigour Index-I	Seedling Vigour Index-II
0	10.79	9.83	20.63	8.23	1.11 (1.26)*	91.75 (73.33)**	1892.53	755.31
0.5	10.85	9.85	20.70	8.26	1.24 (1.27)	91.50 (73.08)	1894.42	756.16
1	10.98	10.23	21.21	8.36	1.10 (1.26)	91.25 (72.81)	1935.65	763.00
1.5	11.37	10.51	21.89	8.38	1.10 (1.26)	92.00 (73.59)	2013.87	770.45
2	10.97	10.24	21.21	8.22	1.09 (1.26)	90.50 (72.05)	1929.62	745.24
3	10.94	10.10	21.04	8.22	1.10 (1.26)	90.25 (71.84)	1899.32	744.08
SEm±	0.18	0.20	0.24	0.04	0.01	0.54	28.64	3.92
CD (P=0.05)	0.54	0.59	0.72	0.10	NS	1.60	85.09	11.66
CV (%)	3.9	4.6	2.7	1.0	2.2	1.7	3.5	1.2

Table 3. Effect of vitamin priming (Ascorbic acid) at different concentrations on seedling vigour traits of paddy under laboratory conditions

Ascorbic acid Concentration (ppm)	Root length (cm)	Shoot length (cm)	Seedling length (cm)	Seedling dry weight (mg)	Root: Shoot ratio	Germination percentage (%)	Seedling Vigour Index-I	Seedling Vigour Index-II
0	10.79	9.83	20.63	8.23	1.09 (1.26)*	91.75 (73.33)**	1892.53	755.31
10	11.39	9.54	20.93	8.50	1.21 (1.31)	90.5 (72.06)	1894.11	751.51
20	11.41	9.58	21.00	8.54	1.21 (1.31)	91.75 (73.36)	1926.46	775.40
30	11.52	10.01	21.52	8.63	1.16 (1.29)	91.75 (73.33)	1974.40	780.66
40	12.20	10.62	22.82	9.15	1.16 (1.29)	93.75 (75.53)	2139.47	856.78
50	12.45	10.91	23.37	9.21	1.15 (1.29)	93.75 (75.55)	2190.92	859.08
SEm±	0.26	0.19	0.29	0.06	0.02	0.54	21.05	5.87
CD (P=0.05)	0.76	0.55	0.87	0.19	NS	1.61	62.53	17.44
CV (%)	5.3	4.5	3.3	1.4	2.9	1.7	2.5	1.4

Table 4. Effect of hormonal priming (Gibberellic acid) at different concentrations on seedling vigour traits of paddy under laboratory conditions

Gibberellic acid concentration (ppm)	Root length (cm)	Shoot length (cm)	Seedling length (cm)	Seedling dry weight (mg)	Root: Shoot ratio	Germination percentage (%)	Seedling Vigour Index-I	Seedling Vigour Index-II
0	10.79	9.83	20.63	8.23	1.09 (1.26)*	91.75 (73.33)**	1892.53	755.31
50	11.00	10.0	21.06	9.17	1.11 (1.27)	92.75 (74.42)	1953.56	851.91
100	11.13	10.20	21.33	9.17	1.10 (1.27)	93.75 (75.60)	2000.62	867.50
150	12.60	11.41	24.01	9.57	1.12 (1.27)	94.25 (76.21)	2262.12	901.60
200	12.75	11.43	24.18	9.74	1.13 (1.28)	94.50 (76.48)	2284.79	920.22
300	12.78	11.55	24.33	9.74	1.12 (1.27)	94.75 (76.80)	2304.64	922.68
SEm±	0.31	0.23	0.30	0.06	0.02	0.73	30.25	7.46
CD (P=0.05)	0.93	0.69	0.90	0.17	NS	2.18	89.86	22.15
CV (%)	6.4	5.2	3.2	1.4	3.1	2.3	3.4	1.7

Table 5. Effect of botanical extract priming (Neem) at different concentrations on seedling vigour traits of paddy under laboratory Conditions

Neem leaf extracts concentration (%)	Root length (cm)	Shoot length (cm)	Seedling length (cm)	Seedling dry weight (mg)	Root: Shoot ratio	Germination percentage (%)	Seedling Vigour Index-I	Seedling Vigour Index-II
0	10.79	9.83	20.63	8.23	1.09 (1.26)*	91.75 (73.33)**	1892.53	755.31
2	10.88	10.21	21.2	8.43	1.07 (1.25)	91.25 (72.81)	1935.25	768.84
4	10.94	10.21	21.15	8.52	1.08 (1.26)	91.25 (72.83)	1930.37	777.77
6	10.95	10.26	21.21	8.55	1.08 (1.26)	91.50 (73.11)	1940.79	781.50
8	11.15	10.28	21.43	8.56	1.10 (1.27)	91.75 (73.33)	1966.09	785.70
10	11.22	10.35	21.58	8.71	1.10 (1.26)	93.00 (74.81)	2007.01	810.46
SEm±	0.16	0.19	0.18	0.13	0.01	0.78	26.89	14.83
CD (P=0.05)	NS	NS	NS	0.40	NS	NS	79.91	44.06
CV (%)	3.4	4.6	2.0	3.8	2.1	2.5	3.3	4.6

Table 6. Effect of nutri priming (P & Zn) at different concentrations on seedling vigour traits of paddy under laboratory conditions

P & Zn concentration (%)	Root length (cm)	Shoot length (cm)	Seedling length (cm)	Seedling dry weight (mg)	Root: Shoot ratio	Germination percentage (%)	Seedling Vigour Index-I	Seedling Vigour Index-II
0	10.34	9.83	20.63	8.23	1.09 (1.26)*	91.75 (73.33)**	1892.53	755.31
0.1,0.1	10.32	9.82	20.77	8.70	1.12 (1.27)	91.25 (72.81)	1895.30	795.13
0.5,0.1	11.24	10.50	21.75	9.07	1.08 (1.26)	94.00 (75.90)	2060.74	858.10
1.0,0.1	11.63	10.62	22.24	9.09	1.11 (1.27)	94.00 (75.92)	2090.69	859.89
0.5,0.5	11.43	10.52	21.96	9.10	1.10 (1.26)	94.00 (75.84)	2056.55	860.60
1.0,0.5	11.95	10.69	22.64	9.15	1.13 (1.28)	94.50 (76.51)	2114.19	864.70
SEm±	0.34	0.18	0.34	0.04	0.02	0.73	23.77	5.69
CD (P=0.05)	1.00	0.54	1.02	0.13	NS	2.17	70.63	16.90
CV (%)	7.2	4.2	3.8	1.1	3.0	2.3	2.8	1.3

Table 7. Effect of different seed priming agents at standardized concentrations on seedling vigour traits of paddy under laboratory conditions

Seed priming agent	Root length (cm)	Shoot length (cm)	Seedling length (cm)	Seedling dry weight (mg)	Root: Shoot ratio	Germination percentage (%)	Seedling Vigour Index-I	Seedling Vigour Index-II
T_1: No priming	10.08	9.34	19.43	7.60	1.10 (1.26)*	91.25 (72.84)**	1773.39	693.47
T_2: Hydro priming	10.79	9.83	20.63	8.23	1.09 (1.26)	91.75 (73.33)	1892.53	755.31
T_3: Nacl @ 1%	10.98	10.23	21.21	8.36	1.10 (1.26)	91.25 (72.81)	1935.65	763.00
T_4: Ascorbic acid @ 40 ppm	12.20	10.62	22.82	9.15	1.16 (1.29)	93.75 (75.53)	2139.47	856.78
T_5: Gibberellic acid @ 150 ppm	12.60	11.41	24.01	9.57	1.12 (1.27)	94.25 (76.21)	2262.12	901.60
T_6: Neem leaf extracts @ 2 %	10.88	10.21	21.21	8.43	1.07 (1.25)	91.25 (72.81)	1935.25	768.84
T_7: P & Zn (0.5&0.1%)	11.24	10.50	21.75	9.07	1.08 (1.26)	94.00 (75.90)	2060.74	858.10
SEm±	0.24	0.20	0.32	0.05	0.01	0.84	28.87	6.55
CD (P=0.05)	0.70	0.58	0.95	0.16	NS	2.49	84.92	19.27
CV (%)	4.2	3.8	3.0	1.2	1.7	2.7	2.9	1.6

**Figures in paranthesis are Square root transformed*
***Figures in paranthesis are Arc sine transformed*

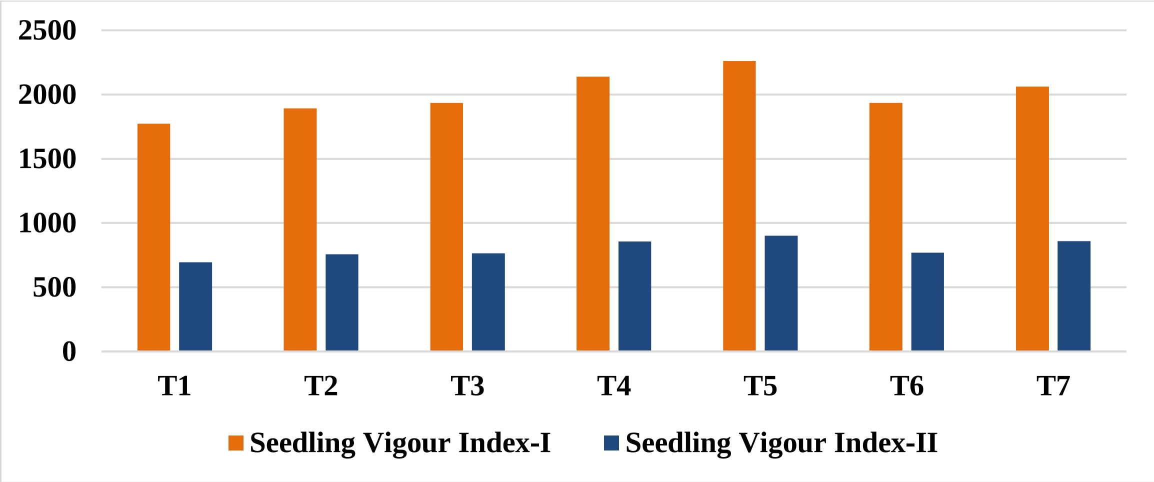

Fig. 1. Effect of different seed priming agents at standardized concentrations on seedling vigour traits of paddy under laboratory conditions

The data pertaining to the effect of different concentrations of nutri priming on seedling vigour traits of paddy was presented in the Table 6. Except root:shoot ratio, all the other vigour traits of the seedlings at various concentrations differed significantly. It was found that, 0.5 & 0.1, 1.0 & 0.1, 0.5 & 0.5, 1.0 & 0.5 % of P & Zn respectively were at par with each other and found to be superior over control and 0.1 & 0.1 % of P & Zn respectively taking into the consideration of optimal resource use, among all the concentrations, 0.5 & 0.1 % of P & Zn respectively was optimum concentration.

Standardized concentration of each priming agent obtained from Table.2 to.6 were analyzed statistically and the results were presented in Table.7, Fig 1. A perusal of the data indicates significant difference among different priming concentrations w.r.t all the vigour traits except root:shoot ratio. Among different priming treatments, vigour traits (root length, shoot length, seedling length, seedling dry weight, germination percentage, SVI-I and SVI-II) were found highest for the seeds primed with gibberellic acid @ 150 ppm. This was followed by ascorbic acid @ 40 ppm and 0.5 & 0.1 % of P & Zn which were at par with each other in terms of both the vigour indices. Next to these, neem leaf extract @ 2%, Nacl @ 1 % and hydro priming performed better and were at par with each other and least was found with the control w.r.t vigour indices.

The study revealed that seed priming with hormone (GA_3) followed by vitamin (ascorbic acid) and nutrients (P&Zn) contributed to a significant improvement in vigour of rice seedlings. In turn, these treatments are followed by priming with salt (Nacl), leaf extracts (Neem) and water which were found superior over control (no priming). Generally, when the seeds are primed with water, quick metabolic turn out in the seed leads to faster and vigorous growth of the seedlings. Further, beneficial effects are found when seeds are primed with chemicals such as gibberellic acid, ascorbic acid and nutrients. Faster increase of seedling length due to priming with GA_3 was already reported by Turner et al. [18], Helms et al. [19] and Kumar et al. [20]. Gibberellic acid triggers the production of higher levels of alpha-amylase which helps to break down the complex starch macromolecules of the seed into simpler sugars. Due to additional energy supplied through sugars, the primed seeds will germinate and produce vigorous seedlings. Whereas, adequate nutrient supply through nutripriming can improve seed germination and seedling vigour [21].

4. CONCLUSION

Seed priming techniques such as hydropriming, halopriming, vitamin priming, hormonal priming, botanical extract priming and nutripriming performed better in terms of inducing better vigour in the paddy seedlings compared no priming. Hormonal priming with gibberellic acid @ 150 ppm outperformed all the other seed priming techniques selected for paddy variety BPT 5204 in terms of seedling vigour traits. Thus, selection of better priming agent at right concentration is critical for achieving successful results.

COMPETING INTERESTS

Authors have declared that no competing interests exist.

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