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Influence of Seed Invigoration on Seed Quality in Upland Rice Genotypes

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ABSTRACT: A lab experiment was conducted to study the Influence of seed invigoration technique on seed quality in rice genotypes in the department of seed science and technology during 2020-21. Results revealed that among elite genotypes 'BD 1' and 'BA 4' found significantly superior in enzymatic activity and seed quality parameters. Among different treatments, the seed priming treatment with gibberllic acid @ 60 ppm for 12 hours recorded significantly higher values for enzymatic activity and seed quality parameters. Followed by seed treatment with ascorbic acid 10 ppm for 48 hours followed by seed treatment with azospirillum @ 1:50 dilution for 18 hours followed by seed treatment with distilled water for 48 hours and control. Among different treatments and genotypes combinations, seed priming treatment with gibberllic acid @ 60 ppm for 12 hours in genotype BD1 and BA4 registered significantly superior seed quality parameters. Decreased enzymatic activity *viz*., alpha amylase (1.98 cm to 0.38 cm), dehydrogenase (1.054 to 0.724 OD @ A_{480} nm), catalase (94.05 to 45.15 µmoles H_2O_2 decomposed/ min/g of seed fresh weight), peroxidase (5.91 to 2.78 µmoles/min/g of seed fresh weight) and SOD activity (22.27 to 12.59 Units/min/g seed fresh weight respectively) it is associated with decline in seed quality parameters *viz*., germination (88.50% to 77.5%), seedling shoot length (25.42cm to 13.3 cm), seedling root length (28.54 cm to 16.07 cm) and seedling dry weight (161.75 mg to 144.9 mg), field emergence (83.5% to 72.5%), seedling vigour index I (2374 to 1138), seedling vigour index II (4332 to 2371).

Keywords: Rice, gibberllic acid, ascorbic acid, azospirullum, priming

INTRODUCTION

Rice belongs to the Poaceae family, genus Oryza, which has 22 species, of which only two are cultivated *viz*, *O. sativa (*with sub species *Japonica)* and *O. glaberrima*. Rice is the primary staple food for billions of people, over two billion in Asia and hundreds of millions in Africa and Latin America are relying on rice for food. Asia is contemplated as the world's 'Rice Basket', as over 90% of rice produced and consumed in Asia, a continent with high population density. Rice is cultivated in 167.20 million hectares worldwide, yielding 769.6 million tonnes of rice [1]. In India it is grown on 43.77 million hectares with a production of 117.47 million tonnes and 2570 kg ha^{-1} yield respectively. In Karnataka rice is grown on 1.24 million hectares with 3.54 million tonnes of production and 2670 kg ha^{-1} of productivity per hectare [2].

Maintenance of seed quality from harvest to planting is of utmost importance for the seed industry. The seed quality depends on various factors *viz*., genotype, prevailing environmental conditions and practices during seed production, post-harvest operations, storage environment. Development of improved genotypes suited to upland and dry situation is one important strategy to enhance overall production. Constraints encountered include limited input of cultivation technology, especially in terms of the use of quality seeds and control techniques of plant-disturbing organisms. In addition, the decline in rice production is also attributed to the decrease in productive wetland due to the transfer of functions to the interests of industry, housing and other non-agricultural land uses. The development of upland rice in dry land can be a solution to increase production. However, upland rice received less attention because of its low productivity. Development of upland rice (especially local upland rice varieties) needs in addition to marginal land issues, implementation of cultivation techniques of upland rice, especially in the use of quality seeds. The use of high quality seeds is an important prerequisite for profitable crop production. Therefore, preparation and treatment of seeds to improve the quality is very important to, get

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uniform germination, healthy and vigorous seedling which can thrive well under dry situation.

In the recent past, many studies proved the efficiency of seed priming technique to enhance the germination rate, uniformity of seedling growth and to reduce the time of seedling emergence. Different seed priming treatments have been used to improve seed quality. Seed priming is a pre-sowing treatment in which seeds are soaked in water and or other chemicals for a definite period of time and then dried back to original moisture content before sowing. Seed priming stimulates various biochemical changes in seed which are essential to commence germination process besides improving its physiology [3]. It synchronizes germination after breaking dormancy, diminishes the lag time required for imbibition, hydrolyses or metabolises inhibitors, activates enzymes, mobilises reserved food and enhances embryonic tissue outgrowth. Starch metabolism is of great importance during seed priming which influences seedling vigour under stress. This metabolism is brought about by α-amylases which hydrolyse the starch reserves into metabolisable sugars providing energy to the developing embryo. Seed priming enhances α-amylase and dehydrogenase activity that could hydrolyse the starch macromolecules into smaller and simple sugars with increased ATP production and respiration. Priming is able to repair the age related cellular and sub-cellular damage of low vigour seeds that may accumulate during seed development [4]. Priming technique is the need of present time to get the enhanced germination and establishment in rice in order to utilize the soil moisture and solar radiation to a maximum extent. Seedling emergence from primed seeds is found to be more vigorous and faster as compared to unprimed seeds. Seed priming with growth regulators, plant extract, compatible solutes or inorganic salts causes improvement in germination.

This investigation was conducted to study the best priming treatment for quick and uniform emergence of paddy seeds. Seed quality parameters performance is investigated in this experiment by the use of seeds taken from aged seed lot, using priming techniques to increase the germination percentage. keeping these things in view, the present investigation entitled “Influence of seed invigoration on seed quality in upland rice genotypes” was carried out with the objective of Investigations on seed invigoration technique on seed quality in rice genotypes.

MATERIAL AND METHODS

An experiment entitled “Investigations on influence of seed invigoration on seed quality in upland rice genotypes”, was carried out during 2020 in the laboratary of seed science and technology department, College of Agriculture, University of agricultural sciences, Dharwad. The details of the materials used and methodology adopted during the course of investigation include 13 rice genotypes viz BA3 (G_1), BA 4 (G_2), BA 5 (G_3), BA 6 (G_4), BA 10 (G_5), BD 1 (G_6), BD 4 (G_7), BD 5 (G_8), BD 6 (G_9), BD 7 (G_{10}), BPT 5204 (G_{11}), Antrasali (G_{12}) and MGD 101 (G_{13}) and five priming treatments namely P_1: GA_3 60 ppm for 12 hours, P_2 :Ascorbic acid 10 ppm for 48 hours, P_3 : Azospirillum @ 1:50 dilution for 18 hours, P_4 : Distilled water for 48 hours, P_5: Control. The observation on seed quality parameters were recorded and the results were subjected to statistical analysis adopting factorial completely randomized design.

Methodology

1. Preparation of chemical solution

Chemical solutions, GA_3 (60 ppm) 60 mg of chemical was added to alcohol because GA_3 does not dissolve readily in water it hence to be dissolved in and volume have to be made up 1000 ml by adding distilled water. and Ascorbic acid (10 ppm) 10 mg of chemical was added with 1000 ml water.

2. Catalase (CAT) activity

Catalase activity was measured at 25 °C according to Aebi (6) with some minor modifications. The UV (Ultra Violet) light absorbance of hydrogen peroxide solution can be measured between 230 and 250 nm. On decomposition of hydrogen peroxide by Catalase, the absorption decreases with time.

Three ml of reaction mixture contains 50 µl enzyme extract, 1.5 ml of 100 mM Phosphate buffer (pH 7.0), 0.5 ml of 75 mM H_2O_2 and 950 µl of distilled water. The control contains enzyme extract and phosphate buffer devoid of H_2O_2. Catalase activity was estimated by the decrease in absorbance of H_2O_2 at 240 nm and was expressed as µmol H_2O_2 decomposed/min/g seed fresh weight.

3. Peroxidase (POX) activity

Peroxidase activity was assayed as increase in optical density due to oxidation of guaiacol to tetra-guaiacol by following Castillo *et al*. [7] with minor modifications at 470 nm absorbance using a reaction mixture containing 12

mM hydrogen peroxide and 96 mM guaiacol in phosphate buffer pH 7.0.

Three ml reaction mixture contains one ml of 100 mM phosphate buffer (pH 7.0), 0.5 ml each of 96 mM Guaiacol and 12 mM H_2O_2, 50 µl of enzyme extract and 950 µl of distilled water. Absorbance due to the formation of tetra-guaiacol was recorded at 470 nm and enzyme activity was calculated as per the extinction coefficient of its oxidation product, tetra –guaiacol E= 26.6 nM/cm. Enzyme activity was expressed as µmoles/cm/min/g seed fresh weight.

4. Superoxide dismutase (SOD) activity

Superoxide dismutase activity was measured according to Beauchamp and Fridovich [8] with some minor modifications.

One gram of decoated seed was homogenized in 15 ml of 100 mM Potassium phosphate buffer (pH 7.8) with a pinch of PVP (Poly Vinyl Pyrrolidone). The extract was centrifuged at 15000 rpm for 10 min at 4 °C. The supernatant was collected and used as enzyme extract.

One ml of 100 mM potassium phosphate buffer (pH 7.8); 100 µl of 2.25 mM nitro blue tetrazolium (NBT); 100 µl of 3 mM Ethylene Diamine Tetra acetic Acid (EDTA); 200 µl of 200 mM L-methionine and 1.75 ml distilled water; 200 µl enzyme extract and 150 µl 0.075 mM riboflavin was mixed in test tubes and shaken properly. Glass test tubes containing the reaction mixture was then placed at a distance of 30 cm below two 15 W fluorescent lamps for 15 min. switching off the light stopped the reaction and the tubes was immediately covered with a black cloth. A non-irradiated reaction mixture containing enzyme extract, which does not develop colour, was used as blank. Control was lacking enzyme in reaction mixture and developed maximum colour.

The absorbance at 560 nm was recorded in spectrophotometer. One unit of SOD was defined as the enzyme activity which inhibited the photo reduction of NBT to blue formazan by 50% and SOD activity of the extracts was expressed as Units/min/g seed fresh weight.

5. α amylase activity

Pipette out 1ml of starch solution and 1 ml of properly diluted enzymes in a test tube and incubated @ 270C for 15 min. Stopped the reaction by the addition of 2ml of dinitrosalicylic acid reagent and heat the solution in a boiling water-bath for 5 min. While the tubes are warm add 1 ml potassium sodium tartrate solution and cool it under running tap water. Madeup the volume to 10ml by addition of 6ml water. Read at absorbance at 560nm and terminate the reaction at zero time in the control tubes. The absorbance read from the spectrophotometer was converted into α-amylase activities using a modified form of the formula used by Beleia and Varriano-Marston [9]. One unit of α-amylase was defined as the amount of micromoles of maltose was produced per milliliter of α-amylase solution per minute under the conditions of test.

$$\text{Activities (U/ml)} = \frac{[\text{mg/ ml (maltose)} * 10^3]}{[\text{Mw.maltose} * \text{time(min)}]} \times [2]$$

The alpha amylase activity was analyzed as per the method suggested by Simpson and Naylor (1962). Two gram of agar shreds and one gram of potato starch was mixed together in water to form paste and volume was made up to 100 ml with distilled water. The homogenous solution of agar-starch mixture after boiling was poured into sterilized petri-dishes and allowed to settle in the form of gel after cooling. The pre-soaked (for 8 hr) and half cut seeds (with their half endosperm and embryo portion intact) were placed In the petri-dishes in such a way that the endospermic part remained in contact with agar-starch gel. The petri-dishes was closed and kept in dark at 30°C. After 48 hr the petri-dishes were uniformly smeared with potassium iodine solution (0.44 g of iodide in 500ml distilled water) and excess solution was drained off after few minutes. The diameter of hallow (clear) zone formed around the seed was measured in mm and reported as alpha amylase activity.

6. Dehydrogenase activity

Dehydrogenase activity was determined by using the tetrazolium (TTC) staining method. Twenty five representative seeds from each treatment in two replication was taken and preconditioned by soaking in water overnight at room temperature. Embryos was exercised from the seeds and was steeped in 0.5 per cent solution of 2, 3, 5 triphenyltetrazolium chloride and kept in dark for two hours at 40°C for staining. The stained seeds were thoroughly washed with water and then soaked in 10ml of 2 methoxy ethanol (methyl cellosolve) and kept overnight the change in colour was observed to red colour forming formazan. The intensity of red colour was measured using UV-VIS spectrophotometer using blue filter at 470 nm wave length and methoxy ethanol will be used as blank. The OD value obtained was recorded [10].

RESULTS AND DISCUSSION

Among elite genotypes 'BA 4, BD 1' registerd significantly higher (0.868, 0.867 OD @ A_{480} nm respectively) level of dehydrogenase activity and α amylase activity (1.298, 1.32 cm respectively) associated with higher values for seed germination (84.8, 84.8% respectively), seedling wet weight (824.1, 824.35mg respectively), seedling dry weight (155.18, 155.3 mg respectively), seedling vigour index I (1924, 1986 respectively), seedling vigour index II (3519, 3635 respectively) and field emergence (79.5, 79.3% respectively) were superior in over other elite genotypes. However, they were on par with the check which registered 'Antrasali' which registered higher (0.864 OD @ A_{480} nm) level of dehydrogenase activity and α amylase activity (1.302 cm) associated with higher values for seed germination (84.7%), seedling wet weight (824.3 mg), seedling dry weight (155.14 mg), seedling vigour index I (1942), seedling vigour index II (3555) and field emergence (79.2%), as compared to other checks.

Among the different elite genotypes, recorded the higher Catalase activity in "BD 1, BA 4" followed by check "Antrasali" recorded significantly higher catalase activity (77.73, 76.92 and 75.4 H_2O_2 decomposed/min/g of seed fresh weight respectively). Whereas, the lowest Catalase activity was recorded in "BA 6, BD 7 and check BPT 5204" (57.99, 58.66 and 60.39 H_2O_2 decomposed/min/g of seed fresh weight respectively). Similarly with respect to peroxidase activity in "BD 1, BA 4, check Antrasali" recorded significantly higher peroxidase activity (4.44, 4.42, 4.41 µmoles/min/g of seed fresh weight respectively). Whereas, the lowest peroxidase activity was recorded in check "BPT 5204, BA 6 and BD 7" (3.66, 3.71 and 3.72 µmoles/min/g of seed fresh weight respectively). Similarly with respect to SOD activity in "BA 4, BD 1, BD 4 and check Antrasali" recorded significantly higher SOD activity (20.06, 19.81, 19.51 and 19.50 Units/min/g seed fresh weight respectively). Whereas, the lowest SOD activity was recorded in "BA 6, BD 7 and check BPT 5204" (15.26, 15.77 and 15.51 Units/min/g seed fresh weight respectively).

The seed invigoration treatment with gibberlic acid @ 60 ppm for 12 hours recorded significantly higher (0.995 OD @ A_{480} nm) value for dehydrogenase activity, α amylase activity (1.807 cm), catalase activity (84.27 µmoles H_2O_2 decomposed/ min/g of seed fresh weight), peroxidase activity (5.43 µmoles/min/g of seed fresh weight) and SOD activity (20.71 Units/min/g seed fresh weight) associated with higher values for germination (84.69%), seedling root length (25.28 cm), seedling shoot length (23.13cm), seedling wet weight (844.44 mg/10 seedlings), seedling dry weight (159.07 mg/ 10 seedlings), seedling vigour index (2054), seedling vigour index II (3853) and field emergence (79.62%). GA_3 plays a vital role in the endosperm cap weakening. A high level of gibberellin is needed for the counteraction of ABA activity in seeds to promote dormancy release and radical protrusion during seed germination [11]. The embryo produces bioactive which are then transported to an aleurone layer, triggering the expression of alpha-amylase. During seed germination, the aleurone layer is unable to synthesize GA_3 but perceives the GA_3 signals. Through the exogenous application of GA_3, the expression of alpha-amylase gene is upregulated, increasing seed vigor, thus leading to seed germination [12].

Followed by the seed invigoration treatment with Ascorbic acid 10 ppm for 48 hours recorded significantly higher (0.866 OD @ A_{480} nm) value for dehydrogenase activity, α amylase activity (1.35 cm), catalase activity (77.91 H_2O_2 decomposed/min/g of seed fresh weight), peroxidase activity (4.19 µmoles/min/g of seed fresh weight) and SOD activity (19.08 Units/min/g seed fresh weight) associated with higher values for germination (83.08%), seedling root length (23.86 cm), seedling shoot length (20.83 cm), seedling wet weight (829.82 mg/ 10 seedling), seedling dry weight (155.72 mg/ 10 seedling), seedling vigour index I (1859), seedling vigour index II (3482) and field emergence (77.54%). Mondal *et al*. [13] observed that the effect of seed priming in rice was co-related with an enhancemnt in endospermic amylase activity resulting from the increase insoluble sugar content of the primed seeds.

Dehydrogenase activity is an indicator of root vitality and used as comprehensive assessment index reflecting the metabolic activity level and root's ability to absorb nutrients and water enhanced source to sink relation leads to better accumulation of food reserves like protein and carbohydrates [14], higher the dehydrogenase activity higher is the germination. These result is in accordance with findings of Mahakham *et al.* [14] in paddy seeds.

Iqbal *et al*. [15] reported that during seed deterioration, free radicals produced as a result of lipid peroxidation cause damage to the enzymes required to convert the reserve food into a usable form in the embryo and thus affect the development of normal seedling and these free radicals also degrade the mitochondrial membrane

Table 1. Influence of seed invigoration treatments on α-amylase, Dehydrogenase, catalase, peroxidase and super oxide dismutase (SOD) enzymatic activity in rice genotypes

Treatments	α-amylase (cm)	Dehydrogenase (OD @ A_{480} nm)	Catalase (µmoles H_2O_2 decomposed/ min/g of seed fresh weight)	Peroxidase (µmoles/cm/min/ g of seed fresh weight)	SOD (Units/min/g seed fresh weight)
Genotypes (G)					
G_1 : BA 3	1.10	0.845	73.55	4.15	19.18
G_2 : BA 4	1.30	0.868	76.92	4.42	20.06
G_3: BA 5	1.03	0.835	72.67	4.04	17.67
G_4: BA 6	1.21	0.812	57.99	3.71	15.26
G_5: BA 10	1.00	0.825	67.67	3.88	16.14
G_6: BD 1	1.32	0.867	77.73	4.44	19.81
G_7: BD 4	1.14	0.842	74.87	4.25	19.51
G_8: BD 5	1.02	0.824	69.43	3.87	16.73
G_9: BD 6	1.08	0.833	73.17	4.09	18.57
G_{10}: BD 7	1.08	0.813	60.39	3.72	15.77
G_{11}: BPT 5204	0.88	0.816	58.66	3.66	15.51
G_{12}: Antrasali	1.30	0.864	75.40	4.41	19.50
G_{13}: MGD 101	0.98	0.832	70.84	3.94	16.66
Mean	1.11	0.837	69.95	4.04	17.72
S. Em.±	0.07	0.005	0.33	0.009	0.196
CD 1%	0.26	0.02	1.25	0.035	0.735
Invigoration treatments (T)					
T_i: Control	0.49	0.740	52.99	3.08	14.99
T_2: Hydro priming	0.72	0.772	60.25	3.56	15.86
T_3: Bio priming	1.19	0.810	74.32	3.97	17.96
T_4: Ascorbic acid priming	1.35	0.866	77.91	4.19	19.08
T_5: GA_3 priming	1.81	0.995	84.20	5.48	20.71
Mean	1.11	0.837	69.95	4.04	17.72
S. Em.±	0.04	0.003	0.21	0.006	0.12
CD@ 1%	0.16	0.01	0.78	0.02	0.46
Interaction effect (T × G)					
T_1G_1	0.44	0.749	54.99	3.06	16.27
T_1G_2	0.60	0.758	57.63	3.33	18.19
T_1G_3	0.43	0.740	54.59	3.10	15.11
T_1G_4	0.41	0.724	45.15	2.87	12.59
T_1G_5	0.42	0.735	54.26	3.01	13.50
T_1G_6	0.60	0.758	57.98	3.35	16.89
T_1G_7	0.53	0.745	55.75	3.21	16.53
T_1G_8	0.47	0.733	52.95	2.91	14.51
T_1G_9	0.52	0.738	54.67	3.04	15.06
T_1G_{10}	0.52	0.729	46.24	2.78	12.93
T_1G_{11}	0.38	0.727	45.46	2.86	12.91
T_1G_{12}	0.61	0.755	56.22	3.33	16.70
T_1G_{13}	0.49	0.739	52.99	3.12	13.65
T_2G_1	0.69	0.776	62.99	3.53	18.00
T_2G_2	0.86	0.795	65.49	3.90	19.50
T_2G_3	0.67	0.770	61.99	3.61	15.37
T_2G_4	0.63	0.756	51.11	3.37	13.10
T_2G_5	0.65	0.764	60.19	3.49	13.91
T_2G_6	0.87	0.793	65.97	3.91	18.29
T_2G_7	0.75	0.779	63.39	3.69	17.89
T_2G_8	0.70	0.764	60.35	3.30	14.06

Contd...

Treatments	α-amylase (cm)	Dehydrogenase (OD @ A_{480} nm)	Catalase (µmoles H_2O_2 decomposed/ min/g of seed fresh weight)	Peroxidase (µmoles/cm/min/ g of seed fresh weight)	SOD (Units/min/g seed fresh weight)
T_2G_9	0.71	0.772	62.81	3.58	17.12
T_2G_{10}	0.71	0.756	52.39	3.20	13.69
T_2G_{11}	0.57	0.756	51.47	3.24	13.58
T_2G_{12}	0.86	0.788	63.95	3.89	17.28
T_2G_{13}	0.63	0.762	61.10	3.52	14.40
T_3G_1	1.22	0.831	78.43	4.09	19.19
T_3G_2	1.45	0.839	82.34	4.31	19.95
T_3G_3	1.15	0.814	77.77	3.97	17.53
T_3G_4	1.02	0.782	61.24	3.64	15.12
T_3G_5	1.10	0.796	69.82	3.80	15.99
T_3G_6	1.47	0.840	83.41	4.33	20.31
T_3G_7	1.23	0.831	80.91	4.15	20.30
T_3G_8	1.11	0.796	73.14	3.86	16.84
T_3G_9	1.16	0.817	77.98	3.98	19.74
T_3G_{10}	1.16	0.783	62.96	3.73	15.61
T_3G_{11}	0.94	0.780	61.46	3.64	15.28
T_3G_{12}	1.43	0.839	81.46	4.29	20.65
T_3G_{13}	1.08	0.790	75.21	3.82	17.07
T_4G_1	1.39	0.878	82.25	4.41	21.08
T_4G_2	1.62	0.895	86.16	4.67	20.40
T_4G_3	1.26	0.870	81.59	4.06	19.91
T_4G_4	1.20	0.835	63.56	3.70	16.39
T_4G_5	1.24	0.850	73.64	3.92	17.15
T_4G_6	1.68	0.895	87.23	4.71	21.36
T_4G_7	1.41	0.867	83.73	4.48	21.14
T_4G_8	1.26	0.847	76.96	4.05	18.04
T_4G_9	1.30	0.855	81.80	4.29	20.20
T_4G_{10}	1.30	0.833	66.78	3.83	17.05
T_4G_{11}	1.08	0.846	65.78	3.73	16.58
T_4G_{12}	1.65	0.896	84.28	4.63	20.69
T_4G_{13}	1.13	0.892	79.03	4.02	18.01
T_5G_1	1.75	0.990	89.07	5.67	21.35
T_5G_2	1.96	1.054	92.98	5.86	22.27
T_5G_3	1.66	0.984	87.41	5.49	20.42
T_5G_4	2.81	0.965	68.88	4.95	19.13
T_5G_5	1.59	0.979	80.46	5.18	20.16
T_5G_6	1.98	1.051	94.05	5.89	22.19
T_5G_7	1.78	0.990	90.55	5.70	21.66
T_5G_8	1.58	0.979	83.78	5.24	20.21
T_5G_9	1.71	0.983	88.62	5.55	20.71
T_5G_{10}	1.71	0.967	73.60	5.06	19.57
T_5G_{11}	1.43	0.969	69.10	4.84	19.20
T_5G_{12}	1.96	1.044	91.10	5.91	22.19
T_5G_{13}	1.57	0.977	85.85	5.24	20.19
Mean	1.11	0.837	69.95	4.04	17.72
S. Em.±	0.15	0.01	0.75	0.02	0.44
CD@ 1%	0.25	0.017	1.25	0.04	0.74

T_1 : Control T_2 : Distilled water for 48 hours

T_3 : Azospirillum @ 1:50 dilution for 18 hours T_4 : Ascorbic acid 10 ppm for 48 hours T_5: GA_3 60 ppm for 12 hours

resulting in a reduction in the energy supply needed for germination, resulting in seed germination failure, but after the treating seeds with invigoration technique like priming, priming leads to production of scavenger enzymes like alpha amylase, dehydrogenase, catalase, peroxidase, SOD, etc. these enzymes detoxifies the free radicles, stabilize the membrane integrity, convert food reserve into usable form in the embryo and thus leads to development of normal seedlings [15].

Seed bio invigoration treatment with Azospirillum @ 1:50 dilution for 18 hours recorded significantly higher (0.8103 OD @ A_{480} nm) value for dehydrogenase activity, α amylase activity (1.194 cm), catalase activity (74.32 H_2O_2 decomposed/min/g of seed fresh weight), peroxidase activity (3.97 µmoles/min/g of seed fresh weight) and SOD activity (17.97 Units/min/g seed fresh weight) associated with higher values for germination (82.12%), seedling root length (22.28 cm), seedling shoot length (18.78 cm), seedling wet weight (823.82 mg/ 10 seedling), seedling dry weight (152.49 mg/ 10 seedlings), seedling vigour index I (1688), seedling vigour index II (3132) and field emergence (76.50%) compare to untreated seeds that is without priming treatment seeds recorded significantly lower (0.741 OD @ A_{480} nm) level of dehydrogenase activity, α amylase activity (0.494 cm), catalase activity (52.99 H_2O_2 decomposed/min/g of seed fresh weight), peroxidase activity (3.08 µmoles/min/g of seed fresh weight) and SOD activity (14.99 Units/min/g seed fresh weight) associated with lower values for germination (78.42%), seedling root length (19.10 cm), seedling shoot length (16.18 cm), seedling wet weight (803.58 mg/ 10 seedling), seedling dry weight (147.61 mg/ 10 seedling), seedling vigour index I (1385), seedling vigour index ^{22} (2606) and field emergence (73.35%) respectively.

The bioagents might have enhanced the shelf life of the genotype to some extent as it is evident from the dehydrogenase activity and also they are known to aid in maturity of embryo and better seed filling and development. These results are in accordance with previous reports in chick pea [16] and rice [17, 18].

Among different invigoration treatment and genotypes combinations, the seed priming treatment with gibberlic acid @ 60 ppm for 12 hours in genotype 'BD 1', 'BA 4' and 'check Antrasali' registered significantly highest (1.051, 1.054 and 1.044 OD @ A_{480} nm respectively) amount of dehydrogenase activity and α amylase activity (1.98 cm, 1.96 cm and 1.96 cm respectively) associated with higher seed germination (88.5%, 88% and 88.5% respectively), wet weight (846.6, 846.6and 846.5 mg respectively), seedling dry weight (161.75, 161.75 and 161.6 mg respectively), seedling vigour index I (2341, 2313 and 2374 respectively), seedling vigour index II (4279, 4279 and 4333 respectively) as compared to other invigoration treatment and genotypes combinations. The present findings may be attributed to better growth and development, promoted by the combined effect of method of priming, genotype and genetic factor.

Among different invigoration treatment and genotypes combinations, Higher Catalase activity was recorded in GA_3 priming @ 60 ppm for 12 hours in genotypes 'BD 1', 'BA 4' and ' check Antrasali' (92.98, 94.05 and 91.10 H_2O_2 decomposed/min/g of seed fresh weight respectively). Similarly with respect to peroxidise activity GA_3 priming @ 60 ppm for 12 hours in genotypes 'BD 1', 'BA 4' and ' check Antrasali' were recorded higher peroxidise activity (5.86, 5.89 and 5.91 µmoles/min/g of seed fresh weight respectively). Similarly with respect to SOD activity GA_3 priming @ 60 ppm for 12 hours in genotypes 'BD 1', 'BA 4' and ' check Antrasali' were recorded higher SOD activity (22.27, 22.19 and 22.19 Units/min/g seed fresh weight respectively). GA_3 induces alpha-amylase activities in early post-germination growth of rice, which is consistent with the increasing growth of coleoptile, mesocotyl, and first leaf. It also enhances catalase, peroxidase and SOD simultaneous production and consumption, where starch degradation in the seed and utilization of soluble sugars support seedling growth [19].

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