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Effect of seed priming on biochemical changes in rice under anaerobic condition

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Abstract
Present investigation was carried out to study the "Effect of seed priming on anerobic germination of rice" during wet season, 2016-17. Experiment was laid out in randomized block design with three replication and two varieties Sambha Mahsuri and Sambha Mahsuri Sub1 in cemented pond (size; 20×20×1.25 meter). Primed seeds were direct seeded and field was completely submerged/ flooded, up to 25-30 cm water depth was maintained for one month. Recommended dose of N, P and K @ 120:40:40 Kg ha-1 was reframed with time schedule (1/4 dose of N and full dose of P2O5 and K2O were applied as basal and rest N was applied in there splits i.e. 30, 60 and 90 DAS respectively). Treatments comprised of (T1) Seed priming with GA3 @ 25 ppm, (T2) Seed priming with GA3 @ 50 ppm, (T3) Seed priming with JLE @ 2%, (T4) Seed priming with KNO3@ 0.5%, (T5) Seed priming with KCl @ 0.2%, (T6) Seed priming with NaCl @ 0. 5%, (T7) Seed priming with IAA @ 0.2%, (T8) Seed priming with CaCl2 @ 0.1%, (T9) Control with distilled water.
Results indicated that all the priming treatments increased the biochemical parameters like total sugar content, reducing and non reducing sugar, protein content and increases days to effect of GA3 @ 50 ppm was found more pronounced followed by GA3 @ 25 ppm on various parameters in both the variety but Sambha Mahsuri Sub1 showed more response of seed priming as compared to that of Sambha Mahsuri.
Seed priming with various chemicals and its concentration may be used as a tool to mitigate the adverse effect caused by anaerobic condition in rice or direct seeded rice early flooding in recurrent phenomena.

Keywords: Mustard, GA_3, SA, and ABA

Introduction

Rice (*Oryza sativa* L., 2n= 24), belongs to the family Poaceae (Graminae). Rice farming is about 10,000 year old and largest single use of land for producing food. Rice fields covers 11% of Earth's entire aerable land. Two rice species are important cereals for human nutrition *i.e. Oryza sativa* grown worldwide and *Oryza glaberrima* grown in parts of West Africa. Varieties of growth duration ranging from 70 to 160 days exist in diverse environments. Rice is the most important cereal food crop of India. It occupies about 23.3% of gross cropped area of the country and plays vital role in the national food grain supply.

Traditionally rice is grown by transplanting nursery seedlings into the puddle field, which requires a continuous supply of water throughout its growth (Farooq *et al.*, 2007) [1]. In addition to high water inputs, it demands a high labour cost, particularly at the critical time of transplanting, which not only increases the cost of production but can also result in delayed transplanting due to labour unavailability (Farooq *et al.*, 2011) [2].

Submergence stress is a major constraint to rice production during the monsoon flooding season in the rainfed lowlands in South and Southeast Asia, which causes annual losses of over US$1 billion and affects disproportionately the poorest farmers in the world (Xu *et al.*, 2006) [3]. Excessive flooding poses risks to human life and is a major contributor to the poverty and vulnerability of marginalized communities especially women and children in poor families (Douglas *et al.* 2009) [4]. It is estimated that the flood-affected area has more than doubled in size from about 5% (19 million hectares) to about 12% (40 million hectares) of India's geographic area. Adding to these already high risk areas, the climate projections suggest that temperatures, precipitation and flooding and sea level rise are likely to increase, with adverse impacts on crop yield and farm income in Southeast Asia (Unnikrishnan *et al.*, 2006) [5] By flooding during submergence, plant germination is greatly affected by depth of water and by its physico-chemical characteristics (oxygen and carbon dioxide concentration, pH, turbidity, temperature etc.

Seed priming is a simple and low cost hydration technique in which seeds are partially hydrated to a point where pre-germination metabolic activities start without actual germination and then re-dried until close to the original dry weight.

Seed priming is employed for better crop stand and higher yields in a range of crops including rice (Farooq *et al*., 2009; Kaymak *et al*., 2009) [6, 7].

Primed seeds when seeded usually emerge faster with better, uniform and vigorous crop stand persistent under less than optimum field conditions. Crop stands from primed seeds lead to earlier flowering and higher grain yield than non-primed seeds. In some other studies, Farooq *et al.* (2006) [8] reported that early emergence and seedling growth, better crop stand, allometric response, increased kernel yield, harvest index, and improved quality from seeds primed with KCl and $CaCl_2$ in coarse and fine rice, respectively.

Priming has been used to improve the performance of germination at the field and potassium nitrate (KNO_3) is a promising compound for this purpose. Besides, the priming could also activate the response of the antioxidant system, becoming the primed seeds more prepared for possible stresses Lara *et al*. (2014) [9].

Rehman *et al*., (2014) [10] conducted an experiment to study the seed priming influence on early crop growth, phonological development and yield performance of Linola in Pakistan. Seeds were treated with 50 mmol/L salicylic acid, 2.2% $CaCl_2$ and 3.3% Moringa Leaf Extract (MLE) including untreated dry and hydro-priming control. Results showed that osmo-priming with $CaCl_2$ reduced emergence time and produce the highest seedling fresh and dry weight including chl.a content. It also reduced crop branching and flowering and maturity time and had the maximum plant height, number of branches, tillers, pods and seeds per pod followed by MLE. An increase in seed weight, biological and seed yields were 9.30, 34.16 and 39.49%, harvest index (4.12%) and oil content (13.39%) with $CaCl_2$ osmo priming. It concludes that seed osmo-priming with $CaCl_2$ and MLE can play significant role to improve early crop growth and seed yield of linola.

Materials and methods

The present investigation was carried out in Kharif season, during 2015-2016 at the Research Farm of Department of Crop Physiology, Narendra Deva University of Agriculture and Technology, Kumarganj, 224229; Faizabad (U.P.) India. The present site is located at main campus of University. The geographical situation of Faizabad district lies between a latitude of 26047' north and longitude of 82012' east, on altitude of 113 meters above sea level in the gangetic alluvium of eastern Uttar Pradesh, India. The Faizabad district falls in semiarid zone, receiving a mean annual rainfall of above 1100 mm, in which about 80% of the total precipitation is required during monsoon season, July to end of September with few showers in winter. The winter months are cold and occasionally frosts occurs during this period and summers are hot and dry. The total soluble sugar of, 2nd, 4th, 6th and 8th, 10th, 12th, 14th, 18th, 18th day old seedling was estimated by the method described by Yemm and Willis (1954)

Sulphuric acid hydrolyse glycosydic bonds of carbohydrate which produces monosaccharides which are dehydrate to furfural and its derivatives. Furfural reacts with anthrone to give a blue-green complex.

The reducing sugar of 2nd, 4th, 6th and 8th, 10th, 12th, 14th, 18th, 18th day old seedling was estimated by the following method Dubois *et al*., (1951).

Dinitrosalicylic acid reagent (DNS) reagent was prepared by dissolving 1.0 gm DNS in 20 ml NaOH.

The non reducing sugar of 2nd, 4th, 6th and 8th, 10th, 12th, 14th, 18th, 18th day old seedling was calculated by subtracting the amount of reducing sugar from the total sugar.

Results and discussion

The perusal of data of effect of seed priming on total soluble sugar content in two rice varieties i.e. Sambha Mahsuri Sub 1 and Sambha Mahsuri under anaerobic condition were recorded at different time intervals have been presented in Table-4.2.1(a). 4.2.1 (b) show that total sugar content increased with time and at 18th day of germination maximum soluble sugar content was observed in their respective treatments. Priming with different chemical and its concentration enhance the total soluble sugar content in both varieties however Sambha Mahsuri Sub 1 showed higher effect of seed priming as compared to that of Sambha Mahsuri. Among various seed priming treatment in both the variety (Sambha Mahsuri Sub 1 and Sambha Mahsuri) maximum soluble sugar content was found in GA3 @ 50 ppm (50.06, 47.93 respectively) followed by GA3 @ 25 ppm (49.89, 47.16 respectively) while the minimum total soluble sugar content was noticed in IAA @ 0.2% (48.41, 46.74) as compared to all other seed priming treatments.

The data pertaining to effect of seed priming on reducing sugar content in two rice varieties i.e. Sambha mahsuri Sub 1 and Sambha mahsuri under anaerobic condition are recorded at different time intervals have been presented in Table-4.2.2(a), 4.2.2 (b) and show that reducing sugar content increased with the increase in time intervals in both the varieties and maximum was found at 18th day in both the variety in their respective treatments. Different seed priming treatment increases the reducing sugar contents in both the varieties (Sambha mahsuri Sub 1 and Sambha Mahsuri) however the magnitude of increase in reducing sugar content was higher in Sambha mahsuri Sub 1 as compared to Sambha mahsuri in all the treatments.

Among various priming treatments increased the reducing sugar content in both the varieties but in Sambha Mahsuri Sub 1 but effect of GA3 @ 50 (19.95, 18.33) ppm was more effective followed by GA3 @ 25 (19.62, 18.01) ppm while the minimum increase in reducing sugar. The data presented in Table 4.2.3 (a), 4.2.3 (b) clearly indicate that all the seed priming treatments increases the non-reducing sugar content in both the varieties i.e. Sambha mahsuri Sub 1 and Sambha mahsuri under anaerobic condition. The data show that non-reducing sugar content increased with the increase in time intervals and maximum was found at 18th day in both the varieties in their respective treatments. Different seed priming treatments increases the non-reducing sugar contents in both the varieties (Sambha mahsuri Sub 1 and Sambha Mahsuri) however the magnitude of increase in non-reducing sugar content was higher in Sambha mahsuri Sub 1 as compared to Sambha mahsuri in all the treatments.

In both the varieties (Sambha mahsuri Sub 1 and Sambha Mahsuri), the effect of GA3 @ 50 ppm (29.90, 29.10) was more effective followed by GA3 @ 25 ppm (29.07, 28.70) while the minimum increase in non-reducing sugar content was noticed in IAA @ 0.2% (27.67, 27.40) as compared to other seed priming treatments.

Table 4.2.1 (a): Effect of seed priming on total sugar content (mg g $^{-1}$ dry wt.) Under anaerobic condition on rice seedling

Samba Mahsuri Sub-1									
Treatments\ Days	**2**	**4**	**6**	**8**	**10**	**12**	**14**	**16**	**18**
T_1GA_3@25ppm	44.46	44.84	45.92	46.52	47.43	47.70	48.7	49.28	49.89
T_2GA_3@50ppm	44.72	45.43	45.95	46.76	47.19	47.31	47.45	49.66	50.06
T_3JLE@2%	43.66	43.96	44.84	45.18	45.23	46.41	47.97	48.19	48.94
T_4KNO_3@0.5%	44.23	44.93	45.45	45.99	46.57	46.73	47.58	48.48	48.97
T_5KCL@0.2%	44.07	44.95	45.35	45.84	46.41	46.90	47.24	48.15	49.28
T_6NaCL@0.5%	43.59	44.69	45.23	45.92	46.21	46.73	47.29	47.66	49.68
T_7IAA@0.2%	44.60	44.95	45.30	45.90	46.40	46.95	47.20	47.78	48.41
T_8CaCL_2@0.1%	43.87	44.55	45.08	45.88	46.18	46.82	47.18	47.74	48.68
T_9Control	43.80	43.99	44.67	45.12	45.98	46.46	47.18	47.97	48.39
SEm±	0.17	0.16	0.07	0.06	0.17	0.17	0.17	0.09	4.96
CD or LSD	0.50	0.48	0.20	0.18	0.52	0.52	0.52	0.28	14.87

Table 4.2.1 (b): Effect of seed priming on total sugar content (mg g-1 dry wt.) Under anaerobic condition on rice seedling

Samba Mahsuri									
Treatments\Days	**2**	**4**	**6**	**8**	**10**	**12**	**14**	**16**	**18**
T_1GA_3@25ppm	42.72	43.15	44.15	44.41	45.10	46.44	46.63	46.68	47.16
T_2GA_3@50ppm	42.57	43.56	44.37	44.97	45.43	45.44	46.11	46.78	47.93
T_3JLE@2%	41.85	42.57	42.96	43.31	43.75	44.34	44.42	44.59	46.57
T_4KNO_3@0.5%	42.02	42.63	43.29	43.60	44.70	44.97	46.19	46.38	46.60
T_5KCl@0.2%	42.46	42.81	43.27	43.76	44.15	45.71	45.95	46.11	46.16
T_6NaCl@0.5%	41.47	41.60	41.95	42.20	42.45	42.90	43.40	44.50	45.90
T_7IAA@0.2%	42.41	42.94	42.98	43.39	44.35	45.57	45.88	46.06	46.74
T_8CaCl_2@0.1%	42.29	42.38	42.74	43.72	44.08	45.37	45.60	45.73	46.61
T_9Control	41.68	42.69	43.18	43.22	43.26	44.28	44.54	44.66	44.72
SEm±	0.31	0.25	0.45	0.41	0.29	0.51	0.35	0.38	0.51
CD or LSD	0.94	0.785	1.34	1.23	0.86	1.52	1.05	1.14	1.54

Table 4.2.2 (a): Effect of seed priming on reducing sugar content (mg g-1 dry wt.) Under anaerobic condition on rice seedling

Samba Mahsuri Sub -1									
Treatments\Days	**2**	**4**	**6**	**8**	**10**	**12**	**14**	**16**	**18**
T_1GA_3@25ppm	15.73	16.29	16.82	17.12	17.74	18.32	18.99	19.45	19.62
T_2GA_3@50ppm	15.73	16.36	16.74	17.20	17.57	18.55	18.89	19.38	19.95
T_3JLE@2%	15.34	15.73	16.20	16.53	16.97	17.48	18.07	18.88	19.20
T_4KNO_3@0.5%	15.65	16.28	16.46	17.21	17.90	18.31	18.94	19.23	19.50
T_5KCl@0.2%	15.65	16.15	16.45	17.10	17.95	18.30	18.83	19.20	19.52
T_6NaCl@0.5%	15.47	16.07	16.47	17.1	17.79	18.51	18.92	19.25	19.46
T_7IAA@0.2%	15.74	16.31	16.58	17.11	17.80	18.31	18.95	19.21	19.55
T_8CaCl_2@0.1%	15.82	16.48	16.70	17.08	17.63	18.18	18.36	19.18	19.35
T_9Control	15.48	15.69	15.95	16.55	17.00	17.50	18.29	18.63	19.16
SEm±	0.07	0.10	0.21	0.06	0.13	0.12	0.10	0.07	0.10
CD or LSD	0.21	0.30	0.63	0.18	0.40	0.36	0.30	0.21	0.29

Table 4.2.2 (b): Effect of seed priming on reducing sugar content (mg g-1 dry wt.) Under anaerobic condition on rice seedling

Samba Mahsuri									
Treatments\Days	**2**	**4**	**6**	**8**	**10**	**12**	**14**	**16**	**18**
T_1GA_3@25ppm	15.01	15.51	15.83	16.26	16.56	16.90	17.53	17.75	18.01
T_2GA_3@50ppm	15.28	15.55	15.71	16.45	16.67	17.04	17.42	18.02	18.33
T_3JLE@2%	14.25	14.33	14.47	15.18	15.48	15.68	16.38	16.89	16.99
T_4KNO_3@0.5%	15.00	15.62	15.96	16.19	16.56	17.00	17.37	17.42	17.73
T_5KCl@0.2%	14.60	15.00	15.23	15.76	15.97	16.31	16.82	17.11	17.67
T_6NaCl@0.5%	15.48	15.63	15.83	16.29	16.62	16.76	16.91	17.23	17.55
T_7IAA@0.2%	14.71	15.3	15.79	15.94	16.34	16.78	17.01	17.52	17.76
T_8CaCl_2@0.1%	14.39	14.82	15.66	16.16	16.44	16.71	17.00	17.02	17.30
T_9Control	14.16	14.32	14.68	15.05	15.62	15.96	16.33	16.74	16.98
SEm±	0.05	0.06	0.12	0.09	0.08	0.14	0.06	0.10	0.05
CD or LSD	0.15	0.18	0.37	0.26	0.23	0.42	0.19	0.30	0.14

Table 4.2.3 (a): Effect of seed priming on non- reducing sugar content (mg g-1 dry wt.) Under anaerobic condition on rice seedling

Samba Mahsuri Sub-1									
Treatments\Days	**2**	**4**	**6**	**8**	**10**	**12**	**14**	**16**	**18**
T_1GA_3@25ppm	27.36	27.93	28.19	28.23	28.50	28.55	28.62	28.70	29.01
T_2GA_3@50ppm	27.54	27.62	27.75	28.07	28.14	28.24	28.35	28.45	29.90
T_3JLE@2%	26.10	26.35	26.42	26.50	26.97	27.10	27.25	27.38	27.61
T_4KNO_3@0.5%	27.01	27.14	27.21	27.23	27.33	27.40	27.47	27.65	27.99
T_5KCl@0.2%	27.00	27.03	27.16	27.30	27.46	27.64	27.72	27.82	28.27
T_6NaCl@0.5%	26.71	26.94	26.97	26.99	27.19	27.32	27.67	27.90	28.10
T_7IAA@0.2%	26.15	26.27	26.40	26.49	26.91	27.10	27.22	27.40	27.67
T_8CaCl_2@0.1%	26.66	26.74	26.95	27.11	27.20	27.37	27.42	27.51	27.76
T_9Control	27.08	27.13	27.43	27.51	27.56	27.61	27.70	27.77	27.57
SEm±	0.14	0.18	0.34	0.08	0.14	0.20	0.19	0.11	0.17
CD or LSD	0.41	0.54	1.01	0.23	0.43	0.59	0.57	0.33	0.51

Table 4.2.3 (b): Effect of seed priming on non- reducing sugar content (mg g-1 dry wt.) Under anaerobic condition on rice seedling

Samba Mahsuri									
Treatments\Days	**2**	**4**	**6**	**8**	**10**	**12**	**14**	**16**	**18**
T_1GA_3@25ppm	26.30	27.90	28.10	28.20	28.38	28.51	28.67	28.70	28.70
T_2GA_3@50ppm	26.41	27.92	28.15	28.25	28.42	28.60	28.72	28.75	29.10
T_3JLE@2%	25.75	26.05	26.35	26.50	26.67	26.97	27.15	27.38	27.57
T_4KNO_3@0.5%	26.95	27.08	27.10	27.21	27.23	27.31	27.40	27.47	27.65
T_5KCL@0.2%	26.66	27.00	27.03	27.16	27.28	27.45	27.12	27.72	27.82
T_6NaCL@0.5%	26.70	26.85	26.94	26.96	26.99	27.10	27.30	27.67	27.75
T_7IAA@0.2%	24.90	26.15	26.26	26.35	26.40	26.81	27.05	27.20	27.40
T_8CaCL_2@0.1%	26.15	26.65	26.71	26.95	27.11	27.22	27.37	27.40	27.51
T_9Control	25.08	26.13	26.43	26.51	26.56	26.61	26.70	26.77	26.86
SEm±	0.32	0.21	0.42	0.43	0.32	0.51	0.28	0.44	0.51
CD or LSD	0.95	0.64	1.26	1.29	0.97	1.53	0.83	1.32	1.54

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