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Influence of Seed Priming Treatments on Biochemical Parameters of Dry Direct Sown Rice (*Oryza sativa* L.)

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A B S T R A C T

The present investigation was carried out at S.V Agricultural College, Tirupati to know the effect of seed priming on biochemical parameters of dry direct sown rice (*Oryza Sativa L.*). A laboratory experiment was conducted in completely randomized design and replicated thrice with six popular aerobic rice genotypes i.e. MTU 1010, MTU 7029, JGL 20171, NLR33671, MTU 1075 and MTU 1112. Rice seeds were subjected to different concentrations of gibberillic acid with, 200, 500 and 1000 PPM and combination of both gibberillic acid and KNO_3 treatments i.e. GA_3 (200PPM, 500PPM, 1000PPM + KNO_3 @3%), KNO_3 @3% and control (Hydropriming/Water soaking), in order to know the effect of seed priming (Gibberllic acid and KNO_3) on various biochemical parameters like Reducing sugars (mg/g), Starch (mg/g) and Alpha amylase (%) activity were recorded at 2, 4 and 6 DAT. The results revealed that in all the biochemical parameters among Varieties MTU 1010, JGL 20171, NLR 33671 had recorded higher amount of reducing sugars, starch and Alpha amylase activity compared to MTU 7029, MTU 1075 and MTU 1112. Among treatments 1000 PPM GA_3, GA_3 (200 PPM, 500PPM, 1000PPM+KNO_3@3%) are found to be best.

Introduction

Rice (*Oryza sativa* L.) is the most important cereal food crop of the developing world and the staple food of more than half of the world's population. Globally rice is grown over an area of 161.83 million ha with an annual production of 717.8 million tonnes (IRRI, 2017). Irrigated rice is the major consumer of fresh water. It was estimated that by 2025, about 15-20 million hectares of irrigated rice will be affected due to water scarcity which threatens the productivity. Combining the growing demand for food with increasing water scarcity, rice producers in Asia need to produce more rice with less water.

A major challenge in rice production is therefore to save water while maintaining or even increasing the grain yield (Yang and Zhang, 2010). Many water-saving technologies are currently used in rice production, including alternate wetting and drying irrigation, the rice intensification system, aerobic rice and the ground cover rice production systems (GCRPSs) (Qin *et al*., 2006). Among these aerobic rice is gaining

popularity as a strategy for water saving agriculture.

Aerobic rice can achieve yields of 4-6 tons per hectare with 50 - 70% less water compared to lowland rice it does not require flooded wetland (Qin *et al*., 2010). In aerobic rice production, the seeds are direct-seeded in aerobic soil without any standing water layer, which minimizes water use and boosts up water productivity by eliminating continuous seepage and percolation, reducing evaporation and eliminating wetland preparation (Nie *et al*., 2012; Singh *et al*., 2008). Season-long weed infestation in aerobic rice may cause yield reduction up to 80 % or complete failure of crop in extreme cases (Jayadeva *et al*., 2011; Sunil *et al*., 2010).

Therefore, the aerobic rice cultivars should have the capacity of early seedling establishment, quick crop growth and yield stability. Identification of strong weed competitive rice cultivar is a feasible solution to inhibit the growth of weeds and it is a cost-effective and safe tool for weed management (Zhao *et al*., 2006).

Seedling vigour is a physiological trait and a sign of potential seed germination, seedling growth and tolerance to adverse climatic factors. On the other hand, it significantly improves the speed, uniformity and the final percentage of germination, and leads to ideal field appearance with good potential grain yield under direct-seeded conditions (Foolad *et al*., 2007).

Thus, to suppress weed growth, early seedling vigour of an elite variety should be achieved. Seed priming is a viable option to attain this target. Seed priming, which is also called seed hardening, is a physiological seed enhancement method. It is a presowing treatment in which seeds are soaked in an osmotic solution that allows them to imbibe water and go through the first stages of germination, but does not permit radicle protrusion through the seed coat. Subsequently, the seeds can be dried to attain their original moisture content and stored or planted using conventional techniques.

Thus in this paper the influence of seed priming with GA_3 and KNO_3 either alone or in combination was tested to know the amount of reducing sugars, starch and amylase content in rice seeds.

Materials and Methods

0.1, 0.25 and 0.5 g each of GA_3 (gibberellic acid, HIMEDIA) was dissolved separately 500 ml each in DDW along with 1.5 g KNO_3 in each case to prepare a series of solutions that gives GA_3 @ 200 ppm + KNO_3 @ 3 %, GA3 @ 500 ppm + KNO_3 @ 3 %, GA_3 @ 1000 ppm + KNO_3 @ 3 %. 3 % KNO_3 was prepared by dissolving 1.5 g of KNO_3 in 500 mL of DDW. Seeds were soaked in the respective treatmental solutions for 24 h and re dried overnight (about 12 h) and placed on petriplates. Seeds are moistened periodically with double distilled water.

Reducing sugars, Starch, alpha amylase were estimated at 48h (2 days), 96h (4 days) and 144h (6 days) after soaking the seeds in different concentrations of GA_3 and KNO_3 treatments. By following standard protocols i.e; Nelson method for reducing sugars, Anthrone reagent method for estimation of starch and DNSA (Dinitro-salycilic acid) method for estimation of α- amylase content in rice seeds were recorded.

Results and Discussion

Reducing sugars (mg g^{-1})

Maintenance of a higher amount of reducing sugar content is an important prerequisite for

faster germination via enhanced metabolic activity of the embryo. Reducing sugar content was measured at 2, 4 and 6 DAT. A significant difference among varieties, treatments and their interaction was observed at all the stages and analyzed statistically and presented in table 1, 2 and 3 and Figure 1, 2 and 3. At 2 DAT it was observed that JGL 20171 (2.54) recorded significantly high reducing sugars followed by NLR 33671 (1.33). Among treatments T_5 (GA_3 @ 200 ppm + KNO_3 @ 3%) (1.93) recorded significantly high reducing sugars followed by T_6 (GA_3 @ 500 ppm + KNO_3 @ 3%) (1.79). T_1 (control) (1.11) recorded lowest reducing sugars not only at this stage but also at 4 and 6 DAT (1.75 and 2.02).

At 4 and 6 DAT the genotypes MTU 1010 (2.86 and 3.28) and JGL 20171 (2.77 and 3.04) recorded significantly highest reducing sugars whereas the lowest value was recorded in MTU 1112 (1.9 and 2.14). Among treatments T_5 (GA_3 @ 200 ppm + KNO_3 @ 3%) (3.07 and 3.19) and T_6 (GA_3 @ 500 ppm + KNO_3 @ 3%) (2.96 and 3.35) showed better results. The interaction effect was found to be significant only at 2 DAT where in V_3T_5 (3.20) recorded highest value.

A progressive increase in reducing sugars content was observed from 2 DAT to 6 DAT. MTU 1010, for example recorded the reducing sugar content of 1.30, 2.86 and 3.28 at 2, 4 and 6 DAT respectively. The corresponding values for JGL 20171 were 2.54, 2.77 and 3.04.

This increased availability of reducing sugars could be linked up with the activity of α-amylase on breakdown of starch. Similar increase in reducing sugar content due to increased starch metabolism was also observed by Brain, 1959; Kuo and Yang, 1967; Basra *et al*., 2005 and Acharya *et al*., 2008.

Starch (mg g^{-1})

Starch represents the resource base of a seed. Higher starch content and its faster breakdown into sugars accelerate the metabolic processes of the embryo which leads to rapid cell division and cell expansion.

Data on influence of different seed priming treatments on starch content at 2, 4 and 6 DAT were presented in tables 4, 5 and 6 and Figure 4, 5 and 6. From the data a gradual decrease in starch content was observed from 2 DAT to 6 DAT. Starch content in the genotype MTU 1010 for example at 2, 4 and 6 DAT was 41.2, 38.3, and 33.1 respectively. The corresponding values in MTU 1112 were 49.2, 46.2, and 42.1 respectively.

Irrespective of the genotype starch content decreased with time. Among the varieties MTU 1075 and MTU 1112 (50.1 and 49.2) recorded significantly higher and at par values of starch followed by MTU 7029 (43.5), MTU 1010 (41.2), NLR 3367 (41.0) and JGL 2017 (40.9) which were at par. MTU 1075 and MTU 1112 recorded highest starch content even at 4 and 6 DAT. This might be due to slow breakdown of starch in these genotypes.

Among treatments T_8 (KNO_3 @ 3%) (50.1), T_1 (Control) (49.8), T_2 (GA_3 @ 200 ppm) (48.3) and T_3 (GA_3 @ 500 ppm) (46.9) recorded significantly higher and at par starch values where T_5 (GA_3 @ 200 ppm + KNO_3 @ 3%) (38.8), T_6 (GA_3 @ 500 ppm+KNO_3@3%) (39.4), T_7(GA_3@ 1000 ppm + KNO_3 @ 3%) (40) and T_4 (GA_3 @ 1000 ppm) (41.4) recorded significantly low and at par starch content at 2 DAT. This could be due to the influence of these treatments on faster breakdown of starch.

Almost a similar trend was observed at 4 and 6 DAT. The interaction effect was found to be non significant at all the stages.

Table.1 Influence of different seed priming treatments and varieties on reducing sugars (mg g^{-1}) at 2 days after treatment

Variety	T_1	T_2	T_3	T_4	T_5	T_6	T_7	T_8	Mean
MTU 1010	0.98	1.08	1.12	1.40	1.84	1.65	1.30	1.00	**1.30**
MTU 7029	1.00	1.10	1.20	1.34	1.83	1.56	1.48	1.10	**1.33**
JGL 20171	1.89	2.20	2.60	2.98	3.20	3.12	2.30	2.00	**2.54**
NLR 33671	0.89	1.10	1.30	1.40	1.67	1.60	1.50	1.20	**1.33**
MTU 1075	0.87	1.20	1.17	1.40	1.56	1.51	1.34	1.26	**1.29**
MTU 1112	1.00	1.20	1.20	1.23	1.45	1.30	1.32	1.30	**1.25**
Mean	**1.11**	**1.32**	**1.43**	**1.62**	**1.93**	**1.79**	**1.54**	**1.31**	
				V	**T**	**V × T**			
			C.D.	**0.10**	**0.12**	**0.29**			
			SE(d)	0.05	0.06	0.14			
			SEm±	0.04	0.04	0.10			

T_1 : Control	**T_5 : GA_3 @ 200 ppm + KNO_3 @ 3%**
T_2 : GA_3 @ 200 ppm	T_6 : GA_3 @ 500 ppm+ KNO_3 @ 3%
T_3 : GA_3 @ 500 ppm	T_7 : GA_3 @1000 ppm+ KNO_3 @ 3 %
T_4 : GA_3 @ 1000 ppm	T_8 : KNO_3 @ 3%

Table.2 Influence of different seed priming treatments and varieties on reducing sugars (mg g^{-1}) at 4 days after treatment

Variety	T_1	T_2	T_3	T_4	T_5	T_6	T_7	T_8	Mean
MTU 1010	2.34	2.56	2.60	3.15	3.40	3.23	3.10	2.50	**2.86**
MTU 7029	1.54	2.00	2.45	2.60	3.02	2.89	2.20	1.76	**2.31**
JGL 20171	2.19	2.42	2.59	3.13	3.26	3.18	3.04	2.35	**2.77**
NLR 33671	1.89	2.20	2.60	2.98	3.20	3.12	2.30	2.00	**2.54**
MTU 1075	1.34	1.80	2.30	2.54	3.00	2.80	2.10	1.70	**2.20**
MTU 1112	1.21	1.56	2.00	2.10	2.56	2.30	2.00	1.45	**1.90**
Mean	**1.75**	**2.09**	**2.42**	**2.75**	**3.07**	**2.92**	**2.46**	**1.96**	
				V	**T**	**V × T**			
			C.D.	**0.16**	**0.19**	**NS**			
			SE(d)	0.08	0.09	0.23			
			SEm±	0.06	0.07	0.16			

Table.3 Influence of different seed priming treatments and varieties on reducing sugars (mg g^{-1}) at 6 days after treatment

Variety	T_1	T_2	T_3	T_4	T_5	T_6	T_7	T_8	Mean
MTU 1010	2.67	3.00	3.30	3.56	3.56	3.78	3.40	2.98	**3.28**
MTU 7029	2.00	2.10	2.56	2.98	3.10	3.26	2.45	1.98	**2.55**
JGL 20171	2.40	2.64	2.98	3.30	3.40	3.54	3.25	2.80	**3.04**
NLR 33671	2.00	2.54	2.98	3.21	3.30	3.43	2.89	2.80	**2.89**
MTU 1075	1.56	1.90	2.40	2.78	3.00	3.10	2.45	1.90	**2.39**
MTU 1112	1.43	1.76	2.10	2.30	2.76	2.99	2.09	1.65	**2.14**
Mean	2.01	2.32	2.72	3.02	3.19	3.35	2.76	2.35	
				V	**T**	**V × T**			
			C.D.	**0.18**	**0.21**	**NS**			
			SE(d)	0.09	0.10	0.25			
			SEm±	0.06	0.07	0.18			

T_1 : Control	**T_5 : GA_3 @ 200 ppm + KNO_3 @ 3%**
T_2 : GA_3 @ 200 ppm	T_6 : GA_3 @ 500 ppm+ KNO_3 @ 3%
T_3 : GA_3 @ 500 ppm	T_7 : GA_3 @1000 ppm+ KNO_3 @ 3 %
T_4 : GA_3 @ 1000 ppm	T_8 : KNO_3 @ 3%

Table.4 Influence of different seed priming treatments and varieties on starch (mg g-1) at 2 days after treatment

Variety	T_1	T_2	T_3	T_4	T_5	T_6	T_7	T_8	Mean
MTU 1010	46.2	43.1	41.2	40.8	33.6	38.6	37.5	48.5	**41.2**
MTU 7029	51.0	53.0	46.0	39.0	38.5	36.0	33.5	51.0	**43.5**
JGL 20171	43.9	43.8	42.5	37.5	38.5	39.7	36.5	44.8	**40.9**
NLR 33671	47.1	45.2	42.5	39.8	33.6	34.5	39.4	46.2	**41.0**
MTU 1075	56.5	54.8	56.1	44.5	43.9	44.1	45.3	55.5	**50.1**
MTU 1112	53.8	49.8	52.9	46.9	44.5	43.5	47.8	54.6	**49.2**
Mean	49.8	48.3	46.9	41.4	38.8	39.4	40.0	50.1	
				V	**T**	**V × T**			
			C.D.	**2.90**	**3.34**	**NS**			
			SE(d)	1.46	1.68	4.12			
			SEm±	1.03	1.19	2.91			

Table.5 Influence of different seed priming treatments and varieties on starch (mg g^{-1}) at 4 days after treatment

Variety	T_1	T_2	T_3	T_4	T_5	T_6	T_7	T_8	Mean
MTU 1010	44.6	45.7	42.5	31.8	31.6	34.5	32.5	43.5	**38.3**
MTU 7029	51.2	42.5	45.5	33.8	35.6	36.8	40.1	45.5	**41.4**
JGL 20171	43.5	41.7	42.8	30.6	34.5	36.2	33.5	48.2	**38.9**
NLR 33671	45.6	45.8	43.5	34.8	36.9	32.8	37.4	44.8	**40.2**
MTU 1075	44.6	48.5	43.2	36.5	37.8	36.5	38.6	47.2	**41.6**
MTU 1112	56.8	55.6	53.2	38.2	36.3	37.1	36.5	55.7	**46.2**
Mean	**47.7**	**46.6**	**45.1**	**34.3**	**35.5**	**35.7**	**36.4**	**47.5**	
				V	**T**	**V × T**			
			C.D.	**2.70**	**3.12**	**NS**			
			SE(d)	1.36	1.57	3.84			
			SEm±	0.96	1.11	2.72			

T_1 : Control	**T_5 : GA_3 @ 200 ppm + KNO_3 @ 3%**
T_2 : GA_3 @ 200 ppm	T_6 : GA_3 @ 500 ppm+ KNO_3 @ 3%
T_3 : GA_3 @ 500 ppm	T_7 : GA_3 @1000 ppm+ KNO_3 @ 3 %
T_4 : GA_3 @ 1000 ppm	T_8 : KNO_3 @ 3%

Table.6 Influence of different seed priming treatments and varieties on Starch (mg g^{-1}) at 6 days after treatment

Variety	T_1	T_2	T_3	T_4	T_5	T_6	T_7	T_8	Mean
MTU 1010	38.3	37.4	35.5	31.6	29.4	28.6	27.8	36.5	**33.1**
MTU 7029	40.5	41.2	38.9	29.8	30.5	27.8	31.5	35.7	**34.5**
JGL 20171	38.7	35.8	36.3	26.2	24.8	24.5	25.8	39.5	**31.5**
NLR 33671	44.6	40.8	43.9	26.4	27.3	29.8	28.7	42.7	**35.5**
MTU 1075	51.3	45.9	47.8	34.2	30.5	35.2	33.8	46.7	**40.7**
MTU 1112	52.4	50.6	42.3	36.3	38.4	34.8	36.8	45.1	**42.1**
Mean	**44.3**	**42.0**	**40.8**	**30.8**	**30.2**	**30.1**	**30.7**	**41.0**	
				V	**T**	**V × T**			
			C.D.	**2.42**	**2.79**	**NS**			
			SE(d)	1.22	1.41	3.44			
			SEm±	0.86	0.99	2.43			

Table.7 Influence of different seed priming treatments and varieties on alpha amylase (mg g^{-1}) at 2 days after treatment

Variety	T_1	T_2	T_3	T_4	T_5	T_6	T_7	T_8	Mean
MTU 1010	1.9	1.9	1.8	2.4	2.2	2.3	2.3	1.9	**2.1**
MTU 7029	1.7	2.0	1.7	2.2	2.1	2.2	2.2	1.9	**2.0**
JGL 20171	2.0	2.1	2.0	2.4	2.3	2.4	2.4	2.0	**2.2**
NLR 33671	1.7	1.5	1.5	2.2	2.3	2.3	2.1	1.7	**1.9**
MTU 1075	0.9	0.9	0.8	1.3	1.4	1.3	1.3	0.9	**1.1**
MTU 1112	0.7	0.8	0.7	1.3	1.3	1.2	1.1	0.8	**1.0**
Mean	**1.5**	**1.5**	**1.4**	**1.9**	**1.9**	**2.0**	**1.9**	**1.5**	
				V	**T**	**V × T**			
			C.D.	**0.11**	**0.13**	**NS**			
			SE(d)	0.06	0.07	0.16			
			SEm±	0.04	0.05	0.11			

T_1 : Control	**T_5 : GA_3 @ 200 ppm + KNO_3 @ 3%**
T_2 : GA_3 @ 200 ppm	T_6 : GA_3 @ 500 ppm+ KNO_3 @ 3%
T_3 : GA_3 @ 500 ppm	T_7 : GA_3 @1000 ppm+ KNO_3 @ 3 %
T_4 : GA_3 @ 1000 ppm	T_8 : KNO_3 @ 3%

Table.8 Influence of different seed priming treatments and varieties on alpha amylase (mg g^{-1}) at 4 days after treatment

Variety	T_1	T_2	T_3	T_4	T_5	T_6	T_7	T_8	Mean
MTU 1010	2.1	2.2	2.4	2.7	2.7	2.8	2.8	2.2	**2.5**
MTU 7029	2.0	2.0	2.5	2.3	2.5	2.6	2.6	2.1	**2.3**
JGL 20171	2.5	2.4	2.4	2.6	2.8	2.8	2.9	2.3	**2.6**
NLR 33671	1.8	1.6	2.0	2.8	2.6	2.3	2.5	1.9	**2.2**
MTU 1075	1.0	0.9	1.0	1.4	1.4	1.9	1.8	1.2	**1.3**
MTU 1112	0.8	0.8	1.0	1.4	1.5	1.6	1.3	0.8	**1.1**
Mean	**1.7**	**1.6**	**1.9**	**2.2**	**2.2**	**2.3**	**2.3**	**1.7**	
				V	**T**	**V × T**			
			C.D.	**0.13**	**0.16**	**NS**			
			SE(d)	0.07	0.08	0.19			
			SEm±	0.05	0.06	0.14			

Table.9 Influence of different seed priming treatments and varieties on Alpha amylase (mg g^{-1}) at 6 days after treatment

Variety	T_1	T_2	T_3	T_4	T_5	T_6	T_7	T_8	Mean
MTU 1010	1.6	1.5	1.6	2.1	2.2	2.5	2.7	1.2	**1.9**
MTU 7029	1.4	1.7	1.4	2.0	1.8	2.0	2.1	1.3	**1.7**
JGL 20171	1.6	1.8	1.9	2.0	2.0	2.1	1.9	1.2	**1.8**
NLR 33671	1.2	1.6	0.9	1.8	2.1	2.3	1.8	0.8	**1.6**
MTU 1075	0.9	0.8	0.7	1.2	1.6	1.6	1.7	1.0	**1.2**
MTU 1112	0.7	0.9	1.2	1.2	1.4	1.3	1.4	0.7	**1.1**
Mean	**1.2**	**1.3**	**1.3**	**1.7**	**1.9**	**2.0**	**1.9**	**1.0**	
				V	**T**	**V × T**			
			C.D.	**0.10**	**0.12**	**0.29**			
			SE(d)	0.05	0.06	0.15			
			SEm±	0.04	0.04	0.10			

T_1 : Control	**T_5 : GA_3 @ 200 ppm + KNO_3 @ 3%**
T_2 : GA_3 @ 200 ppm	T_6 : GA_3 @ 500 ppm+ KNO_3 @ 3%
T_3 : GA_3 @ 500 ppm	T_7 : GA_3 @1000 ppm+ KNO_3 @ 3 %
T_4 : GA_3 @ 1000 ppm	T_8 : KNO_3 @ 3%

Fig.1 Influence of different seed priming treatments and varieties on reducing sugars (mg g^{-1}) at 2 days after treatment

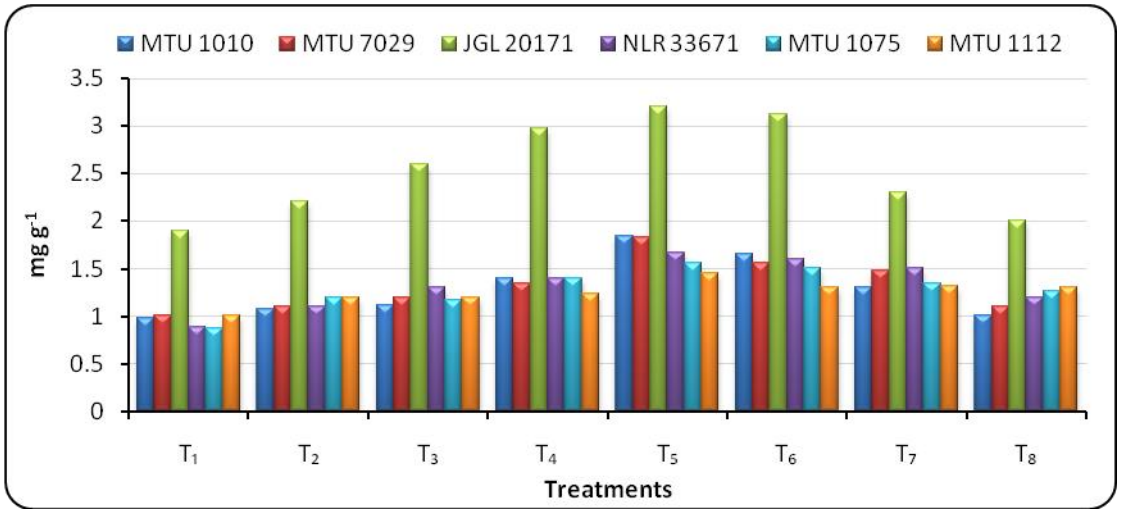


Fig.2 Influence of different seed priming treatments and varieties on reducing sugars (mg g^{-1}) at 4 days after treatment

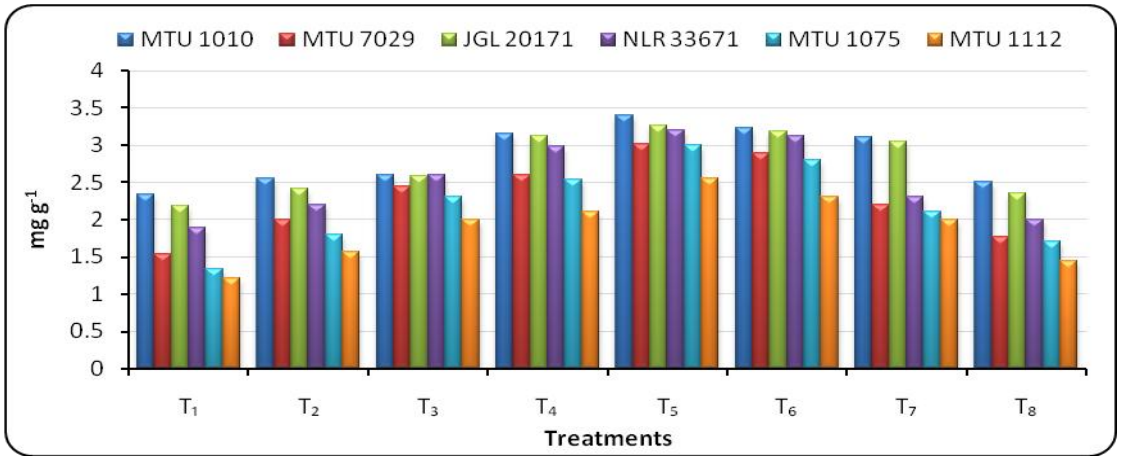


Fig.3 Influence of different seed priming treatments and varieties on reducing sugars (mg g-1) at 6 days after treatment

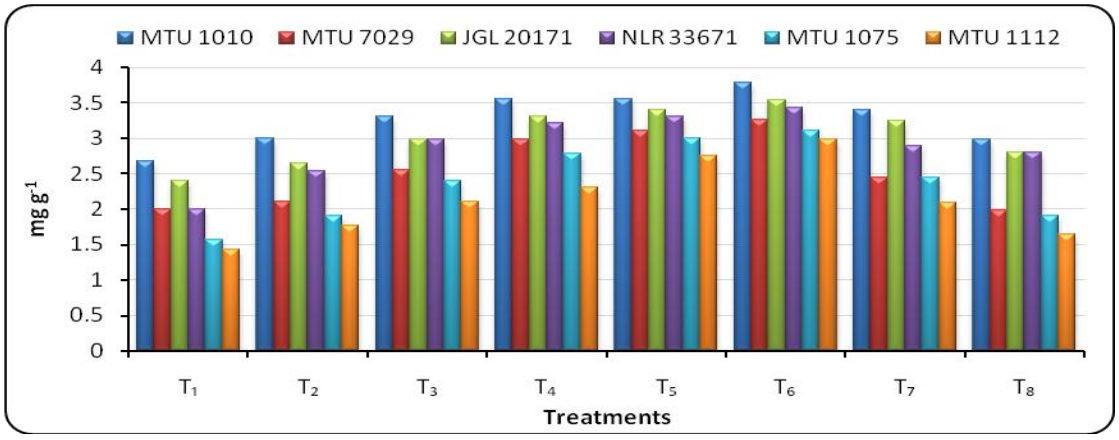


Fig.4 Influence of different seed priming treatments and varieties on starch (mg g^{-1}) index at 2 days after treatment

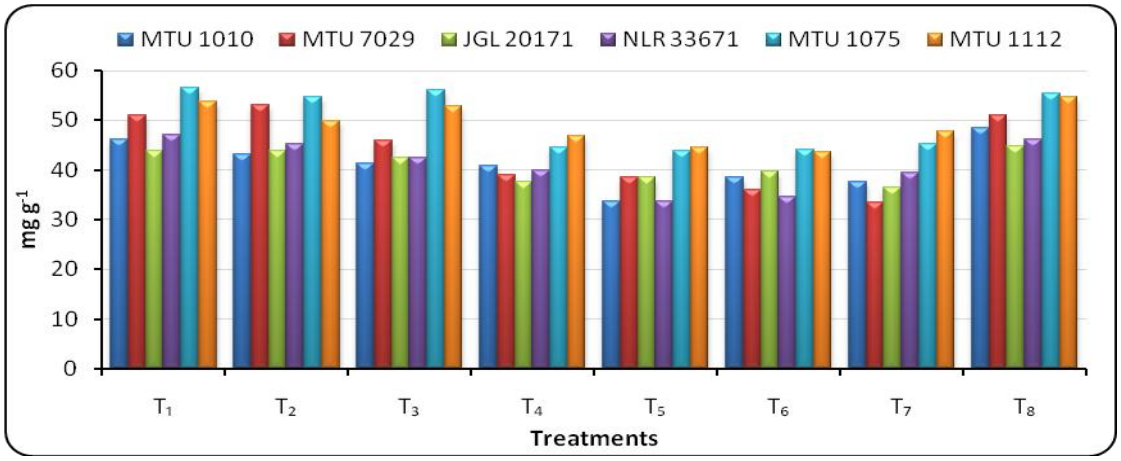


Fig.5 Influence of different seed priming treatments and varieties on starch (mg g^{-1}) at 4 days after treatment

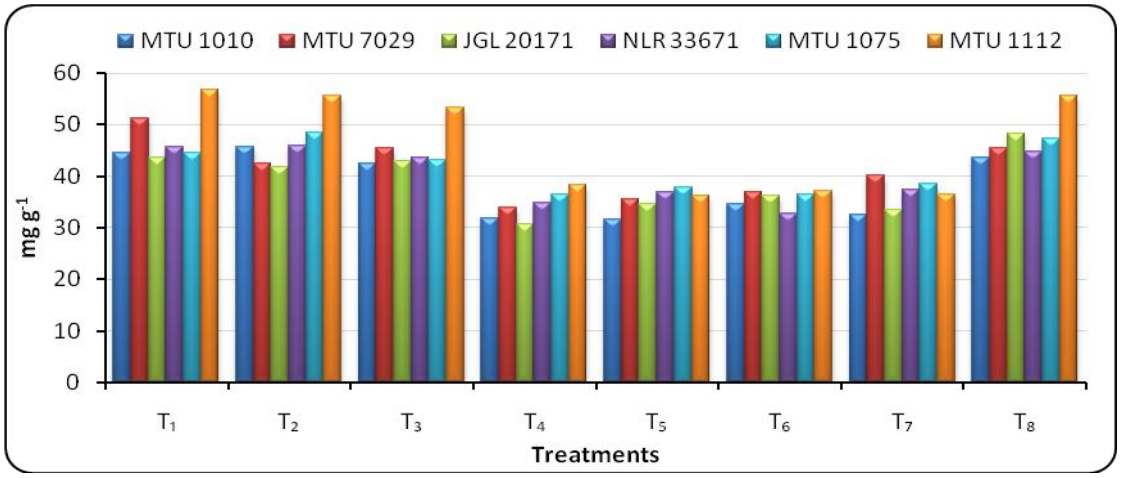


Fig.6 Influence of different seed priming treatments and varieties on starch (mg g-1) at 6 days after treatment

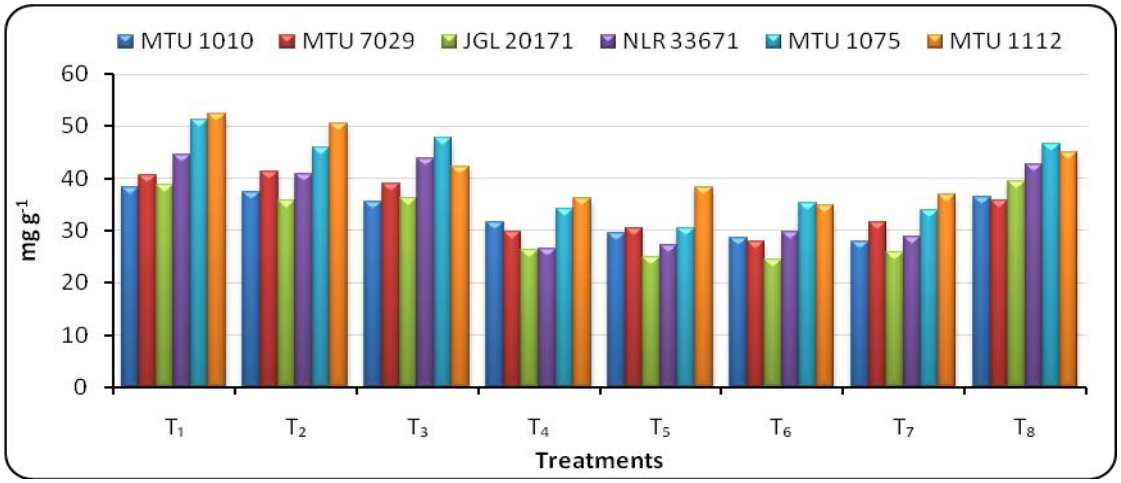


Fig.7 Influence of different seed priming treatments and varieties on alpha amylase (mg g^{-1}) at 2 days after treatment

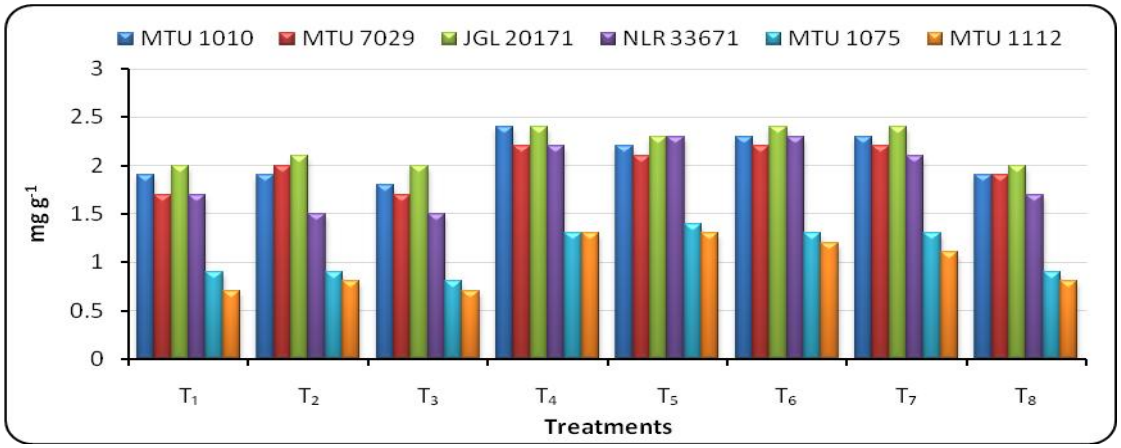


Fig.8 Influence of different seed priming treatments and varieties on alpha amylase (mg g^{-1}) at 4 days after treatment

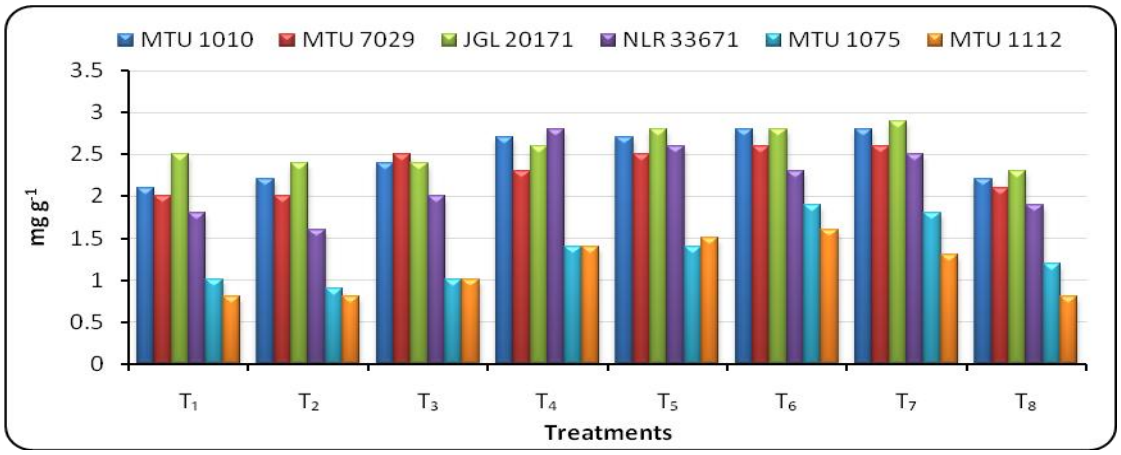
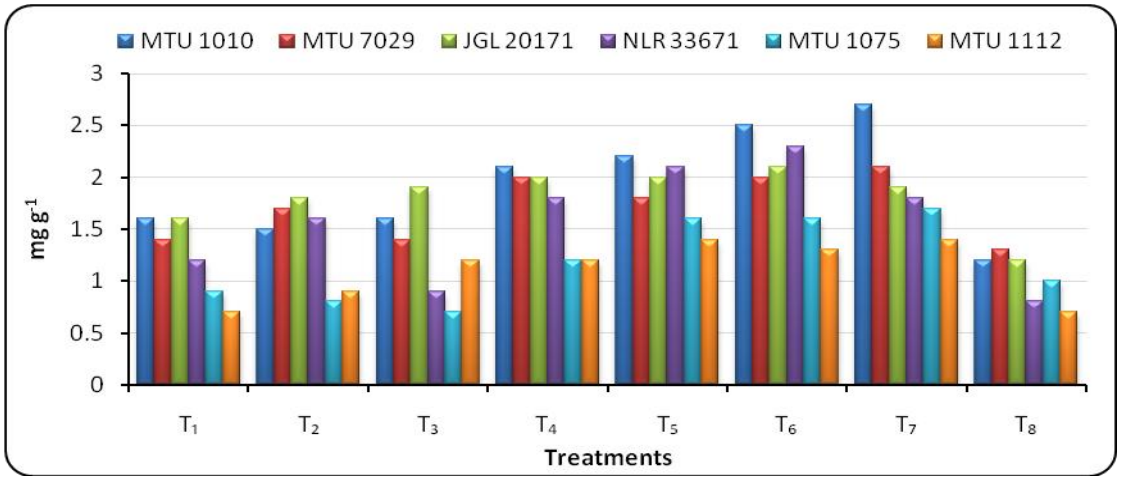


Fig.9 Influence of different seed priming treatments and varieties on alpha amylase (mg g^{-1}) at 6 days after treatment



Robolledo *et al.*, (2012) also reported that stored starch is an important factor for growth and maintenance to balance vigour and moisture stress tolerance in aerobic condition of rice. Further, Luquet *et al.*, (2012) suggested that besides morphological traits, non structural carbohydrates particularly starch could be used as relevant marker to screen genotypes for early seedling vigour in aerobic rice.

Alpha - amylase (mg g^{-1})

De-nova synthesis of α-amylase in aleurone layer is believed to be essential for seed germination in cereal grains, which is tightly regulated by GA_3 synthesized in the embryonic layer.

Data on influence of different seed priming treatments on different aerobic rice genotypes with respect to α- amylase content at 2, 4 and 6 DAT was presented in table 7, 8 and 9 and Figure 7, 8 and 9 respectively.

The results revealed a gradual increase in α-amylase content from 2 DAT to 4 DAT. Later it was decreased at 6 DAT. Among varieties JGL 20171 (2.2, 2.6 and 1.8) and MTU 1010 (2.1,2.5 and 1.9) were found to consist

significantly highest and at par α-amylase content whereas MTU 1112 (1.0) and MTU 1075 (1.1) recorded significantly lowest and at par values at 2, 4 and 6 DAT. It indicates the genotypic difference in α-amylase content.

This higher α-amylase content might be the reason for higher seedling vigour index in these genotypes. This genotypic character was seem to be enhanced further with different treatments.

Among treatments T_6 (GA_3 @ 500 ppm + KNO_3 @ 3%) (2.0) recorded significantly highest α-amylase content which was at par with T_5 (GA_3 @ 200 ppm + KNO_3 @ 3%) (1.9), T_7 (GA_3 @ 1000 ppm + KNO_3 @ 3%)(1.9) and T_4 (GA_3 @ 1000 ppm) (1.9).

T_5 (1.9, 2.2 and 1.9) and T_6 (2.0, 2.3 and 2.0) recorded highest α-amylase content at all the stages whereas T_8 (1.0) followed by T_1 (1.2) recorded significantly lowest values at 6 DAT. This could explain the role of GA_3 either alone or in combination in enhancing α-amylase content.

The interaction effect was found to be non significant at 2 and 4 DAT. However, a significant interaction was observed at 6 DAT. Where in V_1T_7 (2.7) recorded significantly highest α-amylase content and the lowest was recorded in V_6T_1 (0.7).

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