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Biochemical responses associated with drought tolerance in rice genotypes under rainfed and irrigated conditions

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Abstract

The present study was conducted to evaluate the biochemical responses of rice genotypes under irrigated and rainfed conditions. Thirty rice genotypes along with the check variety Poornima (LC) were evaluated during the Kharif season under contrasting moisture regimes. Biochemical parameters including chlorophyll content, proline content, ascorbate peroxidase (APX), superoxide dismutase (SOD), peroxidase (POD) and catalase (CAT) activities were assessed to understand the response of genotypes to water stress. Rainfed conditions generally resulted in a reduction in chlorophyll content, whereas proline accumulation and antioxidant enzyme activities increased under water stress. The magnitude of response varied among genotypes, indicating differences in their stress-adaptation ability. Poornima (LC) and RFU 329 showed comparatively better biochemical responses under rainfed conditions, particularly through higher proline accumulation and antioxidant activity. The results suggest that biochemical traits can serve as useful indicators for identifying rice genotypes with better adaptation to moisture stress. Further evaluation across seasons and locations is required to confirm their drought tolerance potential.

Keywords: Rice, drought stress, rainfed condition, irrigated condition, biochemical traits, chlorophyll, proline, antioxidant enzymes, drought tolerance

1. Introduction

Drought is one of the major abiotic stresses limiting rice productivity, particularly in rainfed production systems. Water deficit affects several physiological processes, including photosynthesis, cellular water balance and metabolic activity, ultimately reducing plant growth and yield. The ability of rice genotypes to maintain physiological functions under water stress is therefore an important component of drought tolerance.

Biochemical responses play an important role in the adaptation of rice plants to drought. Chlorophyll content is closely associated with photosynthetic capacity, whereas proline accumulation contributes to osmotic adjustment and protection of cellular structures under water deficit. Drought stress also induces the formation of reactive oxygen species (ROS), which can cause oxidative damage when their production exceeds the plant's protective capacity. Plants counteract this oxidative stress through antioxidant enzymes such as superoxide dismutase (SOD), ascorbate peroxidase (APX), peroxidase (POD) and catalase (CAT).

Considerable genotypic variation exists in the biochemical response of rice to water stress. Genotypes capable of maintaining higher chlorophyll content and accumulating compatible solutes while enhancing antioxidant enzyme activity may possess better mechanisms for coping with drought. In the present study, these biochemical traits were evaluated in rice genotypes under rainfed and irrigated conditions to identify responses associated with better drought adaptation.

Therefore, the present study was undertaken to evaluate chlorophyll content, proline accumulation and antioxidant enzyme activities (APX, SOD, POD and CAT) in rice genotypes under rainfed and irrigated conditions and to identify genotypes showing superior biochemical responses to water stress.

2. Materials and Methods

The present study was conducted at the Research Cum Instructional Farm, Indira Gandhi Krishi Vishwavidyalaya, Raipur, Chhattisgarh, during the Kharif season of 2019-20.

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Thirty rice (*Oryza sativa* L.) genotypes obtained from IIRR, Hyderabad, under the AICRP on Rice programme, along with the check variety Poornima (LC), were evaluated under rainfed and irrigated conditions. The experiment was laid out in a randomized block design with three replications. The plot size was 3.80×1.40 m, with a spacing of 20×20 cm. A fertilizer dose of $100:45:60$ kg N:P₂O₅:K₂O ha⁻¹ was applied.

Under rainfed conditions, no artificial irrigation was provided after crop establishment, whereas irrigation was supplied according to crop requirement under irrigated conditions. Biochemical observations were recorded for chlorophyll content, proline content, ascorbate peroxidase (APX), superoxide dismutase (SOD), peroxidase (POD) and catalase (CAT) activities. Chlorophyll content was measured using a SPAD meter, while proline and antioxidant enzyme activities were estimated using the respective biochemical procedures followed in the original study.

The biochemical data were analyzed statistically to determine differences among genotypes and moisture regimes. The significance of treatment differences was assessed at the 5% probability level. The responses of individual genotypes under rainfed conditions were compared with those under irrigated conditions to identify genotypes showing better biochemical adaptation to water stress.

3. Results and Discussion

3.1 Chlorophyll content

Chlorophyll content differed significantly among rice genotypes under rainfed and irrigated conditions. (Table 1) A reduction in chlorophyll content was observed under rainfed conditions in almost all genotypes, indicating the adverse effect of water deficit on photosynthetic pigments. The genotype mean chlorophyll content ranged from 16.01 to 31.64 SPAD under rainfed conditions and from 22.17 to 38.27 SPAD under irrigated conditions. Under rainfed conditions, Poornima (LC) recorded the highest chlorophyll content (31.64 SPAD), followed by RFU 329 (29.54 SPAD), whereas RFU 318 recorded the lowest value (16.01 SPAD). The reduction in chlorophyll under water stress may be associated with impaired chlorophyll synthesis, enhanced pigment degradation and damage to the photosynthetic apparatus. However, the maintenance of comparatively higher chlorophyll content in Poornima (LC) and RFU 329 suggests better preservation of photosynthetic capacity under limited water availability. Such genotypes may maintain greater photosynthetic efficiency and consequently exhibit better adaptation to drought stress. Similar reductions in chlorophyll content under water stress and greater pigment retention in tolerant rice genotypes were

reported by Kumar *et al.* (2015)^[2], Singh *et al.* (2018) and Navya *et al.* (2019).

3.2 Proline content

Proline accumulation increased markedly under rainfed conditions compared with irrigated conditions, indicating an active osmotic adjustment response of rice genotypes to water stress. (Table 1) The genotype mean proline content ranged from 9.03 to 17.12 $\mu\text{mol g}^{-1}$ FW under rainfed conditions, compared with 1.97 to 5.02 $\mu\text{mol g}^{-1}$ FW under irrigated conditions. Poornima (LC) recorded the highest proline content under water stress (17.12 $\mu\text{mol g}^{-1}$ FW), followed by RFU 329 (15.55 $\mu\text{mol g}^{-1}$ FW), while RFU 325 recorded the lowest value (9.03 $\mu\text{mol g}^{-1}$ FW). The increased accumulation of proline under drought may contribute to osmotic adjustment, maintenance of cellular water balance and stabilization of proteins and membranes under reduced water availability. Proline also functions as a compatible solute and may protect cellular components from stress-induced damage. The higher proline accumulation observed in Poornima (LC) and RFU 329 indicates a comparatively stronger biochemical response to water deficit and may contribute to their better adaptation under rainfed conditions. Similar increases in proline accumulation in response to drought stress were reported by Kumar *et al.* (2014)^[1] and Sahoo *et al.* (2020)^[7].

3.3 Ascorbate peroxidase activity

Ascorbate peroxidase (APX) activity increased significantly under rainfed conditions compared with irrigated conditions, reflecting activation of the antioxidant defense system in response to water stress. (Table 1) Genotype mean APX activity ranged from 4.41 to 11.41 $\mu\text{mol g}^{-1}$ under rainfed conditions, compared with 1.50 to 4.45 $\mu\text{mol g}^{-1}$ under irrigated conditions. Poornima (LC) recorded the highest APX activity (11.41 $\mu\text{mol g}^{-1}$), followed by RFU 329 (11.00 $\mu\text{mol g}^{-1}$), whereas RFU 320 showed the lowest activity (4.41 $\mu\text{mol g}^{-1}$) under rainfed conditions. Water deficit generally promotes the generation of reactive oxygen species (ROS), including hydrogen peroxide, which may cause oxidative damage to cellular components. APX plays an important role in scavenging hydrogen peroxide and thereby contributes to the protection of membranes and other cellular structures. The enhanced APX activity observed under rainfed conditions therefore suggests a stronger antioxidant response to drought-induced oxidative stress. The comparatively higher activity in Poornima (LC) and RFU 329 may indicate their greater capacity to minimize oxidative damage and maintain cellular stability under water limitation. Similar observations were reported by Nahar *et al.* (2018)^[4], Kumari *et al.* (2019)^[3] and Sahoo *et al.* (2019)^[6].

Table 1: Chlorophyll, proline and APX responses of rice genotypes under rainfed and irrigated conditions

Rice genotype	Chlorophyll (SPAD)		Proline ($\mu\text{mol g}^{-1}$ FW)		APX ($\mu\text{mol g}^{-1}$)	
	Irrigated	Rainfed	Irrigated	Rainfed	Irrigated	Rainfed
RFU 301	28.54	26.70	3.89	9.90	1.78	4.95
RFU 302	28.62	21.27	3.10	10.60	2.08	5.66
RFU 303	30.52	23.10	3.75	10.77	2.48	6.62
RFU 304	32.54	28.50	4.23	14.10	3.64	9.39
RFU 305	22.97	15.05	3.28	9.29	2.08	5.66
RFU 306	23.20	16.44	2.84	9.47	2.63	6.98
RFU 307	28.00	20.37	3.19	9.21	1.78	4.95
RFU 308	30.61	23.27	4.10	10.60	1.94	5.34
RFU 309	29.61	22.47	3.80	10.51	2.35	6.30
RFU 310	30.31	23.18	3.97	10.68	2.13	5.79
RFU 311	24.28	17.24	4.15	10.86	2.48	6.62
RFU 312	25.01	17.85	3.21	11.73	2.58	6.85
RFU 313	24.29	16.90	2.84	9.55	1.94	5.34
RFU 314	31.72	25.00	3.97	10.68	1.82	5.18
RFU 315	27.81	21.58	2.84	9.55	1.75	4.57
RFU 316	22.27	16.45	3.90	13.03	1.70	4.50
RFU 317	23.16	17.55	3.56	13.64	2.60	7.04
RFU 318	22.17	16.01	2.41	9.12	2.35	6.43
RFU 319	22.65	16.50	3.02	9.73	2.47	6.72
RFU 320	24.10	18.04	4.10	12.94	1.50	4.41
RFU 321	31.43	25.37	3.80	11.64	2.83	7.59
RFU 322	30.62	24.28	4.15	11.20	3.51	9.20
RFU 323	28.61	22.77	3.25	12.07	3.37	8.87
RFU 324	31.25	26.45	3.28	10.34	3.45	9.04
RFU 325	29.54	27.60	1.97	9.03	3.24	8.52
RFU 326	30.03	26.40	2.67	9.47	3.16	8.33
RFU 327	33.45	29.10	4.50	14.25	3.99	10.32
RFU 328	31.94	27.45	3.90	12.77	2.94	7.81
RFU 329	35.25	29.54	4.70	15.55	4.28	11.00
Poornima (LC)	38.27	31.64	5.02	17.12	4.45	11.41
Mean	28.43	22.47	3.58	11.31	2.64	7.05
CD	2.04	3.8	0.31	1.51	0.14	0.39
SEm $\pm$	0.72	1.34	0.10	0.47	0.04	0.11

3.4 Superoxide dismutase activity

Superoxide dismutase (SOD) activity showed a significant increase under rainfed conditions. (Table 2) The genotype mean SOD activity ranged from 1.28 to 3.55 U mg^{-1} under rainfed conditions, compared with 0.10 to 0.43 U mg^{-1} under irrigation. The highest SOD activity under rainfed conditions was recorded in Poornima (LC) (3.55 U mg^{-1}), followed by RFU 329 (3.33 U mg^{-1}), whereas RFU 310 recorded the lowest value (1.28 U mg^{-1}). Increased SOD activity indicates enhanced capacity to detoxify superoxide radicals generated during drought stress. Similar results were reported by Sahoo *et al.* (2019) [6].

3.5 Peroxidase activity

Peroxidase (POD) activity was significantly higher under rainfed conditions than under irrigation. (Table 2). The genotype mean POD activity ranged from 5.33 to 11.28 U mg^{-1} protein under the reported moisture conditions, with Poornima (LC) recording the maximum activity (11.28 U mg^{-1} protein), followed by RFU 329 (10.77 U mg^{-1} protein).

RFU 319 recorded the minimum value (5.33 U mg^{-1} protein). The enhanced POD activity under water stress suggests an increased enzymatic antioxidant response to oxidative stress. Similar results were reported by Sahoo *et al.* (2020) [7].

3.6 Catalase activity

Catalase (CAT) activity also increased significantly under rainfed conditions. (Table 2) The genotype mean CAT activity ranged from 0.33 to 0.65 U mg^{-1} protein under rainfed conditions, compared with 0.07 to 0.21 U mg^{-1} protein under irrigation. Poornima (LC) recorded the highest CAT activity under rainfed conditions (0.65 U mg^{-1} protein), followed by RFU 329 (0.58), whereas RFU 315 recorded the lowest value (0.33). The higher CAT activity under water stress indicates an enhanced capacity for detoxification of hydrogen peroxide and protection against oxidative damage. Similar findings were reported by Kumari *et al.* (2019) [3] and Sahoo *et al.* (2020) [7].

Table 2: SOD, POD and CAT activities of rice genotypes under rainfed and irrigated conditions

Rice genotype	SOD (U mg ⁻¹)		POD (U mg ⁻¹ protein)		CAT (U mg ⁻¹ protein)	
	Irrigated	Rainfed	Irrigated	Rainfed	Irrigated	Rainfed
RFU 301	0.23	2.15	2.53	9.14	0.11	0.38
RFU 302	0.12	1.32	2.40	8.26	0.11	0.39
RFU 303	0.24	2.18	2.00	7.42	0.13	0.45
RFU 304	0.32	2.91	2.70	10.12	0.14	0.51
RFU 305	0.28	2.54	1.82	6.91	0.10	0.46
RFU 306	0.30	2.64	2.07	7.51	0.09	0.35
RFU 307	0.31	2.76	1.98	7.35	0.11	0.39
RFU 308	0.20	1.82	2.08	7.68	0.11	0.40
RFU 309	0.21	1.96	1.95	7.28	0.12	0.43
RFU 310	0.10	1.28	1.82	6.91	0.12	0.41
RFU 311	0.23	2.15	2.13	7.89	0.10	0.36
RFU 312	0.18	1.77	2.16	7.98	0.09	0.35
RFU 313	0.29	2.70	2.08	7.68	0.09	0.34
RFU 314	0.29	2.64	1.72	6.63	0.09	0.35
RFU 315	0.20	1.96	1.59	6.14	0.07	0.33
RFU 316	0.24	2.24	1.37	5.58	0.12	0.45
RFU 317	0.28	2.80	1.49	5.84	0.12	0.50
RFU 318	0.24	2.24	1.40	5.68	0.13	0.49
RFU 319	0.28	2.57	1.31	5.33	0.09	0.47
RFU 320	0.26	2.38	1.43	5.65	0.11	0.50
RFU 321	0.22	2.08	2.13	7.93	0.08	0.48
RFU 322	0.18	1.85	1.85	7.02	0.12	0.41
RFU 323	0.11	1.40	1.98	7.40	0.12	0.43
RFU 324	0.15	1.59	2.08	7.72	0.13	0.43
RFU 325	0.13	1.44	1.82	6.95	0.12	0.41
RFU 326	0.12	1.35	2.05	7.65	0.09	0.35
RFU 327	0.35	3.06	2.89	10.26	0.15	0.55
RFU 328	0.26	2.38	1.98	7.40	0.13	0.49
RFU 329	0.39	3.33	3.02	10.77	0.17	0.58
Poornima (LC)	0.43	3.55	3.17	11.28	0.21	0.65
Mean	0.24	2.22	2.03	7.58	0.12	0.43
CD	0.03	0.21	0.13	0.49	0.03	0.06
SEm ±	0.01	0.06	0.05	0.18	0.01	0.02

4. Conclusion

The study demonstrated that rainfed conditions substantially altered the biochemical responses of rice genotypes compared with irrigated conditions. Water stress generally reduced chlorophyll content but enhanced proline accumulation and the activities of APX, SOD, POD and CAT, indicating activation of osmotic adjustment and antioxidant defense mechanisms. Among the evaluated genotypes, Poornima (LC) and RFU 329 consistently showed higher biochemical responses under rainfed conditions, particularly for proline accumulation and antioxidant enzyme activities. These genotypes may therefore possess better biochemical adaptation to water stress and could be useful for further evaluation in drought-tolerance breeding programmes.

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