



Exploring drought mitigation options for improvement of physiology, yield and water use efficiency of aerobic rice (*Oryza sativa*)

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ABSTRACT

An experiment was conducted during summer seasons of 2022 and 2023 in Wetland Farms at Tamil Nadu Agricultural University, Coimbatore, Tamil Nadu to mitigate the ill effects of moisture stress during the flowering stage of rice (*Oryza sativa* L.) by moisture stress mitigating chemicals. The trial was laid out in split plot design (SPD) with 3 main plots and 4 sub-plots, and replicated thrice. In main plot, irrigation at IW/CPE ratio 1.0 from emergence to physiological maturity (M_1); withholding irrigation (drought stress) for 8 days from heading stage (M_2); and withholding irrigation (drought stress) for 12 days from heading stage (M_3) and in sub-plot drought stress mitigating chemicals, viz. no spray (S_1); salicylic acid spray @100 ppm (S_2); sodium selenate @20 ppm (S_3); and pink pigmented facultative methylotrophs (PPFM) @1% (S_4) were tested. Results revealed that irrigation at IW/CPE 1.0 from crop emergence to physiological maturity along with foliar spray of selenium @20 ppm at heading stage had beneficial impact on growth, physiological parameters, yield attributes and yield. Drought stress given for 8 days from heading stage with selenium spray recorded lesser detrimental effect on growth characters, yield attributes and yield when compared to drought stress given for 12 days from heading stage in aerobic rice cultivation. The yield reduction in drought stress for 12 days from heading stage with selenium @20 ppm spray and drought stress for 8 days from heading stage with selenium @20 ppm spray were 23.6 and 16.5% respectively, when compared to irrigation at IW/CPE ratio 1.0 from crop emergence to physiological maturity with selenium spray @20 ppm. Water use efficiency and water profitability also increased by the application of stress mitigating chemical of sodium @20 ppm. From this study, it is recommended that, in aerobic rice cultivation if there is any drought occurrence during heading stage of crop, the selenium application @20 ppm can withstand drought even up to 8 days from heading stage.

Keywords: Aerobic rice, Heading stage, IW/CPE ratio, PPFM, Salicylic acid, Selenium

Rice (*Oryza sativa* L.) is the principal food source for more than half of the world population. In India, total rice cultivation area is 47 Mha with a production of 130 Mt (Indiastat 2022). Among the total rice cultivating area in India, around 18.8 Mha comes under rainfed condition. In Tamil Nadu, 6.80 Mha are cultivated lands, with rainfed crop production accounting for nearly 40–50% of total area and rice cultivating area is around 2.21 Mha producing 7.90 Mt with an average productivity of 3.49 t/ha (Indiastat 2022). Aerobic rice is a system of rice cultivation which specially developed for direct seeded varieties and grown in un-puddled and un-flooded aerobic soil. When compared to lowland rice, aerobic rice can save up to 50% of irrigation

water and increase water-use efficiency (WUE) and water productivity (Venuprasad *et al.* 2007). For production of rice there is a problem around the world due to many abiotic and biotic stresses. Among these, drought stress is a most effective one. Due to moisture stress, there was reduction in average rice grain production i.e. 21.0% during vegetative stage, 50.0% reduction at flowering stage and 21.0% decrease during grain filling stage (Sarvestani *et al.* 2008). The yield characteristics as well as the yield of rice are severely affected by moisture stress, which can result in decreased pollen viability, fewer grains and finally decreased grain yield. The increase in WUE is important to maximize the use of available water to increase photosynthesis and growth of plants under water-deficit conditions. Moisture stress mitigating chemicals, viz. selenium, salicylic acid and PPFM plays a major role to mitigate the negative effects of moisture stress during flowering stage of rice. The objective of the study was to study the effect of drought stress during the reproductive stage on physiology, yield and water productivity and

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also to understand the mechanism of drought mitigating chemicals in aerobic rice cultivation.

MATERIALS AND METHODS

The present experiment was conducted during summer seasons of 2022 and 2023 in Wetland Farms at Tamil Nadu Agricultural University, Coimbatore (77°E longitude and 11°N latitude with an altitude of 426.7 m amsl), Tamil Nadu. The soil of the experimental field was clay loam, alkaline in pH (8.5), normal in EC (0.45 dS/m), low in available nitrogen (235.2 kg/ha), medium in available phosphorus (16.5 kg/ha) and high in available potassium (526.1 kg/ha). The mean maximum temperature during the cropping period was 34.7°C and mean minimum temperature was 23.2°C. The total rainfall received during the cropping period was 77.9 and 54.2 mm, respectively, during the year 2022 and 2023. The experiment was conducted in a split-plot design (SPD) with 3 replications. In main plot, irrigation interval, viz. irrigation @IW/CPE ratio 1.0 from emergence to physiological maturity (M_1); withholding irrigation (drought stress) for 8 days from heading stage (M_2); withholding irrigation (drought stress) for 12 days from heading stage (M_3), and in sub-plots, stress mitigating chemicals, viz. no spray (S_1); salicylic acid @100 ppm (S_2); sodium selenate @20 ppm (S_3); and pink pigmented facultative methylotrophs (PPFM) @1% (S_4) were tested. Paddy variety CO 53 (duration 115 to 120 days) was taken up for sowing on 3rd February 2022 and 21st February 2023, respectively. Seeds were treated with carbendazim @2 g/kg of seeds and then thoroughly soaked and incubated overnight in a gunny bag. The sowing was taken up in lines by dibbling with a row spacing of 20 cm × 10 cm and a seed rate of 75 kg/ha was used. The date of sowing was 18th February 2022. In order to facilitate the good germination of seeds, irrigation was given immediately after sowing. The fertilizer schedule recommended for aerobic rice cultivation is 150:50:50 kg NPK/ha (CPG 2020). Full dose of phosphorus was applied as basal. Nitrogen and potassium were applied as 4 split doses at 15 days after sowing (DAS), tillering, panicle initiation and flowering stages. Pre emergence herbicide pendimethalin @1.0 kg a.i/ha was applied followed by early post emergence herbicide application of bispyribac sodium @30 g a.i/ha. One hand weeding was done on 45 DAS. Irrigation was given based on the IW/CPE ratio of 1.0 with 3 cm depth of water from emergence to physiological maturity stage for the treatment of M_1 . Withholding irrigation (drought stress) for 8 days and 12 days from the heading stage for the treatment M_2 and M_3 , respectively. The known quantity of water (30 mm) was applied when the cumulative pan evaporation reaches a predetermined level. Polythene sheet with a length of 20 m and width of 0.5 m was inserted into the soil and the remaining 25 cm folded on the bunds for arresting the seepage of water for the M_1 , M_2 and M_3 . Mud plastering was done on the bunds along with the polythene sheets. In sub plot, salicylic acid (SA) was applied as a foliar spray (spray volume 500 litre/ha) at the concentration of 100 ppm (100 mg/litre of water or 50

g/ha) during heading stage of the crop (S_2). Sodium selenate (Se) was applied as a foliar spray (spray volume 500 litre/ha) for the source of selenium at the concentration of 20 ppm (20 mg/litre of water or 10 g/ha) during heading stage of the crop (S_3). PPFM was applied as a foliar spray (spray volume 500 litre/ha) at the concentration of 1% (10 ml/litre of water or 5 litre/ha) during heading stage of the crop (S_4). The physiological parameter of chlorophyll stability index (CSI) and biochemical properties of proline content were recorded at initial (90 DAS), middle of the drought (98 DAS), end of the drought (106 DAS) and after recovery of the crop (116 DAS). The chlorophyll stability index was measured using the conventional approach (Murphy 1962) and represented as percentage. Proline content was measured by using the standard procedures described by Bates *et al.* (1973) and expressed as µg/g of fresh weight. The observations on yield attributes, viz. number of filled grains/panicle, grain conversion efficiency, grain yield (kg/ha) were recorded. Water parameter such as water-use efficiency and water profitability were calculated. The water use efficiency was determined by the ratio of grain yield and total water used (Viets 1962) and the water profitability was calculated by function of gross income to the total water used by the crop throughout its growth and expressed in ₹/ha mm. The crop was harvested on 25th March 2022 and 28th March 2023, during the year 2022 and 2023, respectively. The data on various characters were statistically analysed as suggested by Gomez and Gomez (1984). Wherever statistical significance was observed critical difference (CD) @ $P=0.05\%$ level of probability was worked out for comparison of mean data.

RESULTS AND DISCUSSION

Physiological parameters

Chlorophyll stability index (CSI): The chlorophyll stability index (CSI) was influenced significantly by the drought stress and foliar application of stress mitigating chemicals in aerobic rice at heading stage (Table 1). The CSI decline by the imposition of drought stress. Up to 90 days, there was no significant difference among the irrigation treatments. After the drought impose, the treatment M_3 (withhold irrigation up to 12 days from heading) recorded the lower CSI of 71.7, 67.1 and 70.8% on 98 DAS, 106 DAS and 116 DAS, respectively. Higher CSI of 81.3, 81.9 and 82.3% was resulted in M_1 treatment (irrigation @IW/CPE 1.0 from emergence to physiological maturity) on 98 DAS, 106 DAS and 116 DAS, respectively. The CSI indicates the capacity of the plant to endure against drought and it reduced due to increased temperature within plant tissues. Lower CSI was noticed as a result of the drought stress. Withhold irrigation up to 12 days from heading stage (M_3) recorded lower CSI and no drought stress treatment recorded higher CSI. Long-term drought stress might have raised internal temperatures in the plant, which might have led to the instability of the chlorophyll. The ability of plant tissue to regulate temperature was hampered as a result

Table 1 Effect of drought stress and foliar application of stress mitigating chemicals on chlorophyll stability index, total number of grains/panicle, grain conversion efficiency, water use efficiency and water profitability in aerobic rice (pooled data of 2 years)

Treatment	Chlorophyll stability index (%)				Total number filled grains/panicle	Grain conversion efficiency (%)	Water use efficiency (kg/ha/mm)	Water profitability (₹/ha mm)
	At heading stage (90 DAS)	98 DAS	106 DAS	116 DAS				
<i>Drought stress</i>								
M ₁	80.2	81.3	81.9	82.3	76.4	82.5	4.9	96.2
M ₂	82.7	73.8	75.8	78.4	63.2	75.0	4.7	87.2
M ₃	79.2	71.7	67.1	70.8	50.1	66.2	0.03	77.9
SEd	1.8	2.1	1.9	1.7	1.8	-	0.10	2.0
CD (<i>P</i> =0.05)	NS	6.09	5.4	4.7	5.0	-		4.7
<i>Stress mitigating chemicals</i>								
S ₁	78.2	70.9	67.8	70.4	51.1	68.2	4.7	80.5
S ₂	82.4	76.9	77.9	78.8	66.6	76.8	5.1	88.8
S ₃	80.7	80.0	80.3	84.0	76.3	82.8	5.4	94.6
S ₄	81.4	74.4	73.8	75.2	58.8	72.8	4.9	86.9
SEd	2.3	2.2	2.2	2.7	1.9	-	0.02	3.2
CD (<i>P</i> =0.05)	NS	4.72	4.64	4.78	4.2	-	0.05	6.8

Treatment details are given under Materials and Methods. DAS, Days after sowing; NS, Non-significant.

of the increased severity of moisture stress near the root zone. As a result, chlorophyll pigments degraded under higher temperatures and relative water content in plants decreased, which may account for the lower CSI under moisture stress conditions. The CSI values were reduced as a result of increased drought stress in rice (Nahakpam 2017, Yang *et al.* 2019).

Higher value of CSI, 80.0, 80.3 and 84.0% recorded in S₃ treatment due to sodium selenate application @20 ppm at heading stage on 98 DAS, 106 DAS and 116 DAS, respectively. The lowest CSI 70.9, 67.8 and 70.4% was recorded on 98 DAS, 106 DAS and 116 DAS, respectively in no spray treatment (S₁). Selenium had increased CSI in both under normal and drought situations. In order to withstand drought, selenium has enhanced proline synthesis and stabilised chlorophyll production. As a result, rice have been able to withstand drought stress and enhance its CSI by accelerating the anti-oxidative property and selenium application prevented the formation of redox oxygen supply (Khepar *et al.* 2000). This may also contribute to higher chlorophyll stability indexing treatments using selenium spray.

Proline content (µg/g): The proline content was influenced significantly by the drought stress and foliar application of stress mitigate chemicals in aerobic rice at heading stage (Fig. 1). The proline content was noticed to get increased by the imposition of drought stress. Treatment M₃ (withhold irrigation upto 12 days from heading) recorded

the higher proline content of 107.9, 129.7 and 114.2 µg/g on 98 DAS, 106 DAS and 116 DAS, respectively. Lower proline content 75.5, 75.5 and 67.5 µg/g were resulted in M₁ treatment (irrigation @IW/CPE 1.0 from crop emergence to physiological maturity) on 98 DAS, 106 DAS and 116 DAS, respectively. A sign of water stress in plants is the build-up of free proline (Patnaik *et al.* 2020). In comparison to the control, the drought-imposed treatments (M₂ and M₃) had a greater proline content. As a result of drought stress free amino acids like proline, an osmolyte produced in a water shortage environment to help osmotic adjustment (Anjum *et al.* 2017). Under no drought stress treatment (M₁), which was irrigated at 'IW/CPE' ratio 1.0 from crop emergence to physiological maturity, there was less proline production. Treatment M₃ recorded 29.2, 41.8 and 40.9% higher proline content than M₁ due to drought stress at 98, 106 and 116 DAS, respectively. Patnaik *et al.* (2020) and Monisha *et al.*

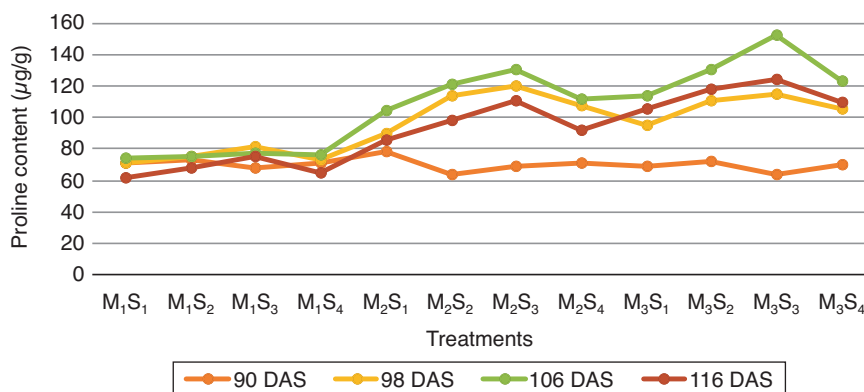


Fig. 1 Effect of drought stress and foliar application of stress mitigating chemicals on proline content (µg/g) of aerobic rice.

Treatment details are given under Materials and Methods. DAS, Days after sowing; NS, Non-significant.

(2021) reported comparable outcomes of higher proline content in drought stress in paddy for osmotic adjustment.

In sub plot, higher value of proline recorded in S_3 due to the sodium selenate application @20 ppm recorded 105.7, 120.1 and 103.3 $\mu\text{g/g}$ on 98, 106 and 116 DAS, respectively. Lower proline content 85.6, 97.6 and 84.1 $\mu\text{g/g}$ were recorded on 98, 106 and 116 DAS, respectively in S_1 . Selenium applied treatment increased proline accumulation in rice plants. S_3 treatment recorded 19.0, 18.7 and 18.6% more proline content than S_1 . Selenium treated plants might have produce more proline as a result of the enzymes function in proline metabolism. Proline has the ability to bond with metal ions due to its chelating characteristic indicates that stressed plants might use this as a defence mechanism (Monisha *et al.* 2021). Foliar application of selenium in rice increase proline concentration (Andrade *et al.* 2018).

Yield attributes and yield

Number of filled grains/panicle: Higher number of filled grains (76.4/panicle) were observed with treatment M_1 (irrigation @IW/CPE ratio 1.0 from crop emergence to physiological maturity) compared to drought stress given up to 8 days (M_2) and 12 days (M_3) from heading stage. The lowest number of filled grains (50.1/panicle) was resulted in 12-days drought stress (M_3) from heading stage of aerobic rice. M_3 treatment recorded 37.2% lesser filled grains/panicle than M_1 treatment. In rice grain filling determines the weight which in turn influences the overall yield. The similar results were obtained in rice by Dada *et al.* (2020). The number of filled grains/panicle decreased as a result of the drought conditions which occurred at the time of blooming. Lack of water during the reproductive phase had negative impact on rice photosynthesis and grain filling (Barnaby *et al.* 2019). In sub plot, foliar spray of sodium selenate @20 ppm (S_3) produced significantly a higher number of filled grains (76.3/panicle) compared to foliar application of salicylic acid @100 ppm (S_2), PPFM @1% and no spray at heading stage. The least number of filled grains (51.1/panicle) recorded in no spray treatment (S_1). Treatment S_3 (sodium selenate @20 ppm) recorded 26.9% higher filled grains/panicle than S_1 (no spray) treatment.

Grain conversion efficiency: Irrigation given at IW/CPE ratio 1.0 (M_1) showed higher grain conversion efficiency (82.5%). While, the lowest grain conversion efficiency (66.2%) was recorded in withhold irrigation up to 12 days from heading (M_3). In foliar application of drought stress mitigating chemicals at heading stage, higher grain conversion efficiency (82.8%) was registered in S_3 (sodium selenate @20 ppm) treatment. Whereas, the least grain conversion efficiency of 68.2% was resulted in no spray S_1 treatment.

Grain yield: The grain yield was influenced significantly by induced drought stress and foliar application of stress mitigating chemicals at heading stage (Table 2). Irrigation at IW/CPE ratio 1.0 from crop emergence to physiological maturity (M_1) recorded higher grain yield which was

Table 2 Interaction effect of drought stress and stress mitigating chemicals on grain yield of aerobic rice (pooled data of 2 years)

Treatment	M_1	M_2	M_3	Mean
S_1	3668	3156	2771	3198
S_2	3942	3470	3004	3472
S_3	4221	3536	3242	3666
S_4	3795	3419	2850	3355
Mean	3906	3395	2967	
	SEd		CD ($P=0.05$)	
M	39		106	
S	31		64	
$M \times S$	59		140	
$S \times M$	52		109	

Treatment details are given under Materials and Methods.

followed by the drought stress given up to 8 days from heading (M_2). The lowest grain yield was recorded in drought stress given up to 12 days from heading stage (M_3). Treatment M_1 (irrigation at 'IW/CPE' ratio 1.0 from crop emergence to physiological maturity) which received constant moisture resulted higher grain yield. The continuous availability of moisture might have enhanced plant photosynthetic capacity and led to a higher build-up of photosynthates in the form of dry matter production, improving yield characteristics and increasing grain yield. The yield reduction due to moisture stress in treatment M_2 (drought stress up to 8 days from heading) and M_3 (drought stress up to 12 days from heading) was 13.1% and 24.0%, respectively, when compared to M_1 (irrigation at IW/CPE ratio 1.0 from crop emergence to physiological maturity). The drought imposition during heading stage (M_2 and M_3) showed lesser filled grains per panicle and increasing sterility might be reason for declining grain yield. The reduced plant nutrient uptake also resulted in decreased growth and photosynthesis in drought imposed treatment (M_2 and M_3). A decrease in the amount of chlorophyll, fewer productive tillers being produced and lesser dry matter accumulation could have all contributed to decline in grain yield in drought imposed during heading stage (Duan *et al.* 2020).

In sub plot, sodium selenate application @20 ppm (S_3) at heading stage resulted higher grain yield. This was followed by salicylic acid @100 ppm (S_2) which was comparable with PPFM @1% (S_4). Application of drought mitigating chemical such as selenium spray increased the grain yield in both normal and drought situations. The antioxidant activities in plant cells might have increased as a result of the selenium treatment, which could have promoted plant tissue growth and increased grain yield (Badawy *et al.* 2021). Selenium enhanced the dry matter accumulation and grain production in rice both in well-watered and drought conditions. Selenium has improved physiological and biochemical parameters, boosted yield qualities (number of filled grains/panicle) and improved drought tolerance.

These improvements have finally resulted in the formation of higher grain yield. There have been reports that under conditions of water scarcity, selenium application boosted production of paddy (Monisha *et al.* 2021).

The interaction had significant effect with irrigation interval and application of stress mitigating chemicals. Higher grain yield was recorded in irrigation at IW/CPE ratio 1.0 with sodium selenate application (M_1S_3) recorded the grain yield of 4221 kg/ha. Lesser grain yield was recorded in withhold irrigation up to 12 days from heading with no spray (M_3S_1) resulted 2771 kg/ha. Drought stress given up to 8 days from heading stage with selenium spray recorded lesser detrimental effect on yield when compared to drought stress given for 12 days from heading stage in aerobic rice cultivation. The yield reduction in the treatment M_3S_3 (drought stress for 12 days from heading stage with selenium @20 ppm spray) and M_2S_3 (drought stress for 8 days from heading stage with sodium selenate spray @20 ppm spray) was 23.2 and 16.3%, respectively, when compared to the treatment M_1S_3 (irrigation at IW/CPE ratio 1.0 from crop emergence to physiological maturity with selenium spray @20 ppm).

Water use efficiency (WUE) and water profitability (WP): The water use efficiency was shown (Table 1) to be much lower and to be decreased under the conditions of drought stress. The ability of the crop to access water in the soil and use it effectively is important to better crop establishment. The higher water use efficiency and water profitability could be increased either by increasing yield or by maintaining the yield level with a reduced quantity of water. Treatment M_1 recorded higher water use efficiency 5.4 kg/ha/mm and water profitability 96.2 ₹/ha mm due to increase in yield. Lower water use efficiency and water profitability were observed in treatment M_3 which led to lower yield. These findings are in accordance with those of Kannan (2017) and Patnaik *et al.* (2020). In sub plot treatment, foliar spraying of stress mitigating chemicals increased the water use efficiency and water profitability in aerobic rice. Application of selenium @20 ppm (S_3) recorded higher water use efficiency (5.4 kg/ha/mm) and water profitability (96.2 ₹/ha-mm). Selenium might have the beneficial effects on plant growth and development. These findings are in accordance with Monisha *et al.* (2021).

From this 2-year study, it can be concluded that irrigation at IW/CPE 1.0 from crop emergence to physiological maturity along with foliar spray of selenium @20 ppm at heading stage had beneficial impact on growth, physiological parameters, yield attributes and yield in aerobic rice cultivation. Drought stress given up to 8 days from heading stage with selenium spray recorded lesser detrimental effect on growth characters, yield attributes and yield when compared to drought stress given up to 12 days from heading stage. The yield reduction in drought stress for 12 days from heading stage with selenium @20 ppm spray and drought stress for 8 days from heading stage with selenium @20 ppm spray was 23.6 and 16.5% respectively, when compared to irrigation at IW/CPE ratio 1.0 from

emergence to physiological maturity with selenium spray @20 ppm. Water use efficiency and water profitability also increased by applying stress mitigating chemical of sodium selenate @20 ppm.

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