

Modulating effect of PGRs on Growth and Biochemical traits in rice (*Oryza sativa* L.) under salt stress

ABSTRACT

An investigation was conducted to study the effect of plant growth regulators on rice (variety Sarju-52) grown under sodic conditions, during the *khari* season 2018 at the students' instructional farm of ANDUA&T, Kumarganj, Ayodhya. The experiment was laid out in randomized block design with nine treatments and three replications, with different concentrations and modes of application of plant growth regulators (PGRs). The PGRs were applied as follows- seed priming of GA₃ @25 ppm & 50 ppm and SA @ 10 ppm & 25 ppm, and foliar application of GA₃ and SA, each @ 100 ppm & 200 ppm at 30 (DAT), along with untreated control. Seed priming was done for 12 hours in different concentrations of PGRs. Thirty five days old seedlings were transplanted to the field from the nursery. Observations were recorded on various growth parameters such as plant height, number of tillers, dry biomass/hill, relative water content and biochemical parameters like chlorophyll, and TSS, at 30, 60 and 90 DAT and at maturity (harvesting stage). Both seed priming and foliar application of PGRs positively influenced the growth characters, chlorophyll content, and total soluble sugars. Moreover, foliar spray of GA₃ @200 ppm was found to have a more pronounced effect in improving the yield of the variety studied, among all the treatments, under sodic conditions. It can be concluded that foliar spray of GA₃ @ 200 ppm is effective in improving the growth and yield of rice grown under salt stress.

Keywords: GA₃, Salicylic acid, TSS, chlorophyll content, salt stress tolerance

INTRODUCTION

There are altogether 24 species of genus *Oryza* of which 22 are wild and two species (*Oryza sativa* L.) and (*Oryza glaberrima* L.) are cultivated. (*Oryza sativa* L.) is grown in all rice growing areas, but (*Oryza glaberrima* L.) is confined to the West Africa only. Thus it indicates that there might have been two centers of origin of our cultivated rice; South-eastern Asia and West Africa. Rice has shaped the culture, diets and economic of thousand millions peoples. For more than half of the peoples "rice is lifeline". Importance of rice is an important staple food crop for more than 60 per cent of the world people. Rice is a critical source of food for more than 3 billion people annually. Rice is cultivated in 45 million hectare which is with a production of 120 million tones (Financial year 2020-2021) and productivity is 2550 kg/ha of our country [7] [3].

Soil sodicity/salinity has become a severe threat to ensure global food security. The total world wide area of the salt affected land is about 190 million ha and about 48 million ha in South and Southeast Asia. In India, sodic soils have occupied 3.77 million ha salt affected area. Sodic soils are essentially located in the Indo-Gangetic plain, arid and semiarid region in western and central India and the peninsular region in the southern India. Salinity stress is the major factors limiting the germination and productivity of crop plants because mostly crop plants are susceptible to salinity caused by higher concentration of salt in the soil and constitutes a issue concerned a significant segment of the plant, mainly in region with dry and hot climates.[5] Excessive salts in the soils may alter its physical and chemical properties, including soil structure and hydraulic conductivity. High exchangeable sodium and pH decrease soil permeability, available water capacity and infiltration rates through swelling and dispersion of clays as well as slaking of soil aggregates. Sodicity reduces the ability of plants to take up water and this causes reductions in growth rate, along with a suite of metabolic changes identical to those caused by water stress, whereas the water available in the salt affected soil is contaminated with anions and cations, inducing osmotic stress. In India, 42.7 million ha of land is under rice cultivation. Salt stresses adversely affect natural

productivity and causes significant crop loss worldwide. Excessive salts in the soils may alter its physical and chemical properties, including soil structure and hydraulic conductivity [11].

Exogenously applied growth regulators can alter the content of endogenous phyto-hormones. The biosynthesis of plant hormones is regulated by both developmental and environmental [19]. Gibberellic acid (GA₃) is the most important growth regulator, which breaks seed dormancy, promotes germination, increases inter nodal length, plant hypocotyl growth and cell division in the cambial zone, flower formation, fruit set and increases the size of leaves. [1][10].

Salicylic acid (SA) is a water-soluble antioxidant compound that can also use regulate plant growth. It also has a role in abiotic stress tolerance such as drought and salt tolerance in wheat. Ameliorative effect of SA on growth of crop plants under abiotic stress conditions may have been due to its role in nutrient uptake, water relations, stomatal regulation, photosynthesis and growth. Salicylic acid (SA) is an endogenous growth regulator and belongs to a group of phenol compounds. It participates in the regulation of physiological processes and also provides protection against biotic and abiotic stress such as salinity. Positive effects of Salicylic Acid for salt tolerance have been studied in many crops [9]. Seed priming techniques have been used to increase germination, improve germination uniformity, improve seedling establishment, induce nuclear DNA synthesis and stimulate vegetative growth in more field crops under stressed conditions. Also, reported that priming by salicylic acid and gibberellins have been used to increase germination characteristics in rye and other crop. [10],[2].

Materials and Methods

The present investigation entitled “**Modulating effect of PGRs on Growth and Biochemical traits in rice (*Oryza sativa L.*) undersalt stress**” was carried out in *kharif* season, during 2018 at the student instructions farm of Narendra Deva University of Agriculture and Technology, Kumarganj, Ayodhya, (U.P.), India. In the present study, Variety sarju-52 under salt stress condition was taken as experimental materials to find out the response of PGRs on growth and biochemical changes traits in rice. The experiment was laid out in RBD with three replications and nine treatments. Healthy seeds of rice variety Sarju-52 were soaked in the solution of various concentration of GA₃ (25 ppm and 50 ppm) & salicylic acid (10 ppm and 25 ppm) distilled water for hydro-priming, for 12 hours along with untreated control. After that seeds were taken out from the solution and distilled water and dried for one hour in shade then seed showing in nursery. Various foliar application of GA₃ (100 ppm and 200 ppm) and salicylic acid (100 ppm and 200 ppm) were applied as foliar application at 30 DAT. Growth parameters viz. plant height, number of tillers hill⁻¹ and dry biomass plant⁻¹ and Relative water content. Bio-chemical parameters viz. Chlorophyll content, Total soluble sugar of crops were also observed were found superior with foliar application of GA₃ 200 ppm followed GA₃ 100 ppm.

OBSERVATIONS RECORDED:

1. Growth and growth analysis: Growth related observations were recorded at four stages of crop growth i.e. 30th, 60th and 90DAT and harvesting stage. Five plants under each treatment were tagged initially which were used for growth measurement.

A) Plant height (cm): Plant height was measured in cm from soil surface up to tip of the plant with the help of a meter scale. Measurement was done on the plants which were initially tagged for this purpose and average height was collected from the replicated data.

B) Number of tillers plant⁻¹: Number of tillers per plant under each treatment was recorded by counting at the appropriate stage. The plants already tagged for this purpose were used and an average tillers plant⁻¹ were computed.

C) Dry Biomass plant⁻¹ (g): Five plants were uprooted from each treatment and roots were removed with the help of a blade. The samples were cleaned and then oven dried at 70±5⁰C till a constant weight was achieved. The weight was recorded with the help of an electronic balance and average biomass plant⁻¹ was recorded.

D) Relative water content (RWC %): The relative water content (RWC) was determined by the method described by Turner and Beg (1981). Leaf discs were cut from the leaves, weighed and saturated by floating on distilled water in Petridis for six hours. The discs were surface dried and weighed. After that discs were kept in oven at 70±5⁰ C for 24 hours. After drying, weight of discs was calculated with the help of electronic balance. RWC was calculated by the following

$$\text{Formula: RWC (\%)} = \frac{\text{Fresh weight} - \text{Dry weight}}{\text{Saturated weight} - \text{Dry weight}} \times 100$$

2. BIOCHEMICAL STUDIES:

All the biochemical measurements was done in leaf sample.

A) Chlorophyll content (SPAD Value):Chlorophyll content was measured with the help of Plant efficiency analyzer (Model X 55/M-PEA) the values were expressed as SPAD value.

B) Total soluble sugar content (mg g⁻¹ dry weight):The total soluble sugar content in shoot was estimated by the method of Yemm and Wills (1954)

Reagents:

Ethanol (80%)

Anthrone (0.2%)

Glucose

Anthrone reagent: 200 mg Anthrone in 100 ml concentrated H₂SO₄

Procedure:

100 mg dried plant sample was homogenized in 10 ml of ethanol (80% concentration) centrifuged at 4000 rpm for 20 minutes. Supernatant was collected and residue was re-extracted with 10 ml of 80% ethanol and again centrifuged at 4000 rpm for 20 minutes. Supernatant was collected and both the supernatants were combined and made up to final volume of 20 ml. 0.2 ml supernatant was dried in a test tube on water bath and cooled to room temperature. One ml of distilled water was added to each test tube and mixed thoroughly. 4 ml of anthrone reagent was added along the wall of the test tube and mixed slowly and heated on water bath at 100⁰C for 10 minutes and cooled rapidly under running cold water. Absorbance was measured at 620 nm against the reagent blank. The amount of total carbohydrate present in the extract was calculated using a standard curve prepared from graded concentration of glucose.

Results and Discussion

Plant height

All treatments showed increasing trend in plant height as compared to control with advancement of stages (Fig.1) As effectiveness of mode of application of PGRs response of foliar application on plant

height was found more pronounced than seed soaking treatment. At 30 DAT, maximum plant height was recorded with treatment T3 (seed soaking GA₃ @ 50 ppm) which is 8.61 % higher than control whereas at 60 & 90 DAT, maximum per cent increase in plant height were obtained with T7 (GA₃ @ 200 ppm) i.e. 27.65 % and 28.03 % respectively. This finding is according to, [15] reported in rice and in mustard [17]. GA₃ is one important plant hormone that regulates various processes of plant growth and development, which is particularly important in stem elongation. GA₃ application, the height was increased significantly under salinity condition. [12]

Plant tiller

Seed soaking and foliar application enhanced tillers production plant⁻¹ as compared to control and varies with growth stages. As effectiveness of mode of application of PGRs response of foliar application on plant height was found more pronounced than seed soaking treatment. At 30 DAT, maximum tillers plant⁻¹ was recorded with treatment T3 (seed soaking 50 ppm GA₃) which is 23.77%, whereas at 60 & 90 DAT, maximum per cent increase in tillers plant⁻¹ were obtained with T7 (GA₃ @ 200ppm) i.e. 34.99% and 31.10% respectively. This finding is in accordance with the finding of [14], [15]. Plant hormones play an important role in regulating the tiller occurrence [8].

Plant dry weight

The maximum plant dry weight was recorded with treatment T3 (seed soaking @ 50ppm GA₃) at 30DAT, which is 43.08% higher over control and at 60 & 90 DAT, maximum per cent increase in plant dry weight were obtained with GA₃ (GA₃@200ppm) i.e. 18.81 % & 30.82 % at 60 & 90DAT respectively. Similar observation was also reported by [14],[17],[6],[8]. The increase in plant dry weight due to foliar spray GA₃ indicated that the photosynthetic activity and efficiency of the leaves have been increased which contributed to dry matter production. [10], [17][13].

Relative water content

All the treatments have positive significant effect on relative water content (RWC%) of rice grown under sodic condition. The effect of spraying of PGRs on leaves, RWC was found to be more obvious as the effectiveness of the method use of PGRs (GA₃ and SA). At 30 DAT seed soaking treatments, were found more effective than control. Maximum relative water content was obtained with seed soaking with GA₃ 50 ppm 70.50% at par with GA₃ @ 25 ppm while 60 and 90 DAT, maximum percent increase in relative water content was achieved with foliar application of GA₃ @ 200 ppm 13.57% and 9 6.44% respectively. Similar observations were also made by [10] [14] RWC tends to decline when transpiration exceeds water absorption under drought condition leading to decrease in cell turgor. Maintenance of high RWC under drought due to relatively more growth of the roots than shoots and/or abscisic acid induced reduction in stomata opening tends to maintain cell turgidity, chlorophyll content and photosynthesis [16].

Bio-chemical studies

Chlorophyll content - It is clear from data presented that activating effectiveness of PGRs is phasic events which regularize the plant growth and development. High salt concentration degrades the chlorophyll content of the plant at all growth stage. Exogenous application of PGRs either seed soaking or foliar application mitigates the adverse effect of salt stress. Sharp decline of chlorophyll content at 60 DAT was noticed in all treatments. Extent of reduction in chlorophyll content at 60 DAT was observed in foliar application of GA₃ @ 200 ppm followed by GA₃ 100 ppm and SA @ 200 ppm in comparison to other treatments. This finding is according to [14][10][18]. This could be predicated as the ability of varieties to maintain higher chlorophyll content and the optimistic response to GA₃. Under sodic condition limited water availability (physiological drought) usually causes a reduction in chlorophyll content. Being positively correlated with yield relatively high chlorophyll content may contribute to the plant productivity under stress condition ([16],[12] [4]).

Total soluble sugar

All treatments recorded rise total soluble sugars as compared to control at varies growth stages . As effectiveness of mode of application of PGRs response of foliar application on soluble sugars was found more pronounced that seed soaking treatment maximum total soluble sugars was recorded with treatment T3 (seed soaking 50ppm GA₃) which is 8.66% higher than control. Whereas at 60& 90 DAT, maximum per cent increase in total soluble sugars were obtained with T7 (GA₃ @ 200 ppm) *i.e.* 22.26% and 19.39% respectively. Similarly [14], [10] reported that- foliar application of GA₃ @ 200 ppm had significantly increase level sugar content.

Conclusions:

This experiment can be concluded that foliar application of PGRs were found more effect method than seed soaking. Seed priming treatment in nursery did not show longer effect to counter act the adverse effect of sodicity on plant growth of rice. Whereas, foliar application of various concentration of GA₃ and SA have significantly improve the crop yield. Among the treatments GA₃@ 200 ppm (foliar application) was found most effective concentration to improve plant growth and yield. Its needs further variation at farmer field.

Table No 1. Modulating effect of PGRs on Growth traits in rice (*Oryza sativa L.*) undersalt stress

	Treatments	Plant height			Number of tillers Plant-1			Dry biomass hill-1 (g)			Relative water content (RWC %)		
		30 DAT	60 DAT	90 DAT & Harvesting	30 DAT	60 DAT	90 DAT & Harvesting	30 DAT	60 DAT	90 DAT & Harvesting	30 DAT	60 DAT	90 DAT & Harvesting
T ₁	Control	51.20	70.62	71.86	6.86	7.06	7.53	2.84	10.79	21.96	69.27	71.47	68.94
T ₂	GA3 @25ppm seed soak	54.09	74.33	76.26	8.46	8.60	8.80	4.49	12.86	24.38	70.27	74.69	71.59
T ₃	GA3 @ 50ppm seed soak	54.93	75.66	77.96	9.00	9.23	9.40	4.99	13.82	24.98	70.50	75.42	72.13
T ₄	SA @10ppm seed soak	52.80	71.60	72.96	7.46	7.80	8.06	3.21	11.57	22.35	69.59	72.67	70.00
T ₅	SA @25ppm seed soak	54.30	73.66	74.73	8.20	8.56	9.13	3.91	12.27	23.51	69.87	73.61	71.17
T ₆	GA3 @ 100ppm FS at 30 DAT	50.70	93.13	95.80	6.26	10.46	10.53	2.24	17.31	28.94	69.34	80.17	75.08
T ₇	GA3 @ 200ppm FS at 30 DAT	50.76	97.06	99.86	6.40	10.86	10.93	2.74	18.81	30.82	69.52	82.70	75.96
T ₈	SA @ 100ppm FS at 30 DAT	50.13	82.26	90.80	7.06	9.46	9.76	2.54	15.21	25.96	69.46	76.83	73.34
T ₉	SA @ 200ppm FS at 30 DAT	50.40	88.66	92.13	6.73	9.80	9.90	2.62	16.15	27.95	69.36	78.29	73.94
	SEm±	0.46	0.78	1.14	0.51	0.27	0.22	0.14	0.33	0.44	0.22	0.30	0.22
	CD at 5%	1.37	2.32	3.41	1.56	0.80	0.67	0.41	1.00	1.31	0.65	0.91	0.66

SA:- Salicylic Acid

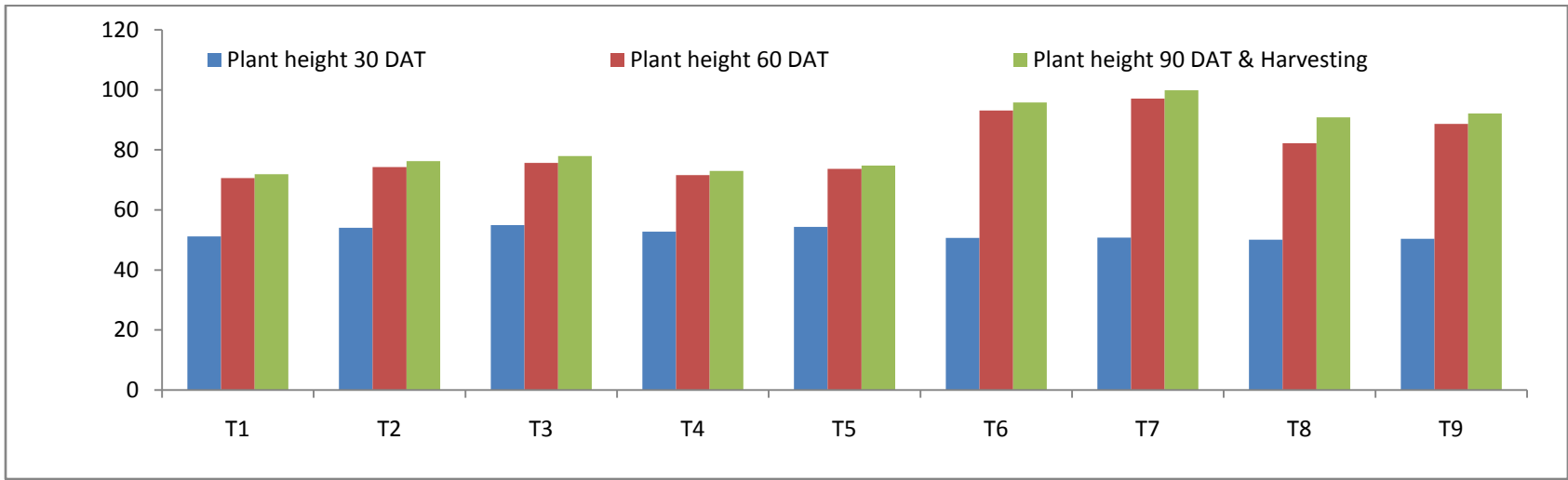
FS:-Foliar spray

Table No 2- Modulating effect of PGRs on Biochemical traits in rice (*Oryza sativa L.*)undersalt stress

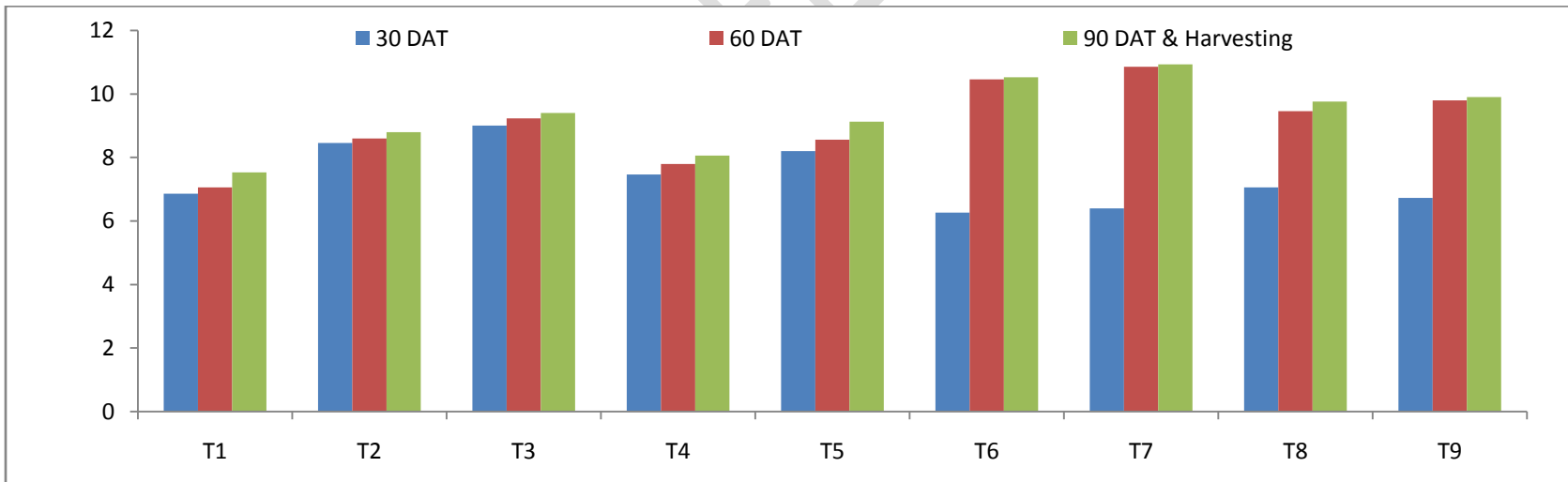
	Treatments	Chlorophyll content (SPAD Value)			Total soluble Sugar		
		30 DAT	60 DAT	90DAT & Harvesting	30 DAT	60 DAT	90 DAT & Harvesting
T ₁	Control	10.35	11.37	6.22	77.95	105.22	91.90
T ₂	GA3 @25ppm seed soak	11.27	13.02	7.55	84.44	120.19	105.26
T ₃	GA3 @ 50ppm seed soak	11.45	13.37	7.50	85.31	125.27	105.29
T ₄	SA @10ppm seed soak	10.49	12.27	6.77	82.68	110.41	101.66
T ₅	SA @25ppm seed soak	11.05	12.84	7.35	83.28	121.94	101.83
T ₆	GA3 @ 100ppm FS at 30 DAT	10.37	14.59	8.03	77.46	132.02	111.32
T ₇	GA3 @ 200ppm FS at 30 DAT	10.53	15.38	8.25	78.43	135.53	114.01
T ₈	SA @ 100ppm FS at 30 DAT	10.19	13.79	7.72	78.73	127.35	108.98
T ₉	SA @ 200ppm FS at 30 DAT	10.25	14.17	7.90	78.01	130.13	109.87
	SEm±	0.19	0.10	0.12	0.54	0.65	0.63
	CD at 5%	0.58	0.29	0.35	1.61	1.94	1.89

SA:- Salicylic Acid

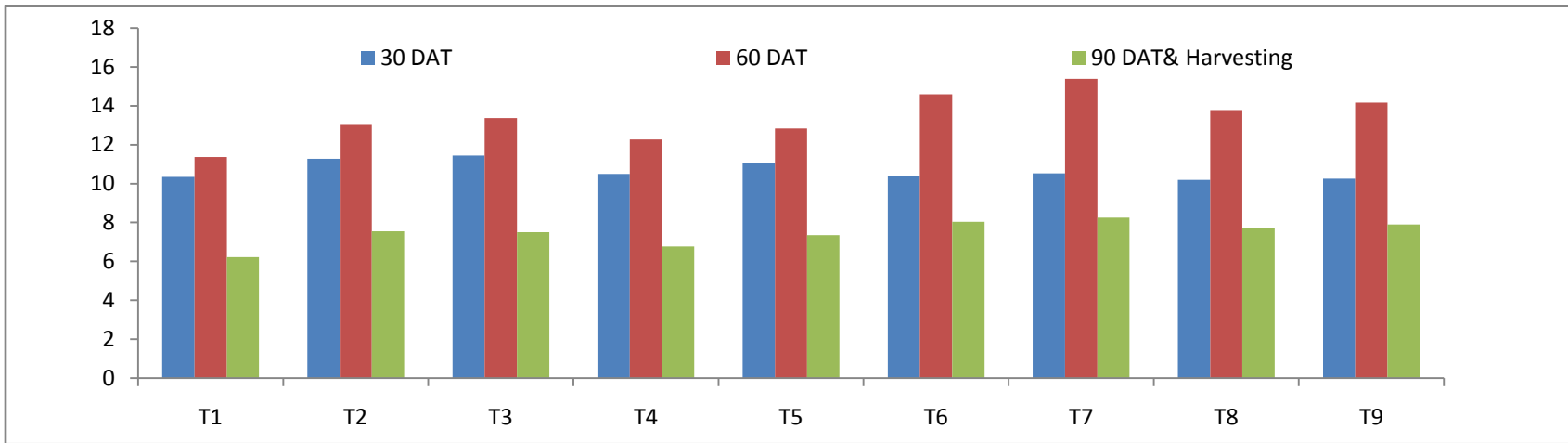
FS:- Foliar spray



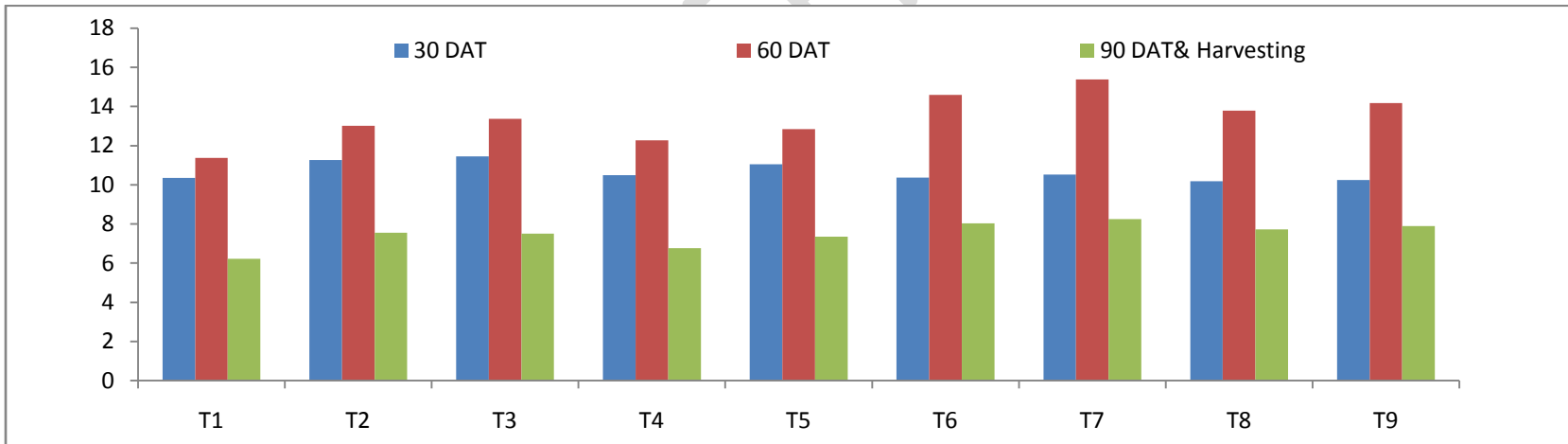
Plant height



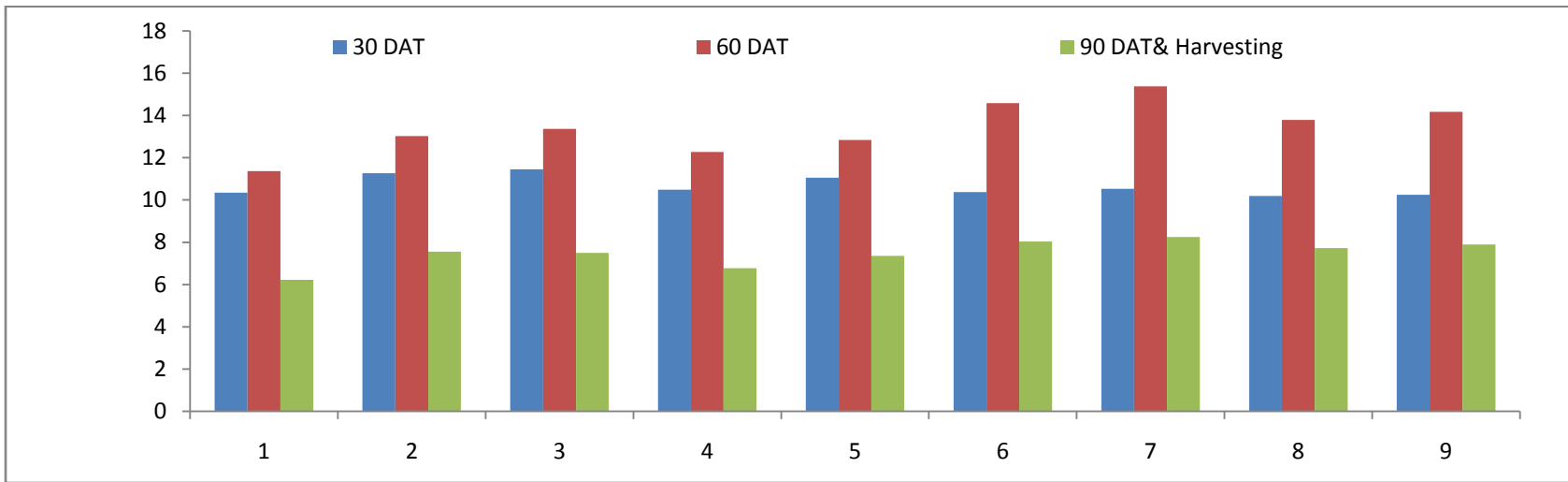
Number of tillers Plant⁻¹



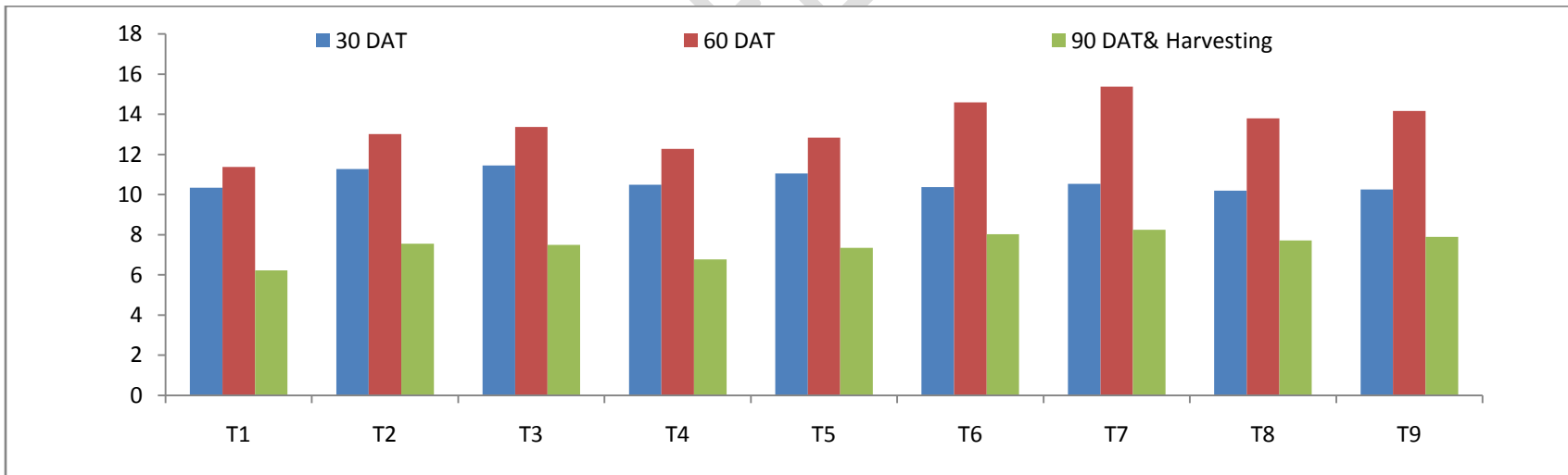
Dry biomass hill⁻¹(g)



Relative water content (RWC %)



Chlorophyll content (SPAD Value)



Total soluble Sugar

References

1. **Afzal I, Shahzad MA, Basra, Ahmad N, Cheema MA, Warraich EA, and Khaliq A**, Effect of Priming and Growth regulator treatments on emergence and seedling growth of Hybrid Maize (*Zea mays* L.). *IJAB*.**2002**; 303–306
2. **Ansari O, and Zadeh FS**, Improve Mountain Rye (*Secale montanum*) seeds Germination and Seedlings Growth under Cold Stress. *IRJBS*.**2012**;3 (8): 1651-1657,
3. Bodie AR, Micciche AC, Atungulu GG, ROthrock MJ, Current Trends of Rice Milling Byproducts for Agriculture Applications and Alternative Food Production Systems . 2019 (3) <https://doi.org/10.3389/fsufs.2019.00047>
4. **Chauhan A, Rajput N, Kumar A, Verma, JS, and Chaudhry, AK**, “Interactive effects of gibberellic acid and salt stress on growth parameters and chlorophyll content in oat cultivars” *JEB*.**2018**; (39) 639-646.
5. **Chauhan A, Bassam A, Amarah A, Kumar A, Verma JS, Ghramh HA, Khan KA, and Ansari MJ**, “Influence of gibberellic acid and different salt concentrations on germination percentage and physiological parameters of oat cultivars” *SJBS*.**2019**;(26) 1298–1304
6. **Dilip, WS, Singh D, Moharana D, Rout S, and Patra SS**, Effect of Gibberellic acid (GA₃) Different concentrations at Different Time Intervals on Seed Germination and seedling Growth of Rangpur Lime. *JANR*.**2017**;157-165
7. **Dwivedi, J.L. (2015)**. “*Rice in Uttar Pradesh*”
8. **Islam. S, Chakraborty, S, Uddin, H, Mehraj. and Uddin, A.F.M .J, (2014)**. Growth and yield of wheat as influenced by GA₃ concentrations. *IJBSSR*(**2**): 74-78
9. **Jini D, Joseph B**, .Physiological Mechanism of Salicylic Acid for Alleviation of Salt Stress in Rice. *Rice Sci.*. **2017**;**24**(2):97–108
10. **Kalpana, Khan AH, Singh AK, Maurya KN, Mubeen, Yadava RK, Singh U. and Gautam AR**, Effect of Different Seed Priming Treatments on Germination, Growth, Biochemical Changes and Yield of Wheat Varieties under Sodic Soil. *IJSR*.**2013**; 2319-7064
11. **Khan AH, Singh AK, Mubeen, Singh U, Yadav RK, Pandey AK, Srivastava AK, Singh S, Singh US, Singh SP, and Ismail AM** “Approaches for Boosting Rice Yield Under Sodic Soil Condition in Gangetic Alluvium of Eastern Uttar Pradesh” .*IJAPSA* Volume.**2016**; 02(**06**):89-103.
12. **Misratia KM, Hakim MA, Musa MH, and Puteh A**, “Effect of salinity and alleviating role of gibberellic acid (GA₃) for improving the morphological, physiological and yield traits of rice varieties” *AJCS*;**2013**.7(11):1682-1692
13. **Pandey G, Nautiyal MK, Chauhan P, and Kandpal G**, “Assessment of appropriate dosage of gibberellic acid (GA₃) for better seed yield of a promising hybrid rice variety” *IJCS* **2017**; 5(4): 1603-1607
14. **Pratap M, Saini PK, Tripathi SK, Yadav RK, Kumar M, and Yadav V**, Effect of Foliar Application of PGRs on Growth and Biochemical Changes of Rice (*Oryza sativa* L.) under Salt Stress Condition” *IJCMAS*. **2014**; 286-291
15. **Ramesh S, Sudhakar P, Elankavi S, Suseendran K, and Jawahar S**, “Effect of gibberellic acid (GA₃) on growth and yield of Rice (*Oryza sativa* L.)” *Plant Archives*.**2019**;Vol. 19(1), pp.1369-1372
16. **Rao SR, Qayyum A, Razzaq A, Ahmad M, Mahmood I, and Sher A**,. Role Of Foliar Application Of Salicylic Acid And L-Tryptophan In Drought Tolerance Of Maize. *TJAPS*, **2012**;22(**3**): 768-772
17. **Saini PK, Yadav RK, and Pratap M**,. Effect of Foliar Application of GA₃, on Yield and Quality of Indian Mustard [*Brassica juncea* (L.) Czern. & Coss.] Under Sodic Soil. *IJCMAS*.**2017**:4156-4159
18. **Shahi S, Yadav RK, Kushwaha SP, Singh M, Singh P.**, Effect of Plant Growth Regulators on Biochemical Studies of Mustard [*Brassica juncea* (L.) Czern & Coss.] under Sodic Soil. *IJPSS*.87074 ISSN: 2320-7035
19. **Yamaguchi S and Kamiya Y.**, Gibberellin Biosynthesis: Its Regulation by Endogenous and Environmental Signals *Plant Cell Physiol.* (2000);41(3): 251-257