

Original Research Article

Effect of Relative Water Content in Rice Genotypes after Gypsum & Bio-compost amended under Sodic Soil

Comment [R1]: Can you change to Effect of Gypsum & Bio-compost applications on Relative Water Content in Rice Genotypes under Sodic Soil

Abstract

Sodic soils have immense productivity potential, if managed through proper technology interventions. Bio-compost (prepared by composting pressmud a sugar industry byproduct received from cane juice filtration) and spent wash received from distilleries through microbial aerobic decomposition) and gypsum (received from waste material of mining) can be used to reclaim sodic soils. In this study, field experiments were conducted during the Kharif season of 2018 and 2019 at the ICAR - Indian Agricultural Research Institute, Sub Regional Station, Pusa (Samastipur), Bihar. The promising results revealed that the mean leaf relative water content at pre-flowering stage in the salt-tolerant genotypes ranged from 69.47 % to 82.20 % during 2018 and 69.52 % to 82.24 % during 2019. The mean leaf relative water content at grain filling stage in all the genotypes varied between 77.55 % to 85.45 % during 2018 and 75.49 % to 85.16 % during 2019. Soil amendments and genotypes interaction was found significantly in both the years at grain filling stage.

Comment [R2]: Is that what you mean?

Comment [R3]: State months

Comment [R4]: Add a few words about the experiment itself

Keywords: Gypsum, Bio-compost, Rice genotypes, Sodic soils, Relative water content.

Comment [R5]: Leaf relative water content

Introduction

Salt affected soil is a major abiotic stress limiting plant growth and development. In crops known as glycophyte or salt susceptible [Hasegawa *et al.* 2000, Qadir *et al.* 2007], it causes yield losses by depressing the uptake of water, and disturbing mineral and normal metabolism. Salt-affected soils are identified by excessive levels of water-soluble salts, especially sodium chloride (NaCl) [Tanji 2002]. NaCl is a small molecule which when ionized by water, produces sodium (Na^+) and chloride (Cl^-) ions. Excess Na^+ in plant cells directly damages membrane systems and organelles, resulting in growth reduction and abnormal development prior to plant death. The toxic ions cause ionic and osmotic stress at the cellular level in higher plants, especially in susceptible germplasm [Mansour and Salama 2000, Chinnusamy 2005]. Salinity reduces plant growth through osmotic effects and reduces the water uptake, thereby causing a reduction in growth.

Comment [R6]: Salinity in soil is – please rephrase

Comment [R7]: Define more accurately

Gypsum and pyrite are the most effective reclamation agents for sodic soils and are received from mining so they are expensive and beyond the reach of poor farmers in rainfed lowland areas. Pressmud, a sugar industry by-product, is readily available in Bihar and Uttar Pradesh (U.P.) and less expensive compared to gypsum. Bio-compost is prepared by composting pressmud (received from cane juice filtration) and spent wash (received from distilleries) through microbial aerobic decomposition. It contains nutrients like N, P, K, Zn and large amounts of organic carbon. Calcium replaces Na^+ from the cation exchange complex, and about 2% - 3% sulphur converts into sulphuric acid and lowers soil pH. In addition, it contains bio-agents like Trichoderma and Azotobacter which protect plants from

Comment [R8]: I recommend this link for rephrasing
https://www.researchgate.net/publication/279507515_How_Salinity_Damages_Citrus_Osmotic_Effects_and_Specific_Ion_Toxicities

Comment [R9]: you are talking about gypsum? Please clarify

several fungal pathogens, enhance growth and development through robust root formation, and enhance soil N availability through atmospheric N₂ fixation. Composition of gypsum and biocompost are shown in Table-1 and Table-2, respectively.

Recently released rice varieties in India, including CSR 30, CSR 63, CSR-27, Suwasini, Rajendra Bhagwati, Boro-3, Rajendra Neelam, CR-3884-244-8-5-6-1-1, CR-2851-SB-1-2-B-1 and Pusa-44, have shown great promise for cultivation in sodic/saline soils of Bihar. In addition, IRR1 made considerable progress in developing a Marker Assisted Backcrossing (MABC) system for the major QTL Saltol, associated with salinity tolerance in rice. Through MABC, this locus is now introgressed into three popular varieties (BR11, BRRI dhan 28, and IR64). Trials conducted under field conditions showed that introgression of this QTL significantly improved the salt tolerance of these varieties, and seeds of these three varieties were now ready for testing in farmers' fields. The availability of these salt-tolerant varieties provides a great opportunity for increasing and stabilizing productivity in salt-affected areas. Particularly when combined with best management practices specific for salt-affected areas, salt tolerant rice varieties could become a great opportunity for improving productivity and soil quality of saline and sodic soils. Considering this background, we conducted experiments to evaluate the benefits of combining bio-compost, gypsum and salt tolerant varieties together to harness the sodic soil potential and increase water uptake by rice crop for metabolic activity.

Comment [R10]: potentials

Comment [R11]: what does it stand for?

Comment [R12]: Please chose a better word

Materials and methods

A field experiments were carried out during 23th June 2018 to 28th November 2018 and 23th June 2019 to 28th November 2019 (two *kharif* seasons). The experiment was conducted at the Indian Agricultural Research Institute, Sub Regional Station, Pusa (Samastipur), Bihar, which lies at 85° 40' 19.7" E latitude 25° 59' 06.2" N longitudes at an elevation of 55.00 meter above mean sea level. The experimental site is hot and humid in summers and extremely cold in winter with average rainfall of 1344 mm of which 70% received during the monsoon period (mid June - mid September, 2018 and 2019).

A field experiment laid out in split plot design with four treatment T₁- Control, T₂- Gypsum @ 100% G.R., T₃- Gypsum @ 50% G.R. + Biocompost @ 2.5 t ha⁻¹, T₄- Biocompost @ 5.0 t ha⁻¹ in main plots and ten genotypes G₁ - Suwasini, G₂ - Rajendra Bhagwati, G₃ - Boro-3, G₄ - Rajendra Neelam, G₅ - CSR-30, G₆ - CSR-36, G₇ - CR-3884-244-8-5-6-1-1, G₈ - CR-2851-SB-1-2-B-1, G₉ - CSR-27, G₁₀ - Pusa-44 in sub plots and replicated in thrice. The main plots and sub plots are permanent plots for both the years (2018 and 2019). Initial representative soil samples were analyzed and accordingly gypsum requirement and organic carbon has been calculated for application in soil. Inorganic and organic amendment applied only first year. After incorporation of inorganic and organic amendments in soil, each plot was little irrigated so that gypsum get dissolved and leaching of gypsum from upper layer to lower layer of soil will take place. Then, field was left for 8-10 days for gypsum leaching of gypsum before rice transplanting. After 8-10 days for transplanted rice, seedlings of different genotypes i.e. Suwasini, Rajendra Bhagwati, Boro-3, Rajendra Neelam, CSR-30, CSR-36, CR-3884-244-8-5-6-1-1, CR-2851-SB-1-2-B-1, CSR-27, Pusa-44 were raised using a seed rate of 30 kg ha⁻¹ and 25 days old seedling were transplanted manually.

Comment [R13]: @ What do you mean?

Comment [R14]: Indicate that you mean gypsum requirements

Transplanted rice genotypes were taken with the recommended dose of N: P₂O₅: K₂O @ 120: 60: 40 in the form of urea, diammonium phosphate (DAP) and muriate of potash (MOP). Fifty per cent of N, and full doses of P₂O₅ and K₂O were applied as basal and the rest fifty per cent of N was applied in two splits at 30 days interval.

Comment [R15]: You mean treated?

Comment [R16]: rephrase

Table-1: Gypsum Composition

Comment [R17]: refer to tables in text

S.No.	Properties	Value
1	Ca (%)	29.2%
2	S (%)	18.6%

Table-2: Biocompost Composition

S.No.	Properties	Value
1	Moisture Content	38%
2	pH	7.68
3	EC (dS m ⁻¹)	12
4	Organic Carbon (%)	24.20%
5	Organic Matter (%)	42.11%
6	C : N ratio	13.5%
7	Available Nitrogen (%)	1.80%
8	Available Phosphorous (P ₂ O ₅) (%)	1.72%
9	Available Potassium (K ₂ O) (%)	1.49%
10	Calcium (%)	3.2%
11	Magnesium (%)	1.1%
12	Available Sulphur (%)	1.3%
13	Available Zn (mg kg ⁻¹)	30.89
14	Available Cu (mg kg ⁻¹)	14.21
15	Available Fe (mg kg ⁻¹)	123.53
16	Available Mn (mg kg ⁻¹)	64.29

Leaf relative water content

Fully expanded youngest leaves were selected from different plants. Ten leaves were sampled and weighed immediately to determine the fresh weight (FW) and afterwards it immersed in distilled water in Petri dishes for 4 hour's in darkness and then turgid weight (TW) was determined. The leaves were dried in an oven at 68°C for 24 hour and the dry weight (DW) was taken. Afterwards RWC was calculated as by using the methodology which was given by Whetherley *et al.* (1950).

$$\text{Relative Water Content (RWC) (\%)} = \frac{(\text{FW} - \text{DW})}{(\text{TW} - \text{DW})} \times 100$$

Statistical analysis

The data recorded for different parameters were analyzed with the help of analysis of variance (ANOVA) technique (Gomez and Gomez, 1984) for split plot design. ANOVA was found significant and accordingly results are presented at 5% level of significance (P=0.05).

Results and discussion

Physico-chemical properties of experimental soil

The soil of the experimental site belongs to ~~the order-Entisol~~ order and characterized by silt loam ~~in~~ texture at surface containing 10.45% sand, 72.06% silt and 17.49% clay. ~~The~~ physico-chemical properties of soil ~~were~~ alkaline of pH 9.69 ~~in reaction, with~~ electrical conductivity of 2.12 dS m⁻¹ and organic carbon 2.6 g kg⁻¹. The soil ~~had~~ available N, P, K and S ~~was recorded~~ were 136.8 kg ha⁻¹, 7.83 kg ha⁻¹, 93.2 kg ha⁻¹ and 3.53 kg ha⁻¹ respectively (Table 3). High pH and low EC of the experimental site might be from excessive accumulation of exchangeable Na⁺ in the soil particles. This indicates that the soil of the experimental site was sodic (USDA 1954). The soil had very low organic carbon content indicating moderate potential of the soil to supply nitrogen to plants through mineralization of organic carbon. ~~Soils in salt-affected landscapes produce less biomass than non-saline soils resulting less in soil organic carbon~~ (Wong *et al.* 2010).

Comment [R18]: rephrase

Table 3: Physico-chemical properties of experimental soil (0-15 cm depth before starting of the experiment)

Properties	Value
Physical properties	
Sand (%)	10.45
Silt (%)	72.06
Clay (%)	17.49
Textural Class	Silt loam
Bulk density(g cm ⁻³)	1.63
Water Holding Capacity (%)	38.62
Wet Aggregate Stability (%)	8.45
Chemical properties	
pH (1:2 Soil : Water) (0 -15 cm depth)	9.69
EC (dS m ⁻¹)	2.12
Organic Carbon (g kg ⁻¹ soil)	2.6
Available Nitrogen (kg ha ⁻¹)	136.8
Available Phosphorous (P ₂ O ₅) (kg ha ⁻¹)	7.83
Available Potassium (K ₂ O) (kg ha ⁻¹)	93.2
Available Sulphur (kg ha ⁻¹)	3.53

Plant water status at pre-flowering stage

It was observed that ~~the~~ all genotypes had significantly higher than the Pusa-44 and Rajendra Bhagwati in both ~~the~~ years (~~found in~~ Table 4). The mean ~~of~~ leaf relative water content at pre-flowering stage in all the genotypes ranged from 69.47 % to 82.20 % during 2018 and 69.52 % to 82.24 % during 2019. All the soil amendments ~~plots~~ had significantly higher leaf relative water content at pre-flowering stage as compared to the control plot in the first year while in the second year it was significantly higher than the control plot and biocompost @ 5.0 t ha⁻¹ applications. The combination of gypsum @ 50% GR and biocompost @ 2.5 t ha⁻¹ had higher value than the other two amendments. However, gypsum @ 100% GR application had higher leaf relative water content at pre-flowering stage than the biocompost @ 5.0 t ha⁻¹ application in both ~~the~~ years.

Comment [R19]: higher in what? Please indicate

Leaf relative water content at pre-flowering stage ranged from 64.86 % to 86.61 % in the first year while in the second year it ranged from 66.83 % to 87.01 %. Amendment and genotype interaction was non-significant during 2018 and 2019.

It might be due to increase in osmotic pressure of cytoplasm which is accompanied by the synthesis of osmolytes which ultimately enhanced water flow into plant organs. Ca^{2+} helps in removal of excess sodium ion and biocompost increase water holding capacity.

Table 4: The influence of organic and inorganic amendments and their combination on leaf relative water content (RWC) (%) at pre-flowering stage in different rice genotypes.

Rice genotypes	2018					2019				
	Organic and inorganic amendments				Mean	Organic and inorganic amendments				Mean
	T ₁	T ₂	T ₃	T ₄		T ₁	T ₂	T ₃	T ₄	
G ₁	66.29	75.69	78.27	74.18	73.61	68.36	76.82	76.45	72.88	73.63
G ₂	66.77	72.59	75.09	72.89	71.84	70.18	73.77	73.20	69.07	71.55
G ₃	68.57	75.68	79.10	73.51	74.22	70.26	78.10	76.64	73.63	74.65
G ₄	70.81	72.80	75.84	73.45	73.23	72.50	74.22	73.19	71.14	72.76
G ₅	70.37	75.11	78.53	73.84	74.46	73.43	76.38	78.19	72.50	75.12
G ₆	74.35	77.68	81.64	77.31	77.75	73.00	81.60	78.10	75.69	77.10
G ₇	71.81	76.94	80.33	74.69	75.94	73.84	78.65	77.77	75.62	76.47
G ₈	70.78	75.21	80.23	75.33	75.39	69.47	76.73	76.45	72.32	73.75
G ₉	76.11	86.61	84.85	81.25	82.20	74.35	84.69	87.01	82.93	82.24
G ₁₀	64.86	69.83	72.73	70.46	69.47	67.40	72.54	71.30	66.83	69.52
Mean	70.07	75.81	78.66	74.69		71.28	77.35	76.83	73.26	
	T	G	T×G	G×T		T	G	T×G	G×T	
CD (P = 0.05)	3.308	2.938	NS	NS		3.269	3.525	NS	NS	
SE(m) ±	0.938	1.040	2.965	2.184		0.927	1.248	2.931	2.542	

Comment [R20]: please state that these are average of replicates or is it not? You should have anova results in a separate table. You calculated CD but you did not have enough comments on the effect of treatment and Variety.

This comment for all tables. Furthermore the effect of time .. you added applications only in first year did it has more or less effect.

Plant water status at grain filling stage

Leaf relative water content at grain filling stage in most of the genotypes were significantly higher than the varietal check Pusa-44 and Rajendra Bhagwati in the first year while in the second year all the genotypes were found significantly higher than the varietal check Pusa-44 (Table 5). The mean of leaf relative water content at grain filling stage in all the genotypes varied between 77.55 % to 85.45 % during 2018 and 75.49 % to 85.16 % during 2019. All the soil amendments plots had significantly higher leaf relative water content at grain filling stage as compared to the control plot in the first year while in the second year it was significantly higher than the control plot and biocompost @ 5.0 t ha⁻¹ applications. The combination of gypsum @ 50% GR and biocompost @ 2.5 t ha⁻¹ had higher value than the other two amendments. However, gypsum @ 100% GR application had higher leaf relative water content at grain filling stage than the biocompost @ 5.0 t ha⁻¹ application.

Soil amendments and genotypes interaction was significant in both the years. Leaf relative water content at grain filling stage varied from 68.68 % to 86.53 % in the first year while in the second year it varied from 65.50 % to 87.29 %. Without application of any amendment all the varieties were found superior ~~of to~~ Pusa-44 and Rajendra Neelam in both the years. The response of gypsum, biocompost and their combination varied between 78.80 % to 86.53 %, 76.36 % to 85.52 % and 81.09 % to 86.43 % in the first year, respectively; while in the second year it was varied between 80.76 % to 85.57 %, 72.04 % to 84.70 % and 79.21 % to 87.29 %, respectively.

Comment [R21]: what is the difference from the previous data

The ~~h~~High salt concentration in the root zone, which causes osmotic stress, restricts water absorption by the plants and causes cellular dehydration, seems to be primarily responsible for decrease in RWC. Furthermore, ~~S~~sodicity induced membrane damage and caused RWC reduction ~~in RWC~~ in leaves. Salt stressed plants exhibit damage of lipid membranes which often results in increased cell permeability and electrolyte leakage from cells. Almost similar results were also reported by Singh *et al.* (2015); Kumar *et al.* (2016) and Taffouo *et al.* (2017).

Table 5: The influence of organic and inorganic amendments and their combination on leaf relative water content (RWC) (%) at grain filling stage in different rice genotypes.

Rice genotypes	2018					2019				
	Organic and inorganic amendments				Mean	Organic and inorganic amendments				Mean
	T ₁	T ₂	T ₃	T ₄		T ₁	T ₂	T ₃	T ₄	
G ₁	78.86	82.90	84.50	80.66	81.73	78.90	84.37	87.29	81.77	83.08
G ₂	78.59	78.80	81.09	80.51	79.75	78.59	80.76	79.21	78.63	79.30
G ₃	80.74	81.39	86.43	82.43	82.75	80.80	83.51	86.50	81.27	83.02
G ₄	70.44	83.92	85.14	83.08	80.65	71.78	83.90	85.34	81.99	80.75
G ₅	81.22	83.92	84.78	83.13	83.26	81.93	84.37	85.70	83.61	83.90
G ₆	80.56	84.76	85.83	84.19	83.83	83.64	84.42	87.29	83.10	84.61
G ₇	82.04	84.70	85.37	84.94	84.26	82.29	85.20	84.95	84.02	84.12
G ₈	82.35	83.34	84.81	83.91	83.60	80.70	84.60	84.91	83.11	83.33
G ₉	83.87	86.53	85.86	85.52	85.45	83.83	85.57	86.55	84.70	85.16
G ₁₀	68.68	79.21	85.96	76.36	77.55	65.50	83.02	81.41	72.04	75.49
Mean	78.74	82.95	84.98	82.47		78.80	83.97	84.92	81.42	
	T	G	T×G	G×T		T	G	T×G	G×T	
CD (P = 0.05)	1.781	2.322	4.753	4.740		3.038	2.924	6.082	6.301	
SE(m) ±	0.505	0.822	1.596	1.639		0.861	1.035	2.723	2.144	

CONCLUSION

Relative water content at pre-flowering stage and grain filling stage ~~had was~~ significantly higher in genotypes CSR-27 followed by CSR-36 and CR-3884-244-8-5-6-1-1. Moreover, ~~and~~ combination of gypsum @ 50% G.R. and biocompost @ 2.5 t ha⁻¹ application had significantly higher RWC followed by gypsum application @ 100% G.R..

REFERENCES

- Gomez, K.A. and Gomez, A.A., 1984. Statistical Procedure for Agricultural Research, 2nd Edition. An International Rice Research Institute Book. A Wiley-Inter-science. Publication, John Wiley and Sons, New York.
- Tanji, K.K., 2002. Salinity in the Soil Environment, In: A. Lauchli and U. Luttge, Eds., Salinity Environment-Plant- Molecules. *Kluwer Academic, Dordrecht*. pp. 21-51.
- Kumar, A., Lata, C., Kumar, P., Devi, R., Singh, K., Krishnamurthy, S.L., Kulshreshtha, N., Yadav, R.K. and Sharma, S.K., 2016. Salinity and drought induced changes in gas exchange attributes and chlorophyll fluorescence characteristics of rice (*Oryza sativa*) varieties. *Indian Journal of Agricultural Sciences*, **86**(6), pp.718-726.
- Mansour, M.M.F. and Salama, K.H.A., 2004. Cellular Basis of Salinity Tolerance in Plants, *Environmental and Experimental of Botany*, Vol. **52**, pp. 113-122. <http://dx.doi.org/10.1016/j.envexpbot.2004.01.009>
- Qadir, M., Oster, J.D., Schubert, S., Noble, A.D. and Sahrawat, K.L., 2007. Phytoremediation of Sodic and Saline-Sodic Soils. *Advances in Agronomy*, Vol. **96**, pp. 197- 247. [http://dx.doi.org/10.1016/S0065-2113\(07\)96006-X](http://dx.doi.org/10.1016/S0065-2113(07)96006-X)
- Hasegawa, P.M., Bressan, R.A., Zhu, J.K. and Bohnert, H.J., 2000. Plant Cellular and Molecular Responses to High Salinity. *Annual Review of Plant Physiology and Molecular Biology*, Vol. **51**, pp. 463-499. <http://dx.doi.org/10.1146/annurev.arplant.51.1.463>
- Singh, A., Sharma, P.C., Kumar, A., Meena, M.D. and Sharma, D.K., 2015. Salinity induced changes in chlorophyll pigments and ionic relations in bael (*Aegle marmelos* Correa) cultivars. *Journal of Soil Salinity and Water Quality*, **7**(1), pp.40-44.
- Taffouo, V.D., Nouck, A.E., Nyemene, K.P., Tonfack, B., Meguekam, T.L. and Youmbi, E., 2017. Effects of salt stress on plant growth, nutrient partitioning, chlorophyll content, leaf relative water content, accumulation of osmolytes and antioxidant compounds in pepper (*Capsicum annum* L.) cultivars. *Notulae Botanicae Horti Agrobotanici Cluj-Napoca*, **45**(2), pp.481-490.
- USDA. 1954. Diagnosis and Improvement of Saline and Alkali soils. United States Salinity Laboratory staff. Agriculture Handbook No. 60, USDA, US. Govt Printing Office, Washington DC, pp.26.
- Chinnusamy, V., Jagendorf, A. and Zhu, J.K., 2005. Understanding and Improving Salt Tolerance in Plants. *Crop Science*, Vol. **45**, pp. 437-448. <http://dx.doi.org/10.2135/cropsci2005.0437>
- Whetherley, P.E., 1950. Studies in the water relation of cotton plants. I. The field measurement of water deficit in leaves. *New Phytologist*, **49**, pp. 81-87.
- Wong, V.N., Greene, R.S.B., Dalal, R.C. and Murphy, B.W., 2010. Soil carbon dynamics in saline and sodic soils: a review. *Soil use and management*, **26**(1), pp. 2-11.

UNDER PEER REVIEW