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A Study of the Effect of Pyrites and Rhizobium Inoculation on Chlorophyll and Sugar Content in Black Gram under Sodicity Stress Condition.

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Abstract- **The study reveals that sodic conditions of soil resulted in poor bio-synthesis in black gram plants and hence both chlorophyll'a', 'b' and reducing and non-reducing sugar contentsdecreased with increasing levels of RSC water treatments. Incorporation of iron pyrites as an ameliorating agent significantly improved the availability of nutrients and enhanced the chlorophyll and sugar content. The Rhizobium inoculated treatments supplemented with 50 g iron pyrites pot-1 adjusted best among all the treatments and plants could withstand irrigation with 2.5 meL-1 RSC water without being adversely affected. Further, 7.5 meL¬-1 proved detrimental for plant growth and recorded maximum reduction in sugar and chlorophyll contents in the treatments which were not supplied with pyrites.**

***Key words:*--RSC water, black gram, iron pyrites, reducing and non-reducing sugar, chlorophyll.**

I. INTRODUCTION

Calcareous soils have resulted in precipitation of $CaCO_3$ from water of some rivers which contain high amount of $CaH(CO_3)_2$. The tropical aridity creats reaction resulting in accumulation of salts. Most of the water of uplands is lost due to runoff and transpiration reducing effective leaching. Precipitation of calcium increases sodium concentration which is further augmented by high RSC of the available water resources. Salinity is the major yield reducing factor in legume cultivation as legumes are salt sensitive crop, especially in the seedling stages where they are most susceptible to damage due to water stress and ion-imbalance. In saline soils, many environmental factors such as soil pH, water deficiency and nutrient deficiency are reported by James et al.[1] and Kopittke and Menzius[2]. Moradi and Ismail[3] reported that the seedling stage suffers most damage due to ion-imbalance and thus, other physiological processes are negatively affected during the growth of the plant especially photosynthesis. Cramer and Nowak[4] reported that during long term exposure to salinity, plants experience premature senescence of adult leaves and root nodules which reduce the net photosynthetic area available to support continued growth of the plant. Thus, bio-synthesis of sugar and nitrogen fixation are negatively affected. In order to overcome this environmental abuse and to raise the plants with better suitability towards changing environmental inputs, various physico-chemical and bio-chemical tools are employed. One such agent that helps in reclamation of saline soil is the use of iron pyrites. Sharma and Manchanda[5] and Singh et al.[6] have suggested that iron pyrites is a good reclaiming agent for calcareous alkali soils.

Application of pyrites has no adverse effects on the soil bacterial and fungal population. Therefore, naturally occurring populations and inoculated strains of rhizobium are not affected. Volatilization losses from ammonium sulphate and urea is considerably reduced by application of pyrites. Rai et al.[7]reported that Pyrites application increases availability of phosphorus, iron and zinc in soil and hence, responses of cereals and pulses to pyrites application in calcareous soils are significant. Inoculation of seeds with rhizobium helps in increasing effective nodulation, especially in those soils that have low population of naturally occurring Rhizobia.

II. METHOD AND MATERIAL:

In order to study the relative effect of the RSC water and pyrite application on chlorophyll and sugar content, a pot experiment was laid down in the School of Chemical Sciences, Chemistry Dept., St. John's College, Agra, using factorial randomized block design and replicated thrice using black gram (*Vigna mungo L.*) C. V. Pant.U-19 as the test crop. The experiment consisted of four levels of RSC water viz. R_0(0 meL^{-1}), R_1(2.5 meL^{-1}), R_2(5.0 meL^{-1}) and R_3(7.5 meL^{-1}), three levels of iron pyrites viz.

S_0(0 gm pot^{-1}), S_1(25 gm pot^{-1}) and S_2(50 gm pot^{-1}) and two levels of Rhizobium inoculation, I_0(uninoculated) and I_1(inoculated). The essential nutrients were supplied through basal application in the soil before sowing the test crop by applying urea, single super phosphate and muriate of potash @ 20, 40 and 60 g pot^{-1}containing 10 kg of soil each. Pyrite was incorporated through basal application before sowing as treatment. Inoculation of seeds was done and dropped at depth of about 6cm equally in all the pots. RSC water was prepared by dissolving the sodium salts containing carbonate and bicarbonate in the best available water (bore well water). The pots were irrigated with RSC water after every 15 days. Control sets were irrigated with best available water only.

Plant material samples were collected from each pot after harvest after 45 DAS for chlorophyll 'a', 'b' and total chlorophyll analysis by Jayaraman's[8] method and after harvest for sugar analysis. Reducing sugar was estimated by method using dinitrosalicilic acid reagent method by Miller[9] and total sugar was estimated by Dubois et al. [10] phenol reagent method . Non-reducing sugar was calculated by subtracting reducing sugar from total sugar. The chlorophyll content was expressed as mg g^{-1} sugar contents were expressed as mg/ 100 g of sample. The results obtained were subjected to statistical analysis with the help of variance, using SIGMASTAT 3.5.

III. RESULT AND DISCUSSION:

It is revealed from the data in table 2 that chlorophyll 'a' and 'b' contents at 45 DAS significantly increased over control with the rise in pyrite application and rhizobium inoculation but decreased with increasing RSC levels. Highest value of chlorophyll 'a' was recorded at $R_1S_2I_1$ which was 3.63% and 3.03% higher over the control respectively for the two consecutive seasonswhereas the lowest chlorophyll 'a' content recorded at $R_3S_0I_1$ was 20.45% and 21.81% lower than the control respectively for both the seasons. And chlorophyll 'b' was recorded a highest at $R_1S_2I_1$ with values of 15.2% and 25% over the control and lowest values recorded at $R_3S_0I_1$ were28.4% and 25.4% respectively for both the seasons. Comparing the treatments, it is obvious that the application of pyrites increased the leaf chlorophyll due to sulphur availability. Similar results were reported by Sinha and Sakal[11].

The highest content of reducing and non-reducing sugar was reported in the treatment $R_1S_2I_1$. Reducing sugar was 1.51% and 2.41% higher over the control for the two consecutive seasons respectively and non-reducing sugar was 3.32% and 2.92% higher over control respectively for both the seasons. Reducing sugar was decreased to the extent of 6.04% and 10.26% and non-reducing sugar was reduced to the extent of 17.49% and 15.57% over the control respectively for the two seasons in the treatment $R_3S_0I_1$. Further, it was also seen that there was a decreasing trend in the values of the sugar content in the second year crop. The decrease in the values in the second year was probably due to the adsorption of the carbonate and bicarbonate ions on the soil surface due to continuous irrigation in the two consecutive years.

Due to the sodic ion toxicity, the physico-chemical properties of the soil as mentioned in table 1and the availability of soil nutrients is affected and the bio-synthesis of starch is reduced due to reduction in the rate of photosynthesis because it is closely related to chlorophyll content of the plant. Incorporation of pyrites as an ameliorating agent increases the sulphur content causing an increased accumulation of the sugar in pulse crop. Reducing and non-reducing sugar content successively increased over control with pyrite application at RSC level of 2.5 meL^{-1}.Dubey and Singh[12] and Flowers[13]explained that the increased accumulation of sugar at elevated salinity levels is one of the ways to combat salinity by an osmotic adjustment. Pattanagul and Thitisaksakul[14] suggested that it allows the plants to maximize sufficient storage reserves to support basal metabolism under stressed conditions. But, there was a decline in the values of sugar accumulation at RSC level of 5-7.5 meL^{-1} probably due to metabolic alterations and mineral deficiency that lead to decrease in growth parameters, premature leaf senescence, reduced chlorophyll content of the leaves and therefore, reduction in the net photosynthetic activity.

IV. CONCLUSION:

It is evident from the foregoing discussions that there was depressive effect on the chlorophyll 'a' and 'b' and sugar content due to irrigation with RSC water and the values reduced significantly over control in the treatments supplied with RSC levels of 5.0 and 7.5meL^{-1} .But the values enhanced over control with pyrite application from 0-50 gm pot^{-1} in all rhizobium treated and untreated plants. In inoculated plants, combination of 2.5 meL^{-1} RSC water supplemented with 50 g pyrite per pot provided most suitable conditions for plant's growth whereas 7.5 meL^{-1} RSC water without pyrite application was detrimental for plant growth

Table1. Physico-chemical properties of soil in field.

Parameters	Season 1	Season 2
pH(1:2.5)	8.00	8.05
ECe (dsm^{-1})	0.46	0.46
% Organic carbon	0.59	0.62
Carbonate ion (meL^{-1})	3.20	3.38
Bicarbonate ion (meL^{-1})	2.00	2.20
Na^+(meL^{-1})	2.20	2.10
K^+(meL^{-1})	0.66	0.64
Ca^{2+}+Mg^{2+}(meL^{-1})	4.30	4.15
RSC (meL^{-1})	0.90	1.25

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***Table. 2 Effect of pyrites and RSC water on chlorophyll content ('a' and 'b') and reducing and non-reducing sugar of Rhizobium inoculated and uninoculated black gram* for two consecutive seasons with variance.**

Treatment	Ch.' a' season 1	Ch.'a' season 2	Ch. 'b' season 1	Ch. 'b season 2	Reducing sugar season 1	Reducing sugar season 2	Non-reducing sugar season 1	Non-reducing sugar season 2
$R_0S_0I_0$	0.660	0.660	0.250	0.228	31.60	31.09	27.64	27.03
$R_0S_1I_1$	0.673	0.670	0.259	0.248	31.77	31.12	27.92	27.09
$R_0S_2I_0$	0.679	0.675	0.265	0.263	32.01	31.17	28.45	27.62
$R_1S_0I_1$	0.660	0.655	0.270	0.264	31.56	30.60	27.85	27.53
$R_1S_1I_0$	0.674	0.670	0.283	0.278	32.03	31.71	28.16	27.62
$R_1S_2I_1$	0.684	0.680	0.288	0.285	32.08	31.84	28.56	27.82
$R_2S_0I_0$	0.620	0.615	0.254	0.224	31.53	29.79	26.79	25.87
$R_2S_1I_1$	0.632	0.628	0.247	0.222	31.79	29.99	26.92	26.00
$R_2S_2I_0$	0.648	0.635	0.235	0.220	31.84	30.05	27.42	26.63
$R_3S_0I_1$	0.525	0.516	0.179	0.170	29.69	27.90	22.83	22.82
$R_3S_1I_0$	0.576	0.560	0.205	0.204	30.46	28.00	25.94	24.66
$R_3S_2I_1$	0.589	0.573	0.220	0.214	31.26	29.30	26.61	25.15
CD at 5%	0.033	0.040	0.035	0.035	1.098	2.232	0.958	1.578
SEm ±	0.011	0.014	0.012	0.012	0.374	0.791	0.327	0.538

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REFERENCES:

1. James JJ, Tiller RL and Richards RH (2005) Multiple resources limit plant growth and function in a saline-alkaline desert community. J. Ecol., 93: 113-126.

2. Kopittke P and Menzies N (2005)Effect of pH on Na induced Ca deficiency. Plant Soil, 269: 119-129.

3. Moradi F and IsmailAM (2007) Responses of photosynthesis, chlorophyll fluorescence and ROS-scavenging systems to salt stress during seedling and reproductive stages in rice. Ann. Bot., 99: 1161-1173

4. Cramer GR and RS Nowak (1992)Supplemental manganese improves the relative growth, net assimilation and photosynthetic rates of salt-stressed barley. Physiol. Plant., 84: 600-605.

5. Sharma SK and Manchanda HR (1989) Effect of irrigation with sodic water of increasing RSC levels on yield and NaCl content of chick pea. J. Indian Soc. Soil Sci. 37: 147-151.

6. Singh H, Singh J and Kumar Ajay (2005)Effect of phosphorus and sulfur on the grain and straw yield of black gram. Annal of Plant Soil Res.7(1):99-100.

7. Rai R, Ashraf MH and Chaudhary S N (1977) Pyrite as an amendment of soil. Ferti. Tech. 14(4):370-374.

8. Jayaraman J (1981) Laboratory manual of bio-chemistry. Willey Eastern Ltd. New Delhi.

9. Miller G L (1956) Use of dinitro-salicylic acid reagent for the determiation of reducing sugar. An. Chem. 31: 426-428.

10. Dubois M, Gilles KA, Hamilton JK, Rebers PA and Smith F (1956) Colorimetric method for determination of sugars and related substances. Anal. Chem., 28: 350-356.

11. Sinha RB and Sakal P (1993)Effect of pyrite and organic manures on sulphur nutrition of crops in calcareous soil. J. Indian Soc.SoilSci.41(2):312-315. .

12. Dubey RS and Singh AK (1999) Salinity induces accumulation of soluble sugars and alters the activity of sugar metabolising enzymes in rice plants. Biol. Plantarum, 42: 233-239.

13. Flowers TJ and Yeo AR(1989)Effects of Salinity on Plant Growth and Crop Yield. In: Environmental Stress in Plants, Cherry, J.H. (Ed.). Springer Verlag, Berlin, pp: 101-119.

14. Pattanagul W and Thitisaksakul M(2008)Effect of salinity stress on growth and carbohydrate metabolism in tree rice (Oryza sativa L.) cultivars differing in salinity tolerance. Indian J. Exp. Biol., 46: 736-742.