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Alleviation of adverse effects of saline water irrigation on maize hybrid by the external application of chemical protectants and organic manures

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Abstract
A field experiment was conducted to alleviate the salinity stress induced through saline water irrigation on the growth and yield of maize hybrid using chemical protectants and organic manures. The maize hybrid CO 6 was grown under saline water irrigation having a water salinity of 6.3dSm^{-1} in a black clay loam soil. Different organic manures (FYM, Vermicompost) and chemical protectants (acetyl salicylic acid (0.1mM), potassium chloride (1%), potassium nitrate (0.5%) and pink pigment facultative methylotrophs (1%) were tested for their significance in mitigating irrigation water induced saline stress on maize hybrid. The study revealed that, foliar spraying of 0.1 mM acetyl salicylic acid thrice at 30, 45 and 60 days after sowing along with basal soil application of 5t vermicompost ha^{-1} registered the highest grain and stover yield in the maize hybrid. Higher crop yield and growth was positively correlated with better biochemical compounds (soluble protein) and antioxidant enzyme activities (catalase, peroxidase, superoxy dismutase). This was closely followed by the foliar addition of 0.1 mM acetyl salicylic acid thrice plus 12.5 t FYM ha^{-1}. Combined effect of organic manures and chemical protectants gave better response than the individual effects.

Keywords: Saline water irrigation, foliar strategies, biochemical changes, maize hybrid, enzyme activity

1. Introduction

Salt stress is one of the major abiotic stresses that affect almost every aspect of physiology and biochemistry of a plant, resulting in the reduction of growth and yield of many crops (Foolad, 2010, Akram and Ashraf, 2011) [11, 2]. It is also well documented that the amount and quality of irrigation water available in the arid and semi-arid regions of the world are the major limiting factors to agricultural productivity (Ashraf and Foolad, 2007) [7]. In India majority of the underground water contain high salts concentration and their continuous use for irrigation adversely affects the crop production (Bali *et al*., 2015) [8]. Salt stress can disturb growth, photosynthetic processes and biochemical metabolisms by causing changes in the accumulation of Na^+, Cl^-, nutrients and disturbance in water and osmotic potential. The increasing concentration of Na^+ and Cl^- in the rooting medium suppresses the uptake of essential nutrients and alters ionic relationships.

To mitigate the adverse effect of saline water irrigation on soil and crops, chemical protectants and organics plays an important role by alleviating salt stress and improving the soil health by supplying adequate nutrients. Physical properties like bulk density, porosity, void ratio, water permeability and hydraulic conductivity were significantly improved when organics like vermicompost, FYM were applied in combination with chemical amendments which results in enhanced rice and wheat yields due water induced salinity (Dagar, 2007) [10].

Foliar spraying of chemical protectants, such as acetyl salicylic acid (ASA), Pink pigmented facultative methanotrophs (PPFM), Potassium chloride (KCl) and Potassium nitrate (KNO_3) can be used to promote growth and yield of plants under various stress conditions. Both foliar and soil application of different amendments positively influenced the plant growth characteristics including plant height, root length, grain and stover yield (Yildirim *et al*., 2008) [28]. Addition of chemical protectants through foliar spray could alleviate salt stress by influencing biochemical mechanisms and antioxidant enzymes (catalase, peroxidase, superoxy dismutase) activities which imparts stress tolerance to plants. Exogenous ASA applications inhibited Na accumulation and stimulated K, Ca and Mg uptake by crops which help to ameliorate the deleterious effects of water salinity on growth and yield (Khan *et al*., 2010) [18]. Positive effects of ASA on many cereal crops have been reported under salinity stress condition (Elwan and El-Hamahmi, 2009) [4] and lesser concentrations of ASA could be tried to enhance the pod yield of groundnut and to derive more economic benefit (Mulgir *et al*., 2014) [22]. In recent years, there has been much interest in the development of strategies to alleviate salt stress hence the present investigation was undertaken to evaluate the role of organic manures and chemical protectants on the growth and yield of maize hybrid and to understand the role of biochemical and antioxidant enzymes activity in mitigating the adverse effect of irrigation water salinity.

Materials and Methods

Field experiment was conducted to study the effect of different organic manures and chemical protectants to alleviate salt stress induced by saline water irrigation in a maize hybrid CO 6. The soil of the experimental field was clay loam in texture with a pH of 8.14, electrical conductivity of 1.98 dS m^{-1} and an irrigation water salinity of 6.30 dSm^{-1}. The treatment structure comprising five chemical protecatatnts as foliar treatments (control, 0.1mM ASA, 1% KCl, 0.5% KNO_3, 1% PPFM) and five sources of organic and inorganic amendments (Absolute control, NPK, 150% K, Vermicompost, FYM). There were about twenty five treatment combinations which were replicated three times in a factorially randomized block design (FRBD). Nitrogen and potassium fertilizers were applied at three splits and other soil amendments were applied as basal. Foliar spraying of chemical protectants was carried out thrice at 30, 45 and 60 days after sowing. The plants were grown to maturity and harvested. Plant samples were collected from each plot as per plan of work and fresh samples were used for various enzymes (SOD, POX and CAT) and biochemical analysis. The yield of maize grain or stover per hectare was recorded at harvest besides observing various growth and yield attributes. The data collected were subjected to statistical analysis following the method proposed by Gomez and Gomez (1984) [13].

Result and discussion

Effect of different organics and chemical protectants were evaluated to alleviate the salt stress induced by saline water irrigation on a maize hybrid. Significant variations were observed with different protectants in maize growth, yield and biochemical parameters.

Plant growth attributes

Plant growth attributes such as plant height, root length and lateral root length showed significant improvement with the application of organic manures, fertilizers and chemical protectants through soil and foliar spray (Table 1). Higher plant height, root length and lateral root length were recorded with the application of 100% NP+150%K+5 t vermicompost ha^{-1} (250, 24.8 and 19.4 cm) followed by 100% NP+150% K + 12.5 t FYM ha^{-1} (241, 23.3 and 17.4 cm) than other treatments. The effect of foliar spraying of 0.10 mM ASA was more pronounced and significant in influencing the growth attributes and closely followed by the foliar spraying of 1 % PPFM. However, combined effect of soil and foliar strategies recorded higher plant height than individual effect of chemical protectants. However irrigation water salinity showed greater reduction in the plant growth attributes which was evidenced in the control treatment.

Hartung (2004) [15] reported that the adverse effects of salinity on plant height and root length may be due o the diverse effects of salinity on meristematic cell division and elongation as well as root penetration. Gama *et al*. (2007) [12] also reported reduction in root length as a result of salinity. The positive effect of ASA could be attributed to the increased antioxidant enzymatic activities such as SOD, POX and catalase, detoxification of reactive oxygen species (ROS), enhanced CO_2 assimilation and photosynthetic rate besides the increased mineral uptake by plants (Yildirim and Dursun, 2009) [27]. Also ASA is a phenolic compound altered the auxin, cytokinine and Abscisic acid (ABA) balances in plants and increased the growth attributes simultaneously which improved the yield under saline water irrigation (Qados, 2015) [23]. Similarly all the chemical protectants actively involved in various plant metabolic activities under saline stress thus increasing the growth and yield of maize than unsprayed control.

Table 1: Effect of organic, fertilizers and chemical protectants on the plant growth attributes at harvest stage of Maize hybrid grown under saline water irrigation

Soil / Foliar	Control	NPK	K150	Vermi	FYM	Mean
Plant height (cm)						
Control	160±7.55	174±9.93	183±6.99	191±5.35	188±7.45	179
0.1mM ASA	207±9.26	229±9.72	233±7.27	250±4.24	241±5.73	232
0.5%KNO_3	175±8.40	190±4.20	199±8.67	224±8.29	211±9.43	200
1% KCl	167±7.91	187±9.01	191±6.65	216±5.86	208±6.95	194
1% PPFM	195±9.13	216±8.44	220±6.59	237±6.22	230±7.09	220
Mean	181±8.49	199±3.34	205±5.20	223±4.38	215±8.54	205
	F	A	FxA			
SEd	0.66	0.65	1.49			
CD (P=0.05)	1.36	1.32	2.96			
Root length (cm)						
Control	11.1±0.77	12.4±0.73	13.6±0.79	15.2±0.81	14.6±0.85	13.4
0.1mM ASA	17.6±0.25	21.4±1.12	22.7±1.30	24.8±1.24	23.3±0.74	22.0
0.5%KNO_3	14.7±1.01	16.2±0.49	17.0±0.84	19.4±1.12	18.8±1.07	17.2
1% KCl	13.6±0.85	14.8±0.88	16.2±0.72	17.9±1.03	17.6±1.00	16.0
1% PPFM	17.9±1.03	19.2±1.09	20.5±1.19	22.1±1.20	21.4±1.22	20.2
Mean	15.0±0.41	16.8±0.17	18.0±0.28	19.9±0.44	19.1±1.07	17.8
	F	A	FxA			
SEd	0.12	0.11	0.28			
CD (P=0.05)	0.25	0.22	0.56			

Lateral root length (cm)						
Control	9.23±0.52	9.80±0.47	11.0±0.41	12.5±0.66	11.6±0.68	10.8
0.1mM ASA	13.4±0.35	15.3±0.62	16.7±0.47	19.4±0.73	17.4±0.54	16.4
0.5%KNO_3	11.0±0.26	12.4±0.42	13.2±0.60	15.5±0.35	14.1±0.84	13.2
1% KCl	10.7±0.68	11.4±0.54	12.2±0.53	13.7±0.31	12.9±0.51	12.2
1% PPFM	12.7±0.44	13.9±0.61	14.8±0.67	17.8±0.48	17.0±0.70	15.2
Mean	11.4±0.13	12.6±0.50	13.6±0.27	15.7±0.41	14.6±0.25	13.6
	F	A	FxA			
SEd	0.06	0.05	0.14			
CD (P=0.05)	0.12	0.11	0.28			

F-Foliar S-Soil A - amendments ASA- Acetyl salicylic acid PPFM – pink pigmented facultative methylotrophs KCl -potassium chloride KNO_3 - Potassium nitrate NS - Non significant

Significant elevations in the growth rate of maize at high potassium application (150% K) in saline fields have been reported by Akram *et al.* (2010) [3] and are linked to continued water uptake and turgor maintenance, as potassium plays a key role in osmoregulation. Spray with potassium result an increase in leaf potassium content which was accompanied by increased rates of photosynthesis, photorespiration and RuBP carboxylase activity. Hence there was a considerable improvement in growth even under saline condition in the present investigation. Similar observation has also been reported in Maize by Khatoon *et al*. (2010) [19].

Grain and stover yield

The data on the grain and stover yield of maize revealed a significant improvement in the yield. Combined application of 100 % NP + 150% K + 5 t ha^{-1} vermicompost as basal and foliar spraying of 0.1mM ASA thrice recorded higher grain (7241 kg ha^{-1}) and stover (9651 kg ha^{-1}) yield followed by 100 % NP+ 150% K + 12.5 t FYM +0.1mM ASA (6699 and 9089 kg ha^{-1} respectively). Irrigation water salinity considerably reduced the grain and stover yield of maize which was reflected in the absolute control (4111 and 6105 kg ha^{-1}, Table 2).

Table 2: Effect of organic, fertilizers and chemical protectants on the stover and grain yield of Maize hybrid grown under saline water irrigation

Soil / Foliar	Control	NPK	K150	Vermi	FYM	Mean
Stover yield (kg ha^{-1})						
Control	6105	6603	6876	7556	7223	6873
0.1mM ASA	7218	8484	8871	9651	9182	8681
0.5%KNO_3	6458	7149	7358	8287	7863	7423
1% KCl	6261	6687	7213	7785	7498	7089
1% PPFM	6971	7689	8136	9089	8676	8103
Mean	6594	7322	7691	8474	8088	7634
	F	A	FxA			
SEd	31.8	28.2	71.1			
CD (P=0.05)	63.1	56.7	141			
Grain yield (kg ha^{-1})						
Control	4111	4833	5019	5309	5199	4894
0.1mM ASA	5143	5830	6290	7241	6901	6281
0.5%KNO_3	4843	5364	5661	5995	5881	5549
1% KCl	4766	5249	5496	5832	5656	5400
1% PPFM	4862	5459	5798	6699	6384	5840
Mean	4745	5347	5653	6215	6004	5593
	F	A	FxA			
SEd	27.9	25.6	62.5			
CD (P=0.05)	55.5	51.2	124			

F-Foliar S-Soil A - Amendments ASA- Acetyl salicylic acid PPFM – pink pigmented facultative methylotrophs KCl - potassium chloride KNO_3 - Potassium nitrate NS - Non significant

Vermicompost and Farmyard manure is a good source of many essential plant nutrients which are released upon decomposition and thus help to promote plant growth. In addition, they might have also helped in improving the buffering capacity of soil which suppressed the effect of salinity on plant growth (Haq *et al*. 2007 [14], Rajkhowa *et al*., 2010 [24]). The obtained results are in agreement with the findings of Kassab (2005) [17] and Thalooth *et al*. (2006) [26]. Aside chemical protectants might have also contributed to suppress the reactive oxygen species production by improving plant biochemical metabolism (Al-Busaidi *et al*., 2010) [5] thus increased crop yield. Increased stress tolerance index was observed with 0.10 mM ASA spray than chemical protectants (Fig. 3).

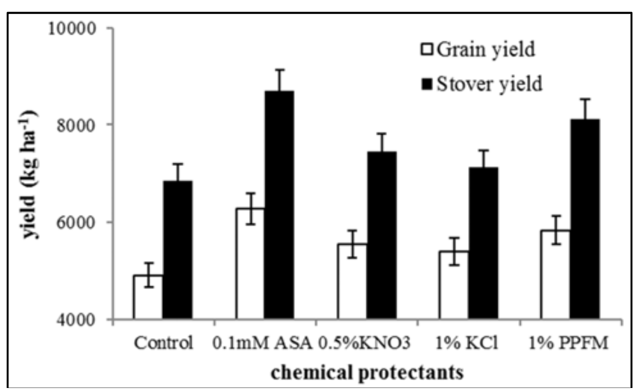


Fig 1: Effect of chemical protectants on the yield of maize hybrid

Biochemical changes

Biochemical parameters such as proline and soluble protein in the plants were adversely affected by the irrigation water induced saline stress. Application of different organic manures, fertilizer nutrients and chemical protectants showed marked influence in alleviating the adverse effect of water salinity. Significant difference was observed with soil and foliar application strategies in influencing the proline and soluble protein content.

Proline is the stress osmoprotectant, which is increasing under saline stress. In the present study inclusion of various organic manures and chemical protectants showed decreasing leaf proline content which confirmed the reduction in plant stress due to water salinity (Table 3). Among the foliar spray of different chemical protectants, 0.1mM ASA play a significant role by registering the lowest proline content (1.95 mg g^{-1}) followed by 1% PPFM (2.08 mg g^{-1}) spray. Soil application of organic manures and fertilizer inputs further complimented the role of chemical protectants in alleviating the plant salinity stress. Addition of 100 % NP+150 %K +5 t vermicompost ha^{-1} performed better in reducing the proline content (1.84 mg g^{-1}) followed by 100 % NP+150%K+ 12.5 t FYM ha^{-1} (1.97 mg g^{-1}). Higher proline accumulation was observed in plants grown with absolute control (2.71 mg g^{-1}).

Table 3: Effect of organic, fertilizers and chemical protectants on biochemical compounds and enzyme activities in the maize leaves at flowering stage under saline water irrigation

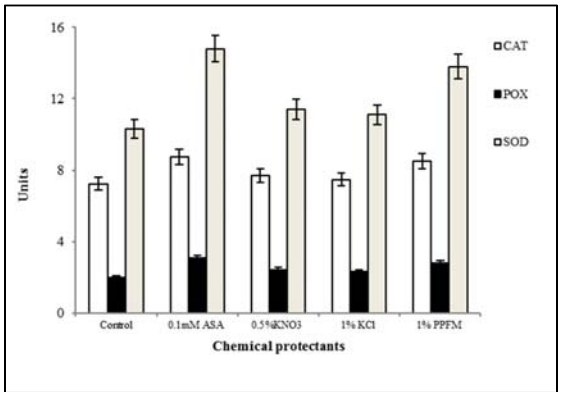
Soil / Foliar	Control	NPK	K150	Vermi	FYM	Mean
Proline						
Control	3.25±0.12	2.82±0.02	2.66±0.10	2.06±0.21	2.22±0.06	2.60
0.1mM ASA	2.26±0.06	2.11±0.02	1.96±0.06	1.67±0.13	1.77±0.03	1.95
0.5%KNO_3	2.58±0.05	2.47±0.03	2.23±0.13	1.78±0.15	1.90±0.15	2.19
1% KCl	3.08±0.11	2.71±0.07	2.50±0.09	1.96±0.19	2.13±0.11	2.48
1% PPFM	2.38±0.02	2.32±0.05	2.14±0.15	1.72±0.17	1.85±0.10	2.08
Mean	2.71±0.02	2.49±0.02	2.30±0.08	1.84±0.02	1.97±0.04	2.26
	F	A	FxA			
SEd	0.01	0.01	0.03			
CD (P=0.05)	0.02	0.03	0.05			
Soluble protein						
Control	5.19±0.18	5.65±0.21	5.95±0.47	6.44±0.45	6.28±0.25	5.90
0.1mM ASA	7.69±0.29	8.30±0.32	8.55±0.28	9.22±0.23	8.67±0.27	8.49
0.5%KNO_3	6.95±0.24	7.23±0.56	7.64±0.41	7.99±0.49	7.78±0.44	7.52
1% KCl	6.41±0.28	6.68±0.51	7.44±0.32	7.96±0.61	7.70±0.28	7.24
1% PPFM	7.41±0.37	7.59±0.59	8.09±0.45	8.64±0.34	8.46±0.33	8.04
Mean	6.73±0.21	7.09±0.16	7.53±0.17	8.05±0.13	7.78±0.04	7.44
SEd	0.01	0.01	0.03			
CD (P=0.05)	0.02	0.03	0.06			
Catalase						
Control	6.23±0.19	6.72±0.16	7.40±0.57	8.20±0.48	7.63±0.52	7.24
0.1mM ASA	8.18±0.12	8.13±0.24	8.91±0.50	9.35±0.43	9.14±0.45	8.74
0.5%KNO_3	7.07±0.15	7.36±0.39	7.77±0.33	8.15±0.38	8.10±0.40	7.69
1% KCl	6.81±0.26	7.11±0.16	7.54±0.37	8.02±0.52	7.90±0.55	7.48
1% PPFM	7.88±0.26	7.75±0.24	8.62±0.48	9.24±0.47	9.02±0.43	8.50
Mean	7.23±0.09	7.41±0.19	8.05±0.39	8.59±0.51	8.36±0.45	7.93
SEd	0.01	0.01	0.03			
CD (P=0.05)	0.02	0.03	0.05			
Peroxidase						
Control	1.80±0.14	1.84±0.17	2.00±0.15	2.14±0.13	2.10±0.15	1.98
0.1mM ASA	2.55±0.13	2.81±0.23	3.19±0.28	3.60±0.30	3.36±0.24	3.10
0.5%KNO_3	1.90±0.15	2.12±0.18	2.52±0.21	2.90±0.10	2.72±0.14	2.43
1% KCl	1.87±0.14	2.03±0.15	2.38±0.16	2.73±0.19	2.66±0.16	2.33
1% PPFM	2.29±0.20	2.57±0.10	2.95±0.35	3.22±0.24	3.09±0.22	2.82
Mean	2.08±0.03	2.27±0.09	2.61±0.19	2.92±0.17	2.79±0.17	2.53
SEd	0.01	0.01	0.02			
CD (P=0.05)	0.03	0.02	0.04			
Superoxy dismutase						
Control	8.83±0.35	9.47±0.69	10.4±0.72	11.8±0.52	11.1±0.47	10.3
0.1mM ASA	12.7±0.38	13.8±0.22	14.9±0.35	17.1±0.32	15.5±0.61	14.8
0.5%KNO_3	9.80±0.68	10.6±0.86	11.3±0.40	12.7±0.75	12.5±0.75	11.4
1% KCl	9.50±0.26	10.3±0.30	11.1±0.62	12.3±0.92	12.1±0.92	11.1
1% PPFM	11.5±0.54	12.5±0.62	13.9±0.59	16.1±0.35	14.8±1.02	13.8
Mean	10.5±0.31	11.3±0.40	12.3±0.22	14.0±0.20	13.2±0.35	12.3
SEd	0.02	0.02	0.04			
CD (P=0.05)	0.04	0.05	0.09			

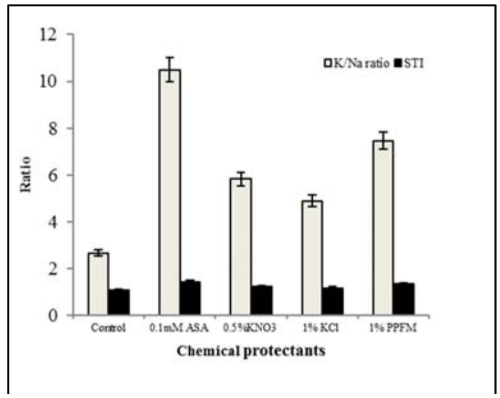
F-Foliar S-Soil A - amendments ASA- Acetyl salicylic acid PPFM – pink pigmented facultative methylotrophs KCl -potassium chloride KNO_3 - Potassium nitrate NS - Non significant

In contrary to proline content, the soluble protein content increased with the addition of 100 % NP+150% K+5 t vermicompost+ 0.1 mM ASA (9.22 mg g^{-1}) followed by 100 % NP+150% K + 12.5 t FYM+0.1 mM ASA (8.67 mg g^{-1}). Application of organic and chemical protectants increases the soluble protein content in the leaves thus the plants can able to tolerate the adverse effect of saline stress which was induced through saline water irrigation. Total soluble protein content decreased with salinity stress. These results have homology with the previous results of Anosheh (2012) [6] who reported that salinity stress reduced the accumulation of total soluble proteins in wheat and the provision of acetyl salicylic acid enhanced the accumulation of total soluble proteins.

Enzyme activities

Antioxidant enzymes play a vital role in plant metabolism under saline stress situations. The enzymes such as peroxidase, super oxy dismutase and catalase activity were widely varied under salinity and enhanced activity of peroxidase and superoxy dismutase activity were reported with the application of organic manures and chemical protectants (Table 3). However higher catalase activity was noted in control plots and decreased with the inclusion of organic manures and chemical protectants. Singh and Usha (2003) [25] reported that chemical protectants like acetyl salicylic acid regulate various physiological and biochemical processes in plants thus used as potential growth regulator to improve plant growth under saline conditions. The increased activity of antioxidant enzymes due to the addition of chemical protectants which is the key indicator to buildup protective mechanism to reduce oxidative damage induced by salt stress (Akankshajaiswal *et al*., 2014) [1].





CAT - $\mu g H_2O_2 min^{-1} g^{-1}$ POX- $EU min^{-1} mg^{-1}$ SOD - $EU g^{-1}$

Fig 2: Effect of chemical protectants on the antioxidant enzyme activities

Fig 3: Effect of chemical protectants on K^+/Na^+ ratioand stress tolerance index of maize hybrid

Basal soil application of 100% NP+150% K+ 5 t vermicompost + 0.10 mM ASA foliar spray thrice registered the highest peroxidase (3.60 EU min^{-1} mg^{-1}) and superoxy dismutase activities (17.1 EU g^{-1}) in the plants. However the lowest activities were obtained with the absolute control (1.80 EU min^{-1} mg^{-1} in POX and 8.83 EU g^{-1} in SOD). Increasing POX and SOD activity was followed the order of : 0.1mM ASA > 1% PPFM > 0.5% KNO_3 > 1% KCl > No spray (Fig 2). Kumara *et al*. (2010) [20] denoted a notable increase in superoxide dismutase activity due to pretreatment of salicylic acid in Gerbera. Singh and Usha (2003) [25] also discovered an exceptional increase in superoxide dismutase movement in wheat seedlings by exogenous provision of acetyl salicylic acid under saline stress.

Catalase activity increased with the application of organic manures and chemical protectants as soil and foliar spray. The highest activity was registered with 100% NP+150% K+ 5 t ha^{-1} vermicompost + 0.10 mM ASA (9.35 μg H_2O_2 min^{-1} g^{-1}). The lowest activity of catalase was recorded with no organic and chemical protectants (6.23 μg H_2O_2 min^{-1} g^{-1}). Significant accumulation of H_2O_2 in the plants under salinity might have decreased the activity of catalase (Mallik *et al*., 2011) [21] and reduction in the adverse effect by foliar feeding of chemical protectants resulted in greater catalase activity. Similar result was observed by Bhattacharjee (2012) [9] in rice cultivated under irrigation salinity.

The ratio of K^+/Na^+ is a good indicator of plant stress which was low in plants under salt stress. But application of ameliorants increased the K^+/Na^+ ratio under salt stress. The lowest K^+/Na^+ ratio was recorded in absolute control and the highest ratio was noted with 100% NP+150% K+ 5 t vermicompost +0.10 mM ASA (8.65) followed by 100 %NP+150% K+ 12.5 t FYM +0.10 mM ASA (6.90) in maize hybrid (Fig 3). Glutathione activity increased by the use of chemical protectants which was integrated into plant primary metabolism and influence the functioning of signal transduction pathways by modulating cellular redox state thus reduces the sodium and potassium ratio (Hemmat, 2007) [16].

Hence it can be concluded from the study that, conjoint application of different organic manures, fertilizers and chemical protectants as basal soil and foliar application is necessary for sustaining the productivity of maize grown under saline water irrigation water. Application of 100% NP+150% K either with 5 t vermicompost or 12. 5 t FYM as basal soil application coupled with foliar spraying of 0.1mM ASA thrice at 30, 45 and 60 days after sowing is found to be a better strategy to alleviate irrigation water induced salinity stress for higher production of maize hybrids.

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