J. Soils and Crops 32 (2) 287-295, December 2022

ISSN 0971-2836 (Print)
ISSN 2582-2756 (Online)

OSMOLYTES ACCUMULATION AND PLANT ENZYME ACTIVITIES IN DIFFERENT SUGARCANE GENOTYPES GROWN IN SODIC SOIL

A. V. Patil[1], A. L. Pharande[2], A.D. Kadlag[3], A.G. Durgude[4] and R. M. Naik[5]

ABSTRACT

A field experiment was conducted to assess the "Osmolyte accumulation and plant enzyme activities in sugarcane genotypes grown on sodic soil" during the preseason of 2015-16 at Post Graduate Instructional Farm, Department of Soil Science and Agricultural Chemistry, Mahatma Phule Krishi Vidyapeeth, Rahuri. The experiment was laid out in randomized block design comprising 27 sugarcane genotypes with two replications. The experimental soil was clay fine montmorillonite, isohyperthermic family of *Sodic Calciustert* and calcareous in nature with pHs 8.64, ECe 3.56 dS m^{-1}, exchangeable sodium percentage (ESP) 18.68 with high cation exchange capacity (CEC) 52.29 C mol (p+) kg^{-1} having swell-shrink property and categorized as sodic soil. The osmolyte accumulation (proline and glycinebetaine), chlorophyll content and enzyme activities *viz*., ascorbate peroxidase (APX), superoxide dismutase (SOD), nitrate reductase (NRA) and lipid peroxidase (LPO) were assayed at 75 and 240 days after planting (DAP).

The results indicated that CoM 0265 recorded significantly higher osmolytes like proline (4.39 and 3.66 µmoles g^{-1} dry wt.) and glycine betaine (1.82 and 1.68 mg g^{-1} dry wt.) which was followed by MS 6847 for proline (4.24 and 3.54 µmoles g^{-1} dry wt.) and glycine betaine (1.77 and 1.56 mg g^{-1} dry wt.) and MS 10001 for proline (3.83 and 3.43 µmoles g^{-1} dry wt.) and glycine betaine (1.73 and 1.48 mg g^{-1} dry wt.) at both 75 and 240 days after planting respectively. Significantly higher chlorophyll content was also recorded by CoM 0265 and MS 6847 than rest of the genotypes.

The NR activity was found significantly higher in the sugarcane genotype CoM 0265 (0.86 and 0.74 µMole nitrite produced g^{-1} fr. wt. hr^{-1}) which was followed by MS 6847 (0.84 and 0.72 µMole nitrite produced g^{-1} fr. wt. hr^{-1}) and MS 10001 (0.79 and 0.68 µMole nitrite produced g^{-1}fr.wt.hr^{-1}) at 75 and 240 DAP respectively. The sugarcane genotypes *viz*., CoM 0265, MS 6847 and MS 10001 recorded higher activity of APX at 75 DAP (2.84, 2.77 and 2.58 µmole ascorbate oxidized mg^{-1} protein min^{-1} respectively) and at 240 DAP (2.62, 2.52 and 2.38 µmole ascorbate oxidized mg^{-1} protein min^{-1} respectively). Further, the SOD activity in MS 6847, Co 99004 and CoM 0261 were higher at 75 DAP (8.64, 7.86 and 7.85 units mg^{-1} protein respectively) and at 240 DAP (6.95, 6.71 and 6.88 units mg^{-1} protein respectively) than rest of the genotypes. However, on the contrary, LPO activity (membrane damaging enzyme) was recorded statistically lower in CoM 0265, MS 6847 and MS 10001 at 75 DAP (0.97, 1.08 and 1.18 µmoles MDA g^{-1} fr. wt.) and at 240 DAP (0.81, 0.91 and 0.99 µmoles MDA g^{-1} fr. wt.) respectively.

The higher cane and commercial cane sugar yield was recorded by CoM 0265 (164.70 and 21.90 MT ha^{-1}), while at par results were obtained by MS 6847 (131.90 and 14.58 MT ha^{-1}), CoM 10051 (128.27 and 15.86 MT ha^{-1}) and MS 10001 (100.91 and 14.84 MT ha^{-1}).

(Key words : Osmolyte, enzyme- SOD, APX, NRA, LPO, sugarcane, sodic soil)

INTRODUCTION

Abiotic stresses (salinity/sodicity, drought, heat/cold, light and other hostile conditions) lead to the over production of reactive oxygen species (ROS) in plants, which are highly reactive and toxic and cause damage to protein, lipids, carbohydrates and DNA, which ultimately result in oxidative stress. In order to overcome oxidative stress, plants have developed two main antioxidants defense

1. Asstt. Professor, Dept. of Soil Science and Agril. Chemistry, College of Agriculture, Pune-411 005. Corresponding author : Email. patilssac1978@gmail.com
2. Ex. Dean, Faculty of Agriculture and Director of Instructions, MPKV, Rahuri, Tal. Rahuri, Dist, Ahmednagar, M.S. (India) 413 722
3. Ex. Head, Dept. of Soil Science and Agril. Chemistry, PGI, MPKV, Rahuri, Tal. Rahuri, Dist, Ahmednagar, M.S. (India) 413 722
4. Analytical Chemist, Micronutrient Scheme, Dept. of Soil Science and Agril. Chemistry, PGI, MPKV, Rahuri, Tal. Rahuri, Dist, Ahmednagar, M.S. (India) 413 722.
5. Ex. Head, Dept. of Biochemistry, PGI, MPKV, Rahuri, Tal. Rahuri, Dist, Ahmednagar, M.S. (India) 413 722

mechanisms that can be classified as non-enzymatic and enzymatic systems. The non-enzymatic system consists of small molecules such as vitamin like A, C and E along with glutathione, carotenoids and phenolics that can react directly with the ROS by scavenging them. Enzymatic system is represented by enzymes, among them; superoxide dismutase, peroxidase and catalase have the capacity to eliminate superoxide and hydrogen peroxide (Gill and Tuteja, 2010).

Under normal condition, there is a balance between ROS production and scavenging, which defines the normal steady-state level of intracellular ROS. In case of abiotic stress (salinity/drought) condition this balance is disturbed and ROS production is enhanced due to stomatal closure and the concomitant limitation on CO_2 fixation. Instead of having an immediate deleterious effect, this rise in ROS production is likely to be beneficial if kept under tight control. In fact, enhanced cellular ROS production is sensed by the plant as an alarm signal, that triggers defense pathway and acclamatory response, enables the plant to adapt to the changing environment (Vranova *et al.,* 2002).

Plants accumulate certain osmolytes like proline (Pro) and glycine betaine (GB), in response to drought and salinity to facilitate water uptake (Hare and Cress, 1997 and Ashraf and Foolad, 2007). In addition to osmotic adjustments, these osmolytes were suggested to be important for protecting cells against increased oxidative stress condition. Proline accumulates in the cytosol and the vacuole during stress (Aubert *et al.*, 1999 and McNeil *et al.,* 1999) and was shown to protest plant cells against damages caused by reduced state of O_2 and HO. (Matysik *et al.,* 2002). Further, nitrate reduction (NR) is also inhibited by salts. As nitrate is taken up by NO_3^- transporter, is reduced to ammonium by the sequential reaction of nitrate reductase in cytosol and nitrite reductase in plastid/ chloroplasts. Nitrate reductase is the first enzyme in the process of nitrate assimilation, catalyses the reduction of nitrate to nitrite while "' Pyrroline-5-Carboxylate Synthase enzyme increases the proline synthesis in stress condition. It was reported that, increasing salt stress decreased plant growth and inhibited the nitrate reduction and ammonium assimilation (Debouba *et al.*, 2006).

In contrast to proline, glycine betaine is not thought to directly scavenge ROS during stresses (Smirnoff and Cumbes, 1989; Chen and Murata, 2008). However, glycine betaine has been shown to protect cells against oxidative damage during abiotic stresses (Park *et al.*, 2007, Chen and Murata, 2008). Glycine betaine accumulates mainly in the chloroplast and is involved in the maintenance of PSII efficiency under stress conditions (Ashraf and Foolad, 2007 and Ben Hassine *et al.,* 2008). Proline is a major component of amino acid pool in plants and the level of proline increases under saline condition.

Sodic soils, due to high ESP, possess poor physical and chemical properties. The high exchangeable Na^+ content of these soils leads to dispersion of fine clay particles resulting into low permeability, crusting and hardening of the surface soil upon drying. As a result, the aeration, soil-water movement and root growth hampered. The soil have poor aggregate stability, low organic matter content, toxic concentration of CO_3^{2-} of sodium, poor microbial activities due to strong alkaline pH and reduced availability of N, K, Zn and Fe which affect the productivity of these soils (Sen, 2003). The presence of high amount of salts in soil profile adversely affect the availability and uptake of mineral nutrients and consequently the crop growth and its quality (More *et al.*, 1994).

As the area of salt affected soils consistently increasing each year in Maharashtra and in almost all the states of India. In order to address this situation, it is necessary to adopt either reclamation measures or to screen the genotypes resistant to abiotic stresses like excess salts. Maharashtra have significant area (9.87 lakh ha) under sugarcane particularly grown on medium and deep black soils. Therefore, it is necessary to screen the sugarcane genotypes to assess their suitability for such salt affected areas. Hence, the experiment was planned with an objective to assess the accumulation of osmolytes along with antioxidant enzyme assays for screening of sugarcane genotypes.

MATERIALS AND METHODS

The experiment was laid out in randomized block design comprising 27 sugarcane genotypes with two replications *viz.,*CoM 0265, CoM 8516, CoM 9516, MS 10001, CoM 10051, Co 99004, CoM 0254, Co 62175, Co 740, Co 96009, Co 94012, CoC 671, Co 94005, Co 94004, Co 94001, MS 6847, CoM 08030, CoM 0261, PDN 13002, PDN 13003, PDN 13004, PDN 13007, CoM 09022, CoPDN 13002, CoPDN 13003, CoM 09057, Co 6032. The planting material of sugarcane genotypes were procurred from the Central Sugarcane Research Station, Padegaon, Post. Nira., Dist. Satara (M.S.). The soil of the experimental site are classified as fine montmorillonite, isohyperthermic family of *Sodic Calciustert*.

The initial representative soil sample was collected before planting of sugarcane and saturation paste extract analysis was carried out for pHs, ECe, SAR, RSC by using standard methods described by Richards (1968). The representative soil sample was also analyzed for ESP and SAR (Jackson, 1973). Further, water (well water) used for irrigation was also analyzed for cations and anions along with derived parameters (Richards, 1968). The soil of the experimental site is classified as fine montmorillonite hyperthermic family of *Sodic Calciustert*. Initial soil samples were collected and analyzed at the start of experiment having low available nitrogen, low phosphorus and very high available potassium. The soil was highly alkaline in reaction with total soluble salt content of soil extract was 2278 mg l^{-1} along with exchangeable sodium percentage (ESP) 18.68, hence it is categorized as sodic soil (Richard, 1968). The analytical data of initial soil properties as stated in Table 1.

The recommended dose of fertilizer (340:170:170 kg N, P_2O_5 and K_2O ha^{-1}) was applied through chemical fertilizers *viz*., urea, single superphosphate and muriate of potash along with farm yard manure (25 t ha^{-1}). The leaf tissue sampling (third leaf from top) of sugarcane were carried out at 75 and 240 DAP for the analysis of proline, glycine betaine, Chlorophyll, ascorbate peroxidase, superoxide dismutase, nitrate reductase and lipid peroxidation rate.

Proline content in leaf tissues of sugarcane genotypes was determined by using the acid ninhydrin reagent as per the method described by Bates *et al*. (1973). Glycine betaine content in leaves of sugarcane genotypes was determined by using Dragendorff reagent as per the method described by Stumpt (1984). Chlorophyll was estimated by extracting the leaf material in 80% acetone and calculated as described by Arnon (1949). The *in vivo* nitrate reductase assay under anaerobic conditions was assayed as per the method of Sawhney *et al*. (1978). The ascorbate peroxidase (APX) activity was assayed according to the method of Nakano and Asada (1998). SOD activity was assayed spectrophotometrically, as the inhibition of photochemical reduction of nitro-blue tetrazolium (NBT) at 560 nm (Beauchamp and Fridovich, 1971). The level of lipid peroxidation rate (LPO) was measured in terms of malondialdehyde (MDA) content, a product of LPO, following the method of Heath and Packer (1968).

RESULTS AND DISCUSSION

Osmolyte accumulation (proline and glycine betaine), total chlorophyll and activities of plant enzymes in 27 sugarcane genotypes grown in sodic soil were assesses periodically (75 and 240 DAP). Osmolytes, plant enzymes and total chlorophyll content in various sugarcane genotypes were significantly affected by sodic soil.

Proline content was found significantly higher in sugarcane genotype CoM 0265 at 75 and 240 DAP (4.39 and 3.66 µmoles g^{-1} dry wt.) and statistically on par with MS 6847 (4.24 and 3.54 µmoles g^{-1} dry wt.) (Table.2). It was numerically followed by MS 10001 (3.83 and 3.43 µmoles g^{-1} dry wt.), CoM 10051 (3.65 and 3.12 µmoles g^{-1} dry wt.) and CoM 08030 (3.46 and 2.98 µmoles g^{-1} dry wt.). The proline accumulation in sugarcane genotypes at 75 DAP was in order of CoM 0265 > MS 6847 > MS 10001 > CoM 10051 > CoM 08030 > Co 94012 > CoM 09022 > CoPDN 13002. The association of free proline with salt stress has been reported by Chu *et al*. (1976) and Stewart and Lee (1974). Accumulation of more proline in salt tolerant genotypes might be due to the increased activity of "' Pyrroline-5-Carboxylate synthase enzyme and reduction of pyrroline dehydrogenase under salinity condition to survive the plant under adverse salt stress condition (Mahajan *et al.*, 2013).The higher content of proline in sugarcane genotypes due to sodicity balanced the decreased osmotic potential in vacular compartment due to excess ionic stress. Proline accumulation triggered to occur as a result of initiation of other responses to salinity stress and also as an indicator of stress damage. Proline may serve as an intracellular osmotic solute for the maintenance of osmotic balance between cytoplasm and vacuole (Kishor *et al.*, 2005). Similarly, proline has a good impact on maintaining the structure of the enzymes and removal of reactive oxygen species (Kumar *et al.*, 2006).

Glycine betaine, a quaternary ammonium compound is another important osmolyte whose level increased substantially under salinity in any crop plants. Glycine betaine found significantly higher at 75 and 240 DAP in CoM 0265 sugarcane genotype (1.82 and 1.68 mg g^{-1} dry wt.) and statistically on par with MS 10001, CoM 10051, MS 6847 at 75 DAP (1.73, 1.67 and 1.77 mg g^{-1} dry wt., respectively) (Table.2). Similarly, it was followed by MS 10001 and MS 6847 at 240 DAP (1.48 and 1.56 mg g^{-1} dry wt., respectively). The higher content of glycine betaine indicated the salt tolerance of sugarcane genotypes under sodic soils. The glycine betaine content in plant correlated with salt tolerance. The salt tolerant genotypes accumulated the highest glycine betaine levels, moderately tolerant genotypes accumulate intermediate levels and sensitive species accumulate low levels or no glycine betaine. The accumulation of glycine betaine might serve as an intracellular osmoticum and closely correlated with the elevation of osmotic pressure (Kavikishore *et al.*, 1995). Glycine betaine and trehalose acts as osmoprotectants by stabilizing quaternary structure of proteins and highly ordered states of membranes (Huang *et al.*, 2000).

Chlorophyll content is related with photosynthesis. The total chlorophyll content of sugarcane genotypes was ranged between 0.90-2.08 mg g^{-1} fr. wt. at 75 DAP and 0.81-2.28 mg g^{-1} fr. wt. at 240 DAP (Table.2). The sugarcane genotype CoM 0265 recorded the highest total chlorophyll content at 75 and 240 DAP (2.08 and 2.28 mg g^{-1} fr. wt.). It was followed by genotype MS 6847 (2.03 and 2.17 mg g^{-1} fr. wt.). However, the sugarcane genotypes MS 10001, CoM 10051 and Co 94012 recorded considerably higher total chlorophyll content at 240 DAP (2.05, 2.01 and 2.10 mg g^{-1} fr. wt., respectively) in sodic soils grown during preseasonal sugarcane. The less amount of total chlorophyll at 75 and 240 DAP were recorded by Co 96009 (0.93 and 0.88 mg g^{-1} fr. wt.), Co 94001 (0.90 and 0.82 mg g^{-1} fr. wt.) and Co 86032 (0.96 and 0.81 mg g^{-1} fr. wt.), respectively. The rest of the sugarcane genotypes also recorded the total chlorophyll content to the extent of 1.15 to 1.98 mg g^{-1} fr. wt. at 75 DAP and 1.14 to 1.98 mg g^{-1} fr. wt. at 240 DAP. The sugarcane genotypes *viz.,* CoM 0265, MS 6847 and MS 10001 recorded higher content of total chlorophyll performed well for their physiological growth. This signifies that lower pigment degradation favoured better photosynthetic activities under sodic condition (Vasantha and Rajlakshmi, 2009). Parbat *et al.* (2018) also reported that the higher amount of total chlorophyll was observed in resistant genotypes of groundnut as compared to susceptible genotypes for tikka disease. The genotypes CoM 0265, MS 10001 and MS 6847 were able to maintain higher levels of chlorophyll might be because of scavenging of ROS which protect the cell

membrane from damage (Joshi and Naik, 1977). Bele *et al.* (2021) also reported in his study that white rice cultivar SKL-3-1-41-8-33-15 recorded higher total chlorophyll content which leads to higher yield and yield contributing characters as compared with other cultivars studied. Hence, cultivar SKL-3-1-41-8-33-15 can be recommended for cultivation.

The sodicity effect was also studied on enzymatic activities in sugarcane genotype at tillering and grand growth stage. The higher ascorbate peroxidase activity which helps to scavenge ROS which is a result of oxidative stress in the plant system are desirable to sustain under biotic or abiotic stress condition. The sugarcane genotypes *viz*., CoM 0265, MS 6847, MS 10001, CoM 10051 and CoM 08030 were recorded significantly higher ascorbate peroxidase activity at 75 DAP (2.84, 2.77, 2.58, 2.49 and 2.34 µmoles ascorbate oxidized mg^{-1} protein min^{-1} respectively) and at 240 DAP (2.62, 2.52, 2.38, 2.17 and 2.05 µmoles ascorbate oxidized mg^{-1} protein min^{-1} respectively) (Table 3). These results indicated that these genotypes tolerate to grow on sodic soils. Similar finding was reported by Rasool *et al.*, (2013). The sugarcane genotypes *viz.,* CoM 0265, MS 6847, CoM 10051 and MS 10001 with high ascorbate peroxidase activity at 75 and 240 DAP showed their adaptability in sodic soil by maintaining higher chlorophyll content and nitrate reductase activity with low lipid peroxidation rate as compared to salt susceptible genotypes *viz*., Co 94001, CoM 0261, Co PDN 13003 and Co 86032. The results shows that antioxidant enzyme protect cell from oxidative burst by scavenging ROS under sodic soil. Thus, it indicated that the plants have developed an antioxidant defense system to cope with oxidative damage under extremely adverse conditions that include H_2O_2 sensitive antioxidative enzyme APX.

Superoxide dismutase enzyme is the first line defense against the salt stress. The higher SOD activity increases the adaptability of plant species under salt stress. The superoxide dismutase enzyme activity was significantly higher in sugarcane genotypes MS 6847 (8.64 and 6.95 units mg^{-1} protein), CoM 99004 (7.86 and 6.71 units mg^{-1} protein), CoM 0261 (7.85 and 6.88 units mg^{-1} protein), MS 10001 (7.34 and 6.50 units mg^{-1} protein), Co 94004 (7.50 and 5.83 units mg^{-1} protein), Co 94005 (7.39 and 6.73 units mg^{-1} protein) at 75 and 240 DAP respectively (Table.3). The higher activity of superoxide dismutase in the sugarcane genotypes probably suggested the better adaptability of sugarcane genotypes in sodic soil. The salt tolerant genotypes *viz*., MS 10001 and MS 6847 have higher superoxide dismutase activity and chlorophyll content with lower lipid peroxidation rate as compared to salt susceptible genotypes *viz*., Co 94001, CoM 0261, CoPDN 13003 and Co 86032, suggest that superoxide dismutase constitutes the first line defense *via* detoxification of superoxide radicals thereby maintaining integrity of membranes of plant tissue. Similar observations were reported by Sairam *et al.* (2002). They found that the SOD activity was increased significantly under salinity stress at all three stages in both the genotypes and more activity was observed at S2 salinity level. SOD activity was highest in Kharchia 65 wheat genotype at 50 % anthesis in both control and salinity treatments when compared with other two stages.

The higher nitrate reductase activity in the plant system are desirable to withstand in adverse salt stress condition. The highest *in vivo* nitrate reductase activity was recorded in the sugarcane genotype CoM 0265 at 75 and 240 DAP (0.86 and 0.74 µmoles nitrite produced g^{-1} fr. wt. hr^{-1}) and on par with sugarcane genotypes MS 10001, MS 6847 and CoM 08030 at 75 DAP (0.79, 0.84 and 0.81 µmoles nitrite produced g^{-1} fr. wt. hr^{-1}) and 240 DAP (0.68, 0.72 and 0.63 µmoles nitrite produced g^{-1} fr. wt. hr^{-1} respectively) (Table.3). These results indicated that the salt tolerant sugarcane genotypes have an ability to absorb the nitrogen from soil under salt stress condition and maintain the nitrate reductase activity in plant system. This is an indication of good adaptability of sugarcane genotypes in sodic soils. These results are in agreement with the findings of Satbhai and Naik *et al.* (2014), who reported that the reduction in chlorophyll content and nitrate reductase activity was less in salt tolerant sugarcane variety CoM 0265 than that of CoC 671, a salt susceptible variety. Mahajan *et al.* (2013) observed that salt tolerant varieties showed higher nitrate reductase activity under sodic soil as compared to normal soil.

The peroxidation of lipids are considered as the most damaging process known to occur in every living organism. The lower lipid peroxidation rate is desirable for plant species to withstand under abiotic stress like salt stress. The lipid peroxidation rate was found significantly lower at 75 and 240 DAP in CoM 0265 (0.97 and 0.81 µmoles MDA g^{-1} fr. wt.) and on par with MS 10001 (1.18 and 0.99 µmoles MDA g^{-1} fr. wt.), MS 6847 (1.08 and 0.91 µmoles MDA g^{-1} fr. wt.) and CoM 10051 (1.47 and 1.23 µmoles MDA g^{-1} fr. wt.) (Table 3). Similar observations were recorded by Mohamed *et al.* (2012). The sugarcane genotypes CoM 0265, MS 10001, MS 6847 and CoM 10051 were able to maintain higher chlorophyll content with lower lipid peroxidation, thus, protect the cell membrane from damage due to oxidative burst. The higher LPO and lower membrane stability with lower chlorophyll content in susceptible genotypes might be due to reduction of photochemical quenching in PSII due to salt stress. Many reports have also noticed same type of observations in different crop species while screening for salt tolerance (Taishi *et al.*, 2000; Sairam and Tyagi, 2004 and Gomathi *et al.*, 2012).

The cane and commercial cane sugar yield of sugarcane genotypes grown on sodic soil in preseasonal sugarcane are significantly influenced by sodicity of soil. The sugarcane genotype CoM 0265 recorded significantly higher cane and commercial cane sugar yield (164.70 and 21.90 MT ha^{-1}) followed by MS 6847 (131.90 and 14.58 MT ha^{-1}), CoM 10051 (128.27 and 15.86 MT ha^{-1}), Co 94012 (102.06 and 15.14 MT ha^{-1}) and MS 10001 (100.91 and 14.84 MT ha^{-1}) (Table 4). The lowest cane and commercial cane sugar yield was recorded by Co 740 (32.73 and 3.73 MT ha^{-1}). The cane yield of sugarcane genotypes was in order of CoM 0265 > MS 6847 > CoM 10051 > Co 94012 > MS 10001

> CoM 08030. Commercial cane sugar yield of sugarcane genotypes was in order of CoM 0265 > CoM 10051 > Co 94012 > MS 10001 > MS 6847 > CoPDN 13002 > CoM 08030. The yield of sugarcane genotypes are the function of nutrient uptake from soil under prevailing soil conditions (Tiwari *et al.*, 2006).

In the present study, the sugarcane genotype CoM 0265 recorded the maximum cane and commercial cane sugar (CCS) yield followed by MS 6847, CoM 10051 and MS 10001 due to higher chlorophyll content, osmolyte accumulation and antioxidant enzyme activity, on the contrary lipid peroxidation rate was reduced. Hence, these genotypes are having higher defense mechanism and suitable to grow under sodic shrink-swell soils. Higher yield of sugarcane genotypes is due to higher osmolyte accumulation and antioxidant enzyme activity under stress condition is due to mechanism of ROS balancing in sodic soil.

Table 1. Characterization of experimental soil and irrigation water

Properties	Values	Properties	Values	
Soil saturation paste extract analysis		**Irrigation water analysis**	**Canal**	**Well**
pHs	8.64	pH	7.70	7.86
ECe (dSm	3.56	EC (dSm^{-1})	0.35	2.70
Cations (me l^{-1})		Cations (me l^{-1})		
i. Calcium	3.10	i. Calcium	1.32	3.43
ii. Magnesium	6.03	ii. Magnesium	0.21	5.07
iii. Sodium	29.42	iii. Sodium	0.11	12.83
iv. Potassium	0.14	iv. Potassium	0.07	0.013
Anions (me l^{-1})		Anions (me l^{-1})		
i. Carbonate	Traces	i. Carbonate	Traces	Traces
ii. Bicarbonate	20.00	ii. Bicarbonate	1.60	10.26
iii. Chloride	10.15	iii. Chloride	0.51	8.52
iv. Sulphate	7.29	iv. Sulphate	0.41	6.13
Sodium Adsorption Ratio (SAR)	13.75	Sodium Adsorption Ratio (SAR)	0.13	6.23
Residual Sodium Carbonate (me l^{-1})	10.87	Residual Sodium Carbonate (me l^{-1})	0.07	1.76
Magnesium : Calcium ratio	1.95	Mg :Ca ratio	0.16	1.48

Soil

Chemical properties			
Soil pH (1:2.5)	8.58	DTPA micronutrients (mg kg^{-1})	
Electrical Conductivity(dSm^{-1})	1.15	i. Iron	5.01
Available Nitrogen (kg ha^{-1})	175.62	ii.Manganese	11.07
Available Phosphorus (kg ha^{-1})	7.30	iii.Zinc	0.54
Available Potassium (kg ha^{-1})	670.42	iv.Copper	2.51
Organic Carbon (%)	0.41	**Physical properties**	
Calcium Carbonate (%)	16.89	Particle size distribution	
Exchangeable Calcium (meq 100^{-1} g soil)	30.91	i. Coarse sand	5.91
Exchangeable Magnesium (meq 100^{-1} g soil)	13.36	ii. Fine sand	12.92
Exchangeable Sodium (meq 100^{-1} g soil)	9.77	iii. Silt	22.94
Cation Exchange Capacity CEC [C mol (p^+) kg^{-1}]	52.29	iv. Clay	57.32
Exchangeable Sodium Percentage (ESP)	18.68	Textural class	Clay
		Bulk density (Mg m^{-3})	1.57
		Hydraulic conductivity(cm hr^{-1})	0.18

Table 2. Effect of sodicity on Osmolytes accumulation and chlorophyll content in sugarcane genotypes at tillering stage (75 days after planting) and grand growth stage (240 days after planting)

Sr. No.	Genotypes	Proline (ìmoles g^{-1} dry wt.)		Glycine betaine (mg g^{-1} dry wt.)		Total chlorophyll (mg g^{-1} fr. wt.)	
		75 DAP	240 DAP	75 DAP	240 DAP	75 DAP	240 DAP
1.	CoM 0265	4.39	3.66	1.82	1.68	2.08	2.28
2.	CoM 8516	2.80	2.46	1.27	0.98	1.73	1.62
3.	CoM 9516	1.56	1.42	0.85	0.73	1.18	1.21
4.	MS 10001	3.83	3.43	1.73	1.48	1.98	2.05
5.	CoM 10051	3.65	3.12	1.67	1.40	1.96	2.01
6.	Co 99004	2.90	2.54	1.32	1.28	1.77	1.83
7.	CoM 0254	2.27	1.96	1.15	0.94	1.65	1.76
8.	Co 62175	2.29	1.92	1.16	0.95	1.68	1.52
9.	Co 740	1.63	1.42	0.89	0.75	1.23	1.14
10.	Co 96009	1.42	1.23	0.75	0.64	0.93	0.88
11.	Co 94012	3.07	2.96	1.40	1.24	1.89	2.10
12.	CoC 671	2.52	2.40	1.16	0.96	1.69	1.62
13.	Co 94005	2.84	2.48	1.31	1.16	1.73	1.84
14.	Co 94004	2.59	2.39	1.20	1.04	1.73	1.70
15.	Co 94001	1.34	1.12	0.71	0.62	0.90	0.82
16.	MS 6847	4.24	3.54	1.77	1.56	2.03	2.17
17.	CoM 08030	3.46	2.98	1.42	1.36	1.92	1.98
18.	CoM 0261	1.51	1.36	0.81	0.71	1.15	1.22
19.	PDN 13002	2.06	1.88	1.03	0.83	1.57	1.48
20.	PDN 13003	2.11	1.83	1.04	0.86	1.60	1.53
21.	PDN 13004	2.53	2.36	1.18	0.98	1.70	1.78
22.	PDN 13007	2.02	1.87	1.02	0.88	1.53	1.47
23.	CoM 09022	3.06	2.82	1.35	1.18	1.85	1.94
24.	CoPDN 13002	2.95	2.78	1.34	1.12	1.77	1.86
25.	CoPDN 13003	2.01	1.88	0.97	0.82	1.30	1.37
26.	CoM 09057	2.84	2.41	1.34	1.23	1.74	1.90
27.	Co 86032	1.47	1.31	0.79	0.66	0.96	0.81
	SE(m) ±	**0.17**	**0.14**	**0.10**	**0.09**	**0.07**	**0.08**
	CD at 5 %	**0.49**	**0.40**	**0.28**	**0.25**	**0.20**	**0.25**

Table 3. Effect of sodicity on APX, SOD, NR activity and LPO content in sugarcane genotypes at tillering stage (75 days after planting) and grand growth stage (240 days after planting)

Sr. No.	Genotypes	APX (ìmoles ascorbate oxidized mg^{-1} protein min^{-1})		SOD (units mg^{-1} protein)		NR activity (ìmoles nitrite produced g^{-1} fr. wt. hr^{-1})		LPO (ìmoles MDA g^{-1} fr. wt.)	
		75 DAP	240 DAP	75 DAP	240 DAP	75 DAP	240 DAP	75 DAP	240 DAP
1.	CoM 0265	2.84	2.62	7.24	6.79	0.86	0.74	0.97	0.81
2.	CoM 8516	1.98	1.83	7.01	6.11	0.53	0.46	2.98	2.76
3.	CoM 9516	1.56	1.44	6.98	4.23	0.42	0.42	3.34	3.16
4.	MS 10001	2.58	2.38	7.34	6.50	0.79	0.68	1.18	0.99
5.	CoM 10051	2.49	2.17	7.27	6.52	0.73	0.62	1.47	1.23
6.	Co 99004	2.07	1.92	7.86	6.71	0.49	0.47	3.18	2.97
7.	CoM 0254	1.91	1.81	7.65	6.49	0.50	0.43	2.12	1.88
8.	Co 62175	1.91	1.77	6.38	5.36	0.52	0.52	2.63	2.43
9.	Co 740	1.63	1.41	5.97	4.52	0.45	0.48	3.77	3.63
10.	Co 96009	1.37	1.39	5.77	4.66	0.41	0.51	4.06	3.84
11.	Co 94012	2.23	2.20	7.09	7.28	0.74	0.56	1.67	1.52
12.	CoC 671	1.91	1.74	6.65	5.51	0.58	0.46	3.24	2.94
13.	Co 94005	2.00	1.78	7.39	6.73	0.51	0.50	2.56	2.37
14.	Co 94004	2.04	1.86	7.50	5.83	0.54	0.48	2.44	2.27
15.	Co 94001	1.36	1.28	5.46	4.32	0.40	0.42	4.29	4.11
16.	MS 6847	2.77	2.52	8.64	6.95	0.84	0.72	1.08	0.91
17.	CoM 08030	2.34	2.05	6.61	5.59	0.81	0.63	1.60	1.44
18.	CoM 0261	1.66	1.46	7.85	6.88	0.41	0.47	3.72	3.58
19.	PDN 13002	1.82	1.71	6.53	5.42	0.56	0.46	2.69	2.46
20.	PDN 13003	1.82	1.62	6.50	5.48	0.52	0.54	2.31	2.06
21.	PDN 13004	1.95	1.70	6.21	5.73	0.61	0.43	2.02	1.81
22.	PDN 13007	1.77	1.66	5.95	5.12	0.58	0.45	2.47	2.32
23.	CoM 09022	2.23	2.12	7.10	6.10	0.62	0.58	1.44	1.24
24.	CoPDN 13002	2.10	1.82	6.22	5.88	0.63	0.54	1.63	1.47
25.	CoPDN 13003	1.75	1.60	6.15	4.74	0.47	0.43	3.68	3.41
26.	CoM 09057	2.05	1.74	7.32	6.20	0.60	0.56	3.63	3.05
27.	Co 86032	1.47	1.32	6.94	5.12	0.43	0.46	4.39	4.16
	SE (m) ±	0.16	0.13	0.12	0.11	0.05	0.05	0.22	0.12
	CD at 5 %	0.46	0.38	0.34	0.32	0.14	0.15	0.64	0.35

Table 4. Effect of sodicity on cane and commercial cane sugar yield of sugarcane genotypes

Sr. No.	Treatments (Genotype)	Yield (MT ha^{-1})		CCS yield(MT ha^{-1})
		Cane	Top	
1.	CoM 0265	164.70	20.19	21.90
2.	CoM 8516	63.27	23.31	8.60
3.	CoM 9516	47.29	17.00	5.97
4.	MS 10001	100.91	16.32	14.84
5.	CoM 10051	128.27	17.15	15.86
6.	Co 99004	66.66	14.84	8.49
7.	CoM 0254	77.66	20.59	10.31
8.	Co 62175	67.21	17.08	7.39
9.	Co 740	32.73	26.55	3.73
10.	Co 96009	47.82	13.84	6.35
11.	Co 94012	102.06	14.28	15.14
12.	CoC 671	77.96	18.21	11.62
13.	Co 94005	76.25	16.49	9.53
14.	Co 94004	81.57	18.87	12.72
15.	Co 94001	34.02	10.27	4.95
16.	MS 6847	131.90	21.74	14.58
17.	CoM 08030	98.12	16.58	13.71
18.	CoM 0261	45.79	12.32	5.38
19.	PDN 13002	67.81	12.59	10.14
20.	PDN 13003	76.34	18.98	10.36
21.	PDN 13004	82.26	19.32	10.56
22.	PDN 13007	60.85	14.80	7.36
23.	CoM 09022	90.98	20.92	10.83
24.	CoPDN 13002	90.61	19.44	14.33
25.	CoPDN 13003	56.92	16.59	6.48
26.	CoM 09057	71.45	14.45	9.98
27.	Co 86032	51.11	14.34	6.72
	SE (m) ±	7.28	1.94	1.24
	CD at 5 %	21.17	5.64	3.60

REFERENCES

Arnon, D.I. 1949. Copper enzymes in isolated chloroplasts. Polyphenoloxidase in *Beta vulgaris*. Plant Physiol. **24**:1-15.

Ashraf, M. and M. R. Foolad, 2007. Roles of glycine betaine and proline in improving plant abiotic stress resistance. Environ. Exp. Bot. **59**:206-216.

Aubert, S., F. Hennion, A. Bouchereau, E. Gout, R. Bringly and A. J. Dorne, 1999. Subcellular compartmentation of proline in the leaves of the subantarctic Kerguelen cabbage *Pringleaantiscorbutica* R. Br. *In vivo* 13C-NMR study. Plant Cell Environ. **22**:255-259.

Bates, L.S., R. P. Waldren and I. D. Teare, 1973. Rapid determination of free proline for water stress studies. Plant Soil. **39**:205–207.

Beauchamp, C. and I. Fridovich, 1971. Superoxide dismutase: Improved assay and assay applicable to acrylamide gels. Anal. Biochem**. 44**:276–287.

Ben Hassine, A., M. E. Ghanem, S. Bouzid and S. Lutts, 2008. An inland and a coastal population of the Mediterranean xerohalophyte species *Atriplex halimu*s (L.) differ in their ability to accumulate proline and glycine betaine in response to salinity and water stress. J. Exp. Bot. **59**:1315-1326.

Bele, K. N., G .R. Shamkumar, R. D. Deotale and A. S. Deshmukh, 2021. Screening of rice genotypes for morpho- characters and yield. J. Soils and Crops. **28(2):** 345-349.

Chen, T. H. and N. Murata, 2008. Glycine betaine: an effective protectant against abiotic stress in plants. Trends Plant Sci. **13**:499-505.

Chu, T. M., D. Aspinall and L. E. Paleg, 1976. Stress Metabolism. VII Specific ion effects on proline accumulation in barley. Aust. J. Plant Physiol. **3**:503-511.

Debouba, M., H. Gouia, A. Suzuki and M. H. Ghorbel, 2006. NaCl stress effects on enzymes involved in nitrogen assimilation pathway in tomato (*Lycopersicon esculantum* L.) seedlings. J**.** Plant Physiol. **163**:1247-1258.

Gill, S. S. and N. Tuteja, 2010. Reactive oxygen species and antioxidant machinery in abiotic stress tolerance in crop plant. Plant Physiol. Biochem. **48**:909-930.

Gomathi, R., G. Manohari and P. Rakkiyapan, 2012. Antioxidant enzymes on cell membrane integrity of sugarcane varieties differing in flooding tolerance. Sugar Tech. **13:**261-265.

Hare, P. D. and W. A. Cress, 1997. Metabolic implications of stress-induced proline accumulation in plants. Plant Growth Regul. **21**:79-102.

Heath, R. L. and Packer, 1968. Photoperoxidation in isolated chloroplasts : I Kinetics and stoichiometry of fatty acid peroxidation. Arch. Biochem. Biophys. **125**:189**–**198.

Huang, J., R. Hirji, L. Adam, K. L. Rozwadowski, J. K. Hammerlindl, W. A. Keller and G. Selvaraj, 2000. Genetic engineering of glycine betaine production toward enhancing stress tolerance in plants: metabolic limitations. Plant Physiol. **122**:747-756.

Jackson, M.L. 1973. Soil Chemical Analysis. Prentice Hall of India Pvt. Ltd., New Delhi, 69-182.

Joshi, G.V. and G. R. Naik, 1977. Salinity effect on growth and photosynthetic productivity in sugarcane Var. Co.740. Indian Sugar, **27**:329-332.

Kavikishore, P.B., Z. Hongdou, G. U. Miao, Hu, C-AH. and D. P. S. Verma, 1995. Over expression of "1- pyroline-5-carboxylase synthetase increases proline production and confers osmotolerance in transgenic plants. Plant Physiol. **108**:1387- 1394.

Kishor, P.B.K., S. Sangam, R. N. Amrutha, P. S. Laxmi, K. R. Naidu and K. S. Rao, 2005. Regulation of proline biosynthesis, degradation, uptake and transport in higher plants: its implications in plant growth and abiotic stress tolerance. Curr. Sci. **88**:424-438.

Kumar, P., P. S. Deshmakh, R. K. Sairam, S. R. Kushvaha and P. Sing 2006. Biochemical and phonological evaluation of chickpea genotypes differing in drought tolerance. Indian J. Plant Physiol. **11**:166-171.

Mahajan, S. T., R. M. Naik and U. S. Dalvi, 2013. Assessment of biochemical markers in differentiating sugarcane genotype for salt tolerance. Sugar Tech. **15**:116-121.

Matysik, J., B. Alia Bhalu and P. Mohanty, 2002. Molecular mechanism of quenching of reactive oxygen species by proline under stress in plants. Curr. Sci. **82**:525-532.

McNeil, S.D., M. L. Nucciom and A. D. Hanson, 1999. Betaines and related osmoprotectants-Targets for metabolic engineering of stress resistance. Plant Physiol**. 120**:945-950.

Mohamed, H., A. A. Hady, M. Mahmoud and S. Abid, 2012. Association of oxidative stress components with resistance to flax powdery mildew. Trop. Plant Pathol. **37**:386-392.

More, P.V., B. B. Desai and U. D. Chavan, 1994. Effect of salt stress on biochemical parameters of sugarcane. J. Maharashtra Agric. Univ. **19**:431-433.

Nakano, Y. and K. Asada, 1998. Hydrogen peroxidase scavenged by ascorbate specific peroxidase in spinach chloroplast. Plant Cell Physiol. **22**:867**–**880.

Park, E. J., Z. Jeknic, M. T. Pino, N. Murata and T. H. Chen, 2007. Glycine betaine accumulation is more effective in chloroplasts than in the cytosol for protecting transgenic tomato plants against abiotic stress. Plant Cell Environ. **30**:994-1005.

Prabat, J. M., R. L. Parate, S. B. Bramhankar, D. G. Wasnik, R. R. Kalaskar and P. N. Rokhande, 2018. Screening of groundnut genotypes against tikka disease. J. Soils and Crops. **28**(2):366-373.

Rasool, S., A. Ahmad, T. O. Siddiqi and P. Ahmed, 2013. Changes in growth, lipid peroxidation and some key antioxidant enzymes in chickpea genotypes under salt stress. Acta Physiol. Plant. **35**:1039-1050.

Richards, L. A., 1968. Diagnosis and improvement of saline and alkaline soils. Agricultural Handbook No. 60, United State Salinity Laboratory Staff, Oxford and IBH Publ. Co. Calcutta.1-156.

Sairam,R.K., K. Veerabhadra Rao and G. S. Srivastava, 2002. Differential response of wheat genotypes to long term salinity stress in relation to oxidative stress, antioxidant activity and osmolytes concentration. Pl. Sci. **163** : 1037-1046.

Satbhai, R. D. and R. M. Naik, 2014. Osmolytes accumulation, cell membrane integrity, and antioxidant enzymes in sugarcane varieties differing in salinity tolerance. Sugar Tech. **16**:30-35.

Sawhney, S. K., M. S. Naik and D. J. D. Nicholas, 1978. Regulation of nitrate reductase by light ATP and mitochondrial respiration in wheat leaves. Nature, **272:**647**–**648.

Sairam, R.K. and A. Tyagi, 2004. Physiology and molecular biology of salinity stress tolerance in plants. Curr. Sci**. 86:**407-421.

Sen, H.S., 2003. Problem soils in India and their management. Prospect and Retrospect. J. Indian Soc. Soil Sci. **51**:388-408.

Smirnoff, N. and Q. J. Cumbes, 1989. Hydroxyl radical scavenging activity of compatible solutes. Phytochem. **28**:1057-2060.

Stewart, G. R. and J. A. Lee, 1974. The role of proline accumulation in halophytes. Planta, **120**: 279-289.

Stumpf, D.K., 1984. Quantification and purification of quaternary ammonium compounds from halophyte tissue. Plant Physiol. **75**:273**–**274.

Taishi, V., S. Katsuyoshi, K. Morino and K. Takao, 2000. Enhancement of salt tolerance in soybean with NaCl pretreatment. Physiol. Plant. **110:**59-63.

Tiwari, T. N., S. K. Singh, S. Sharma, R. R. Singh, D. N. Singh and S. B. Singh, 2006. Performance of sugarcane varieties under sodic soil. Cooperative Sugar, **37:** 45-49.

Vasantha, S. and R. Rajalakshmi, 2009. Progressive changes in biochemical characters of sugarcane genotypes under salinity stress. Indian J. Plant Physiol. **14**:34-38.

Vranova, E., D. Inze and F. Van Breusegem, 2002. Signal transduction during oxidative stress. J. Exp. Bot. **53**:1227-1236.

Rec. on 03.03.2022 & Acc. on 07.05.2022