International Journal of Life Science
International Peer Reviewed Open Access Refereed Journal

Int. J. of Life Science, 2019; 7 (2):343-350
ISSN:2320-7817(p) | 2320-964X(o)

Original Article Open Access

Effect of Salt stress on plant growth and osmolyte accumulation in Black gram (*Vigna mungo* L.) seedlings

Sunanda CH[1] and Ranganayakulu GS[1]*

Department of Botany, Rayalaseema University, Kurnool-518007, Andhra Pradesh, INDIA.
*Corresponding authors: Dr.G. S. Ranganayakulu, Tel. (+91) 9989239242 E-mail: gsranganaayakulu@gmail.com

Manuscript details:

Received: 18.04.2019
Accepted: 05.05.2019
Published: 25.06.2019

Editor: Dr. Arvind Chavhan

Cite this article as:
Sunanda CH and Ranganayakulu GS (2019) Effect of Salt stress on plant growth and osmolyte accumulation in Black gram (*Vigna mungo* L.) seedlings, *Int. J. of Life Science*, Volume 7(2): 343-350.

Available online on http://www.ijlsci.in
ISSN: 2320-964X (Online)
ISSN: 2320-7817 (Print)

ABSTRACT

The effects of salt stress on seedling growth (shoot, root length and biomass), accumulation of osmolytes such as proline, glycine betaine, soluble sugars, free amino acids and polyamines were investigated in two commonly grown Black gram cultivars (Cv. Maruthi and Cv. LBG) during stress period. Seeds were grown at different concentrations of NaCl stress [(0.0 (control), 50, 100 and 150mM] for seven days. Salt stress resulted reduced seedling growth in all cultivars. Nevertheless, the rate of decrease in seedling growth (shoot, root growth and biomass) was significantly more in cv. LBG than cultivar Maruthi. The lowest effect of salinity on seedling growth was found in cv. Maruthi. Salt stress results showed genotypic differences in an accumulation of osmolytes during stress period. The level of osmolytes accumulation was significantly increased with increasing stress severity and duration in all studied cultivars. However, the magnitude increase of osmolytes accumulation was found to be very higher in cv. Maruthi and low in cv. LBG. The osmolyte accumulation in relation to the salt tolerance of these cultivars has been discussed.

Key words: Growth analysis, osmolyte accumulation, tolerant, salt stress, black gram

INTRODUCTION

Soil salinity is one of the most important agricultural problems in arid and semi-arid regions in different parts of the world. Nearly 20-40 % of the world's cultivated area and half of the world's irrigated lands are affected by salinity (Rhodes and Loveday 1990; Flowers, 2004). The economic prosperity of nation like India, where a majority of population is primarily dependent on agriculture, depends on crop productivity. Soil salinity creates extremely unfavorable conditions, and leads to inhibition of plant growth and development. In response to various environmental stresses such as salt and drought, different plant species have developed different physiological and biochemical mechanisms to cope or to tolerate stress (Cushman and Bohnert, 2000; Munns, 2003). The osmotic adjustment, i.e. reduction of cellular osmotic potential by net solute accumulation, has been considered as an important mechanism to salt and drought tolerance in plants. This reduction in osmotic potential in salt tolerant plants can be result of an accumulation of osmolytes such as proline, glycine betaine, soluble sugars, free amino acids and polyamines (Hasegawa et al., 2000; Rontein et al., 2002, Jouve et al., 2004). These osmoprotectants can serve to raise osmotic pressure in the cytoplasm and can also stabilize proteins and membranes when salt levels or temperatures are unfavorable, therefore osmoprotectants can play an important role in the adaptation of cells to various adverse environmental conditions (McNeil et al., 1999). An understanding of these mechanisms of plant stress tolerance will lead to effective means to be breed or genetically engineered salt tolerant crops.

Although the relationship between osmolyte regulation and tolerance is not clear, there is evidence that the osmotic adjustment appears, at least partially, to be involved in the salt tolerance of certain plant genotypes (Giridarakumar et al., 2003).

Black gram (*Vigna mungo* L.) is one of the important pulse crop in India. This crop comes up reasonably well in drought prone areas, where other crops invariably fail to grow. Information is lacking regarding to the relative levels of salt tolerance among the existing Black gram cultivars. Therefore, the objective of the present study was to evaluate the effects of salt stress on plant growth and osmolyte accumulation in two different Black gram cultivars. Further, these lines are using in breeding for salt tolerance; and is seems to be effective and economic improvement.

MATERIALS AND METHODS

Experimental design

The seeds of Black gram (*Vigna mungo* L. cv. Maruthi and LBG) were procured from Acharya N.G. Ranga Regional Agricultural Research Station, Nandyal, Kurnool District, Andhra Pradesh, India. The healthy seeds were surface sterilized with 0.1% mercuric chloride (w/v) solution for 1 minute and thoroughly rinsed with distilled water and germinated in Petri plates lined with filter papers. Salt stress was induced by using sodium chloride solution at different concentrations (50, 100 and 150mM) prepared from half strength Hoagland's nutrient solution. Half strength nutrient solution alone served as control. The Petri plates were kept at room temperature of 25± 4^{o}C in dark for seven days. After seven days total seedlings were used for experimental analysis.

The growth of seedlings in terms of shoot and root length was measured and recorded. For the determination of mass seedlings were dried at 80 ^{0}C in a hot air oven for 48 h and then dry mass were recorded.

Free proline content was estimated in both control and stressed seedlings by (Bates et al., 1973) using ninhydrin reagent. Quaternary ammonium compounds were measured as glycine betaine according to Grieve and Grattan (1983). Soluble sugars were extracted following the method of Highkin and Frankel (1962), boiled 0.5g of dried tissue in 80% ethanol at 80 ^{0}C for 30 and then centrifuged. Supernatant was used for the estimation of total soluble sugars following Nelson's method (Nelson, 1944) as modified by Somogyi (1952).

The extraction and estimation of amino acids were determined by spectrophotometrically according to Moore and Stein (1948) and total polyamine content was estimated according to Seiler and Wiechmann (1967).

Statistical Analysis

The data obtained in all cases were subjected to analysis of variance (ANOVA) and the mean values were compared by Duncan's Multiple Range (DMR) test at 0.05% level (Duncan, 1955).

RESULTS

Seedling growth

The growth of the seedlings in terms of root and shoot length of all studied cultivars in control and stressed plants were measured and results were depicted in figure 1A and 1B. Salt stress showed significant inhibition of root and shoot length in all studied cultivars. However the percent inhibition was high in cv. LBG and low in cv. Maruthi

Biomass accumulation

The fresh weight and accumulated biomass in terms of dry weight was studied in two Black gram cultivars under control and salt stressed seedlings and the results were presented in figure 2A and 2B. From the figure it is found that the seedling biomass was decreased in salt stressed plants compared to their respective controls. The magnitude of decline in seedling biomass accumulation was found to be dependent on the stress severity and duration. However, the percent reduction in biomass yield was less (54%) in cv. Maruthi and more (70%) in cv. LBG at 150mM NaCl concentration compared to other cultivars.

Free Proline content

Free proline content was estimated in both control and NaCl stressed seedlings of six Black gram cultivars and data were presented in Figure 3. The free proline content was significantly increased in stressed plants over control plants. Significant differences were observed in free proline accumulation among the studied cultivars by about 3.7 fold in (cv. Maruthi) and 2.5 fold in (cv. LBG) at 150mM NaCl stress when compared to their respective controls. However, the percent increase was comparatively high in cv. Maruthi and low in cv. LBG.

Glycine betaine

The level of glycine betaine content was significantly increased in two cultivars during stress. However, the level of increase was significantly varied between the cultivars (Figure 4). The magnitude of elevation in glycine betaine content was found to be higher in cv. Maruthi all stress levels when compared to cv. LBG.

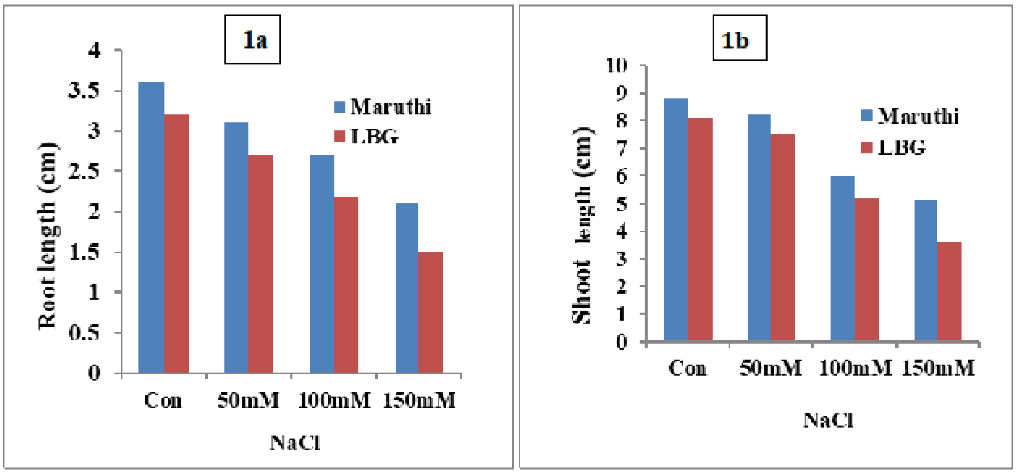


Figure 1 : Root length and shoot length in 7-day old seedlings of two black gram cultivars under control and NaCl stress. (a) Root length (cm) and (b) shoot length (cm). Values are mean from five replications. Vertical bars indicate ±S.D.

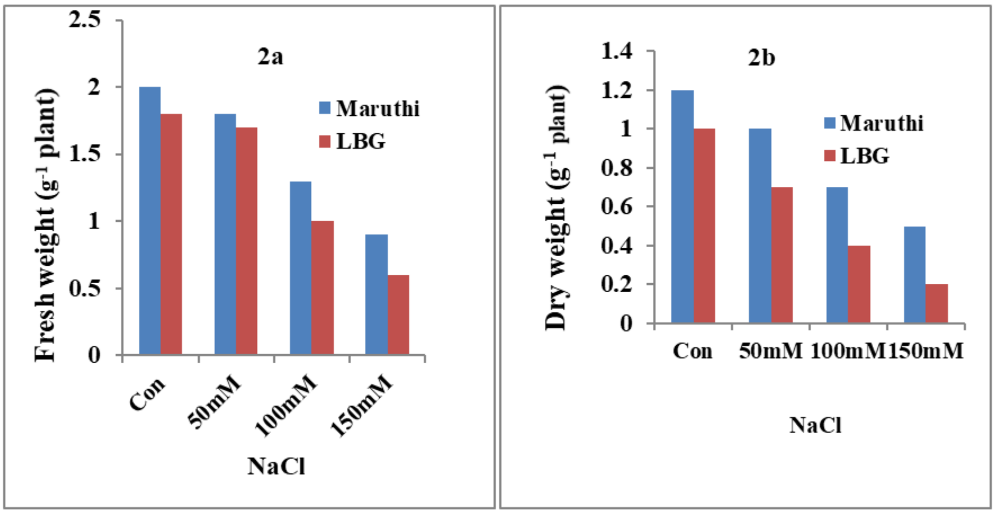


Figure 2 : Fresh weight and dry weight in 7-day old seedlings of two black gram cultivars under control and NaCl stress. (a) Fresh weight (g^{-1} plant) (b) Dry weight (g^{-1} plant). Values are mean from five replications. Vertical bars indicate ±S.D.

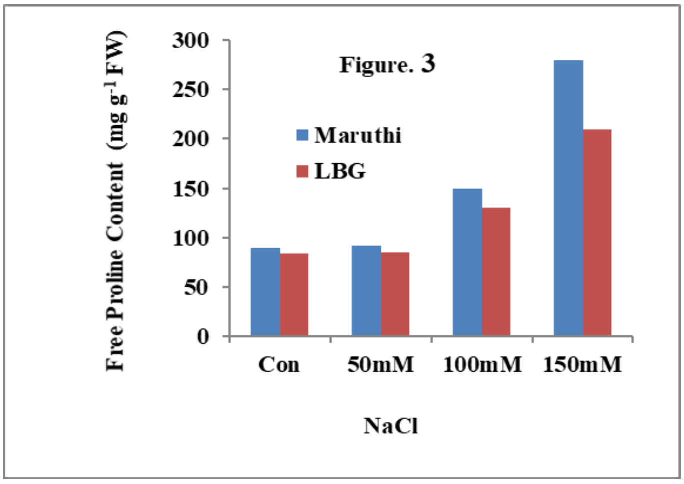
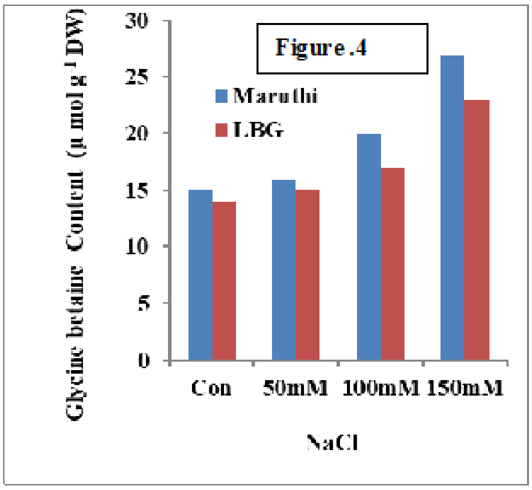


Figure 3: Free proline content (mg g^{-1} FW) in 7-day old seedlings of black gram cultivars under control and NaCl stress. Values are mean from five replications. Vertical bars indicate ±S.D.



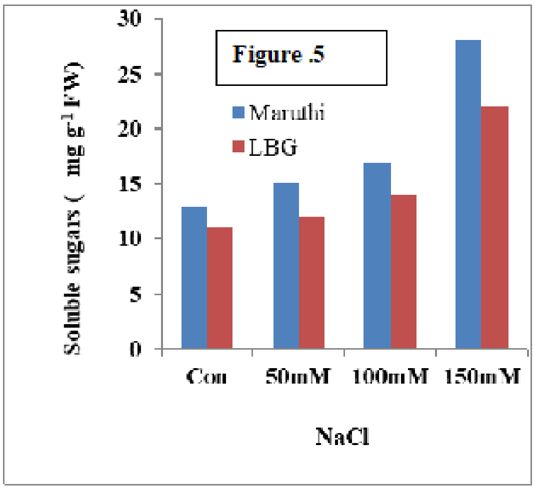
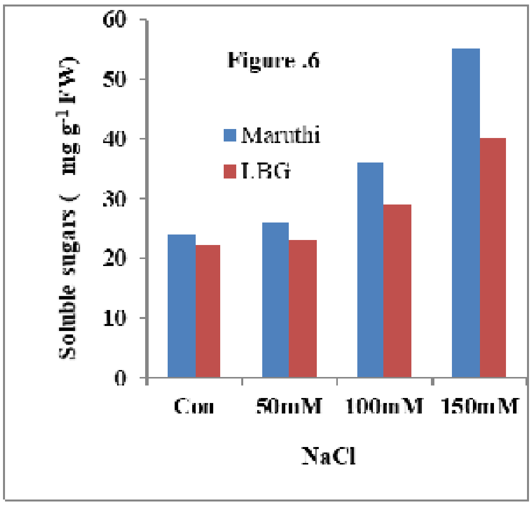


Figure 4 &5: Levels of quaternary ammonium compounds (glycine betaine equivalents) (µ mol g^{-1} DW) and Levels of Soluble sugars (mg^{-g} FW) in 7-day old seedlings of two black gram cultivars under control and NaCl stress. Values are mean from five replications. Vertical bars indicate ±S.D.



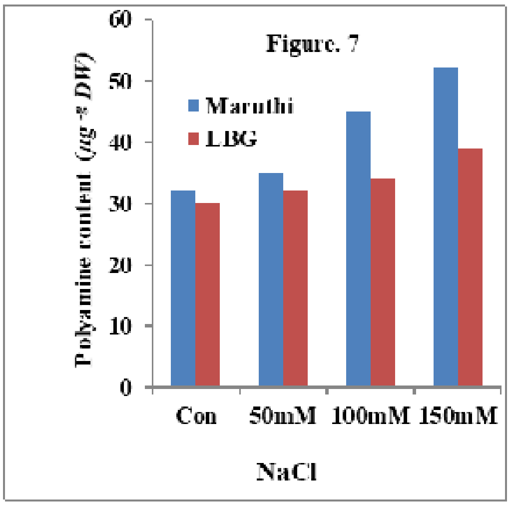


Figure 6&7: Total amino acid content (mg $^{-g}$ DW) and Total polyamine content (µg $^{-g}$ DW) in 7-day old seedlings of two black gram cultivars under control and NaCl stress. Values are mean from five replications. Vertical bars indicate ±S.D.

Soluble sugars

Total soluble sugar content was increased with increasing stress severity in two black gram cultivars results were depicted in (Figure 5). However, the degree of increase was varied between the cultivars. There was, by about 3.7 fold in (cv. Maruthi), and 2.5 fold in (cv. LBG) at 150mM NaCl stress.

Total amino acids

The pool sizes of amino acid levels were increased significantly in two black gram cultivars at all stress levels results were depicted in (Figure 6). However, the degree of increase in free amino acid contents was more in cv. Maruthi compared to cv.LBG. The total amino acids were found to be 2.4 fold in cv. Maruthi and 2.1 fold in cv. LBG at 150mM NaCl stress.

Total Polyamines

Total polyamine content was increased significantly with increasing stress severity in two cultivars results were depicted in (Figure 7). Nevertheless, a difference in the accumulation of total polyamine content was observed between the cultivars. The percent increase of polyamine content was found to be higher in cv. Maruthi at (100 and 150mM) compared to cv.LBG.

DISCUSSION

Salinity has a dual effect on plant growth via an osmotic effect on plant water uptake, and specific ion toxicities. By decreasing the osmotic potential of the soil solution, plant access to soil water is decreased, because of the decrease in total soil water potential. But there is a general agreement that the whole plant growth responses to salinity are multigenic and that a better knowledge of the underlying physiology is required in order to understand why some plant species and varieties are more salt resistant than others. However, this is a complex task since plant growth responses to salinity can vary with degree and duration of stress, plant organ, variety or species and developmental stage (Neumann, 1997). It has been shown that the stress caused by salts present in the soil, alters water status and brings about initial growth reduction of the plant (Yeo 1998, Munns, 2003). One of the classic manifestations of salt stress in many plant species is marked reduction in plant height, due to the osmotic effects of the salt outside the roots and that distinguishes a salt-susceptible plant from a more tolerant one (Munns et al., 1995).

Similarly, in the present study we recorded the reduced growth (root length, shoot length and biomass) during stress conditions and degree of reduction in seedling growth was dependent on severity of stress and species. It revealed that the 150mM NaCl stress treatment has caused significant reduction in two black gram cultivars, but more pronounced reduction was found in cv. LBG and less in cv. Maruthi (Figure 1A &1B). Several investigators (Zidan et al., 1990; Mishra et al., 1996; Claudivan et al., 2006; Veeranagamallaiah et al., 2007) reported the reduced growth in different plant species under salt stress. This reduced growth under salinity stress has been ascribed either due to osmotic or ionic effects; inhibition of cell division and cell elongation process associated with the growth of the seedling and decrease in plastic extensibility of the growing cell walls (Veeranagamallaiah et al., 2007).

The accumulation of compatible solutes may help to maintain the relatively high water content obligatory for plant growth and cellular functions. Osmotic and oxidative stress induced by salinity could be reduced by the production and accumulation of compatible solutes and is known to play a key role in plats for increasing growth and productivity of crop plants subjected to adverse environmental conditions such as salinity and drought. The levels of osmoprotectants increased during exposure to stresses such as salinity, water deficit, and low temperature (McNeil et al., 1999). The prequently observed metabolites with an osmolyte function are proline, glycine betaine, soluble sugars free amino acids and polyamines.. The frequently observed metabolites with an osmolyte function are proline, glycine betaine, soluble sugars, free amino acids and polyamines.

The accumulation of proline is an early response to salt stress (Fedina et al., 2002). Several investigators (Delauney and Verma, 1993; Kavikishor et al., 1995) have demonstrated that the positive correlation was found between the accumulation of proline and osmoprotective role during stress at the whole plant level and cell cultures. However convincing evidence is still lacking as to whether accumulation of proline can provide any biochemical adaptation for plants during stress. Ramanjulu and Sudhakar (2000), Giridarakumar et al., (2003) Veeranagamallaiah et al., (2007) observed genotypic differences in proline accumulation and a positive correlation between magnitude of free proline accumulation and tolerance to salt and water stress. Similarly in the present study we have noticed a positive correlation between salt stress and free proline accumulation between the two Black gram cultivars (Figure 3). However a greater accumulation of free proline content (3.7 fold) was found in cv.Maruthi whereas cv.LBG showed lesser accumulation (2.0 fold). These results were further in agreement with Silveira et al., (2003) Giridarakumar et al., (2003) Khedr et al., (2003) .though free proline accumulation has been suggested as an index for determining salt tolerance potentials between the cultivars (Sudhakar et al., 1993; Igarashi et al., 1997; Giridarakumar et al., 2003), Very few reports are controversial to the above results and in this way Lutts et al., (1999) reported that salt sensitive cultivars accumulated significantly higher levels of proline accumulation compared to the tolerant ones.

Glycine betaine is regarded as an effective compatible solute that accumulates in chloroplast of plants when exposed to environmental stresses (Sawahel, 2003). Here we report a positive correlation between glyine betaine accumulation and NaCl stress in two Black gram cultivars (Figure 4). An increased accumulation was higher in cv. Maruthi (2.0 fold) at 150mM and cv. LBG was found to be registred least account (1.5 fold). Several investigators have noticed that accumulation of glycinebetaine under salt stress was found to be high in salt tolerant species (Sudhakar et al., 1993; Jagendorf and Takabe, 2001; Giridarakumar et al., 2003). Besides osmoregulation, glycine betaine stabilizes the oxygen evolving activity of photosystem-II, protein complexes at high concentration of NaCl. The major role of glycine betaine might be to protect membranes and macromolecules from damaging effects of stress (Sawahel, 2003).

Soluble sugars have been specified as potential osmoregulators (Raggi, 1994). Elevated sugar levels relative to control in salt stressed plants may contribute the turgor maintenance (Sacher and Staples, 1985). In the present study the amount of soluble sugars was relatively higher in tolerant cv.Maruthi and lesser in cv.LBG at severe stress treatments (Figure 5). In analogy, several investigators noticed that soluble sugar levels were increased with increased levels of salt stress (Sairam and Tyagi, 2004; Ashraf and Harris, 2004). Furthermore, Jouve et al., (2004) observed a higher accumulation of soluble sugars in aspen at 150mM NaCl stress.

Salt stress induced changes in amino acid content play an important role in response to salt stress, since these relations were obtained several time in relationship with stress tolerance by Livia et al., (2002) in cereal plants. Survival and growth of plants in saline environments is the result of adaptive processes such as ion transport and compartmentation of osmotic solutes, synthesis and their accumulation is lead to the osmotic adjustment and protein turnover for cellular repair (Munns and Termaat, 1986). In the present study we have noticed that a variation in the accumulation of amino acid levels adjusting between the cultivars, the extent of increase was greater in cv. Maruthi compared with cv. LBG (Figure 6). Similar results were obtained Livia et al., (2002) in cereal plants, Ramanjulu and Sudhakar (1997) in mulbarry. Varietal differences in the accumulation of amino acids have been taken as an index for determining the salt tolerant potentials of many crops. Improved levels of free amino acids together with organic acids and quaternary ammonium compounds serve as compatible cytoplasmic solutes to maintain the osmotic balance under stress conditions (Dubey, 1994).

Polyamines are known to be involved in various cellular processes (Rajam et al., 1998) and they are ubiquitous aliphatic amines that are implicated in many aspects of plant growth and developments in a wide range, and play an important role in stabilizing the plasma membrane under salt stress condition (Galston and Sawhney, 1995). Effect of salt stress on polyamine metabolism is not always clear, since differences in polyamine accumulation in response to salt stress have been reported among and within the species (Prakash et al., 1988; Pedro et al., 2004). Similarly in the present study we have observed a positive correlation between salt tolerance and accumulation of total polyamines between the two Black gram cultivars (Figure 7). However, a greater accumulation was found in cv. Maruthi and lesser accumulation in cv. LBG. Similarly, Chattopadhaya et al., (2002), Zapata et al., (2004) noticed a higher accumulation of polyamine content with varying levels in seven different plant species under salinity stress.

Finding markers of stress tolerant and susceptible lines in commonly cultivated Black gram cultivars under salinity could be useful for plant breeding programmers. From the present study it is clear that cv.Maruthi possesses relatively better salt tolerant nature compared to cv.LBG based on higher accumulation of osmolytes such as proline, glycine betaine, amino acids, soluble sugars and total polyamines. Interesting futures have been found through these results, must be related to salt stress response and should be considered as general salt stress reaction markers for black gram.

In conclusion we evaluate the cv.Maruthi has salt tolerant one and cv.LBG has susceptible cultivar taken into consideration in this study. And further research is needed to unravel the tolerance potentials of these two black gram cultivars at molecular level.

Acknowledgements

We are thankful to Prof. Chinta Sudhakar, S.K. University, Anantapur for providing lab and instrumental facility

REFERENCES:

Ashraf M, Harris PJC (2004) Potential biochemical indicators of salinity tolerance in plants. Plant Sci. 166: 3- 46.

Bates LS, Waldorn RP, Teare ID (1973) Rapid determination of free proline for water stress studies. Plant and Soil. 39: 205-208.

Chattopadhaya MM, Tiwari BS, Chattopadhyay G, Bose A, Sengupta DN, Ghosh B (2002) Protective role of exogenous polyamines on salinity-stressed Oriza sativa plants. Physiol. Plnat. 116:192-199.

Claudivan FL, Jose OAJ, Luiz CALF, Teogenes SD, Francisco VAG, Eneas GF, Jose TP, Marlos B (2006) Morpho-physiolgical responses of cowpea leaves to salt stress. Braz.J.Plant Physiol, 18 (4):455-465.

Cushman JC, Bohnert HJ (2000) Genomic approaches to plant stress tolerance. Curr. Opin. Plant Biol. 3:117-124.

Delauney AJ, Verma DPS (1993) Proline biosynthesis and osmoregulation in plants. Plant J. 4: 215-223.

Dubey RS (1994) Protein synthesis by plants under stressful conditions. In: Mohammad P (eds), Hand book of plant and crop stress. pp.277-299. Marcel Decker Inc, New York.

Duncan M (1955) Multiple range and multiple tests. Biometrics 42:1-47.

Fedina IS, Georgieva K, Grigorova I (2002) Light-dark changes in proline content of barley leaves under salt stress. Biol. Plant. 45(1): 59-63.

Flowers TJ (2004) Improving crop salt tolerance. J. Exp. Bot. 55: 307-319.

Galston AW, Kaur-Sawhney R (1995) Polyamines as endogenous growth regulators. In: Davies, P.J. (eds), Plant Hormones: Physiology, Biochemistry and Molecular Biology, pp.158-178. Kluwer Academic Publishers, Dordrecht.

Giridarakumar S, Matta Reddy A, Sudhakar C (2003) NaCl effects on proline metabolism in two high yielding genotypes of mulberry (Morus alba L.) with contrasting salt tolerance. Plant Sci. 165: 1245-1251.

Grieve CM, Grattan SR (1983) Rapid assay for determination of water soluble quaternary ammonium compounds. Plant and Soil. 70: 303-307.

Hasegawa PM, Bressan RA, Zhu, JK Bohnert, H J (2000) Plant cellular and molecular responses to high salinity. Annu. Rev. Plant Physiol. Plant Mol. Biol., 51: 463-499.

Highkin HR, Frankel F (1962) Studies of growth and metabolism of barley mutant lacking chlorophyll. Plant Physiol. 37: 814-820.

Igarashi Y, Yoshiba I, Yamaguchi-Shinozaki K, Wada K, Shinozaki K (1997) Characterization of the gene for delta 1-pyrroline-5-carboxylate synthetase and correlation between the statement of the gene and salt tolerance in (Oryza sativa L.). Plant Mol. Biol. 33: 857-865.

Jagendorf AT, Takabe T (2001) Inducers of glycine betaine synthesis in barley. Plant. Physol. 127: 1827-1835.

Jouve L, Hoffmann L, Hausman JF (2004) Polyamine, carbohydrate, and proline content changes during salt stress exposure of aspen (Populus tremila L.). Involvement of oxidation and osmoregulation metabolism. J. Plant Biol. 6: 74-80.

Kavikishor PB, Hong Z, Miao GH, Hu CA, Verma DPS (1995) Overstatement of D1-pyrroline-5-carboxylate synthetase increases proline production and confers osmotolerance in transgenic plants. Plant Physiol. 108: 1387-1394.

Khedre AHA, Mohammd AA., Amal A.A.W, Paul Quick W, Abogadallah GM (2003) Proline induces the expression of salt-stress-responsive proteins and may improve the adaptation of (Pancratium maritimum L.) to salt stress. J. Exp. Bot. 54: 2553-2562.

Livia SS, Gabor K, Zoltan S (2002) Effect of salt stress on free amino acid and polyamine content in cereal. Acta Biologica Szegediensis. 46: 73-75.

Lutts S, Majerus V, Kinet JM (1999) NaCl effects on proline metabolism in rice (Oryza sativa L.) seedlings. Physiol. Plant. 105: 450-458.

McNeil SD, Nuccio M, LHanson AD (1999) Betaines and related osmoprotectants. Targets for metabolic engineering of stress resistance. Plant Physiol. 120(4): 945-950.

Mishra SN, Singh DB, Chaudhary A (1996) Native and ammonium effect on Indian mustard seedlings grown under salinity stress. Indian J. Plant Physiol. 1: 93-97.

Moore S, Stein WH (1948) Photometric methods for use in the chromatography of amino acids. J. Biol. Chem. 176: 307-318.

Munns R (2003) Physiological processes limiting plant growth in saline soils: some dogmas and hypotheses. Plant Cell Environ. 16: 15-24.

Munns R, Termaat A (1986) Whole-plant responses to salinity. Aust. J. Plant Physiol. 13: 143-160.

Munns, SR, Schachtman, DP, Condon, AG (1995) The significance of a two-phase growth response to salinity in wheat and barley. Aus. J. Plant Physiol. 22: 561-9.

Nelson N (1944) A photometric adaptation of the somogyi method for the determination of glucose. J. Biol.Chem.153: 375-380.

Neumann PM (1997). Salinity resistance and plant growth revisited. Plant Cell and Environ. 20: 1193-119.

Pedro JZ, Maria S, Terasapretel M, Asuncion A, Angeles Botella M (2004) Polyamines and ethylene changes during germination of different plant species under salinity. Plant Sci. 167:781-788.

Prakash L, Prathapasenan G (1988) Putrescine reduceds NaCl-induced inhibition of germination and early seedling growth of rice (Oriza sativa L.). Aust. J. Plant Physiol. 15: 761-767.

Raggi V (1994) Changes in free amino acids and osmotic adjustment in leaves of water-stressed bean. Physiol. Plant. 91: 427-437.

Rajam MV, Dagar S, Waie B, Yadav JS, Kumar PA., Shoeb F, Kumaria R (1998) Genetic engineering of polyamine and carbohydrate metabolism for osmotic stress tolerance in higher plants. J. Bio Sci. 23: 473-482.

Ramanjulu S, Sudhakar C (1997) Drought tolerance is partly related to amino acid accumulation and ammonia assimilation: A comparative study in two mulberry genotypes differing in drought sensitivity. J. Plant Physiol. 150, 345-350.

Ramanjulu S, Sudhakar C (2000) Proline metabolism during dehydration in two mulberry genotypes with contrasting drought tolerance. Plant Physiol. 157: 81-85.

Rhodes JD, Loveday J (1990) Salinity in irrigated agriculture. In American society of civil Engineers, Irrigation of Agricultural Crops (Steward, B. and Nielsen, D.R., Neds), Am. Soc.Agronomist, Monograph. 30:1089-1142.

Rontein D, Basset G, Hanson A.D (2002) Metabolic engineering of osmoprotectant accumulation in plants. Metabolic Engineering. 4: 49-56.

Sacher RF, Staples RC (1985) Inositol and sugars in adaptation of tomato to salt. Plant Physiol. 77: 206-210.

Sairam RK, Tyagi A (2004) Physiology and molecular biology of salinity stress tolerance in plants. Curr. Sci. 86: 407-421.

Sawahel W (2003) Improved performance of transgenic glycine betaine-accumulating rice plants under drougt stress. Biol. Plant. 47: 39-44.

Seiler N, Wiechmann M (1967) Die mikrobestimmung von spermin and spermidine als 1-dimithylaminonapthali-5-sulpho-derivative. Int. J. Plant Physiol. Chem. 348: 1285-1290.

Silveira JAG, De Almeida VR, Da Rocha IMA, De Oliveira Monteiro Moreira AC, De Azevedo Moreira A, Oliveria JTA (2003) Proline accumulation and glutamine synthetase activity are increased by salt-induced proteolysis in cashew leaves. J. Plant Physiol. 160: 115-123.

Somogyi M (1952) Notes on sugar determination. J. Biol. Chem. 195:19-24.

Sudhakar C, Reddy PS, Veeranjaneyulu K (1993) Effect of salt stress on enzymes of proline synthesis and oxidation in green gram (Phaseolus aures Roxb.) seedlings. J. Plant Physiol. 141: 621-623.

Veeranagamallaiah G, Chandraobulreddy P, Jyothsnakumari G, Sudhakar C (2007). Glutamine synthetase expression and pyrroline-5-carboxylate reductase activity influence proline accumulation in two cultivars of foxtail millet (Setaria italica L.) with differential salt sensitivity. Envi. Exp. Bot, 60:239–244.

Yeo AR (1998) Molecular biology of salt tolerance in the context of whole-plant physiology. J. Exp. Bot., 49: 915-929.

Zapata PJ, Serrano M, Teresapretel M, Amoros A, Angelesbotella M (2004) Polyamines and ethylene changes during germination of different plants species under salinity. Plant. Sci. 167: 781.

Zidan I, Azaizeh H, Neumann PM (1990) Doe's salinity reduce growth in maize root epidermal cells by inhibiting their capacity for cell wall acidification. Plant Physiol. 93: 7-11.