RESEARCH ARTICLE
Legume Research- An International Journal, Volume 45 Issue 4: 429-434 (April 2022)

Antioxidant Responses of Ricebean [*Vigna umbellata* (Thunb.) Ohwi and Ohashi] Seedling under Iso-osmotic Potential of Salinity and Drought Stress

Kousik Atta, Jahnavi Sen, Pravachan Chettri, Anjan Kumar Pal
10.18805/LR-4551

ABSTRACT

Background: Salinity and drought are the major abiotic stresses and both can cause osmotic imbalances. Drought stress directly results in osmotic stress whereas salinity problem firstly disrupts the water balance and eventually induces ion toxicity which results in cyto-toxicity, metabolic impairment, nutrient imbalance and finally poor crop growth and yield. The co-ordinated up-regulation or constitutive expression of antioxidative system in plants is the main defense in plant against these stresses and thus the present experiment was undertaken to study the antioxidant responses under drought and salinity stress at seedling stage in ricebean (Bidhan 1).
Methods: For studying the effect of iso-osmotic potential of salinity and drought stress solutions of NaCl and PEG 6000 with -0.2 MPa (50 mM NaCl and 10% PEG), -0.4 MPa (100 mM NaCl and 12% PEG) and -0.8 MPa (200 mM NaCl and 18% PEG) osmotic potential were used. The experiment was done in the laboratory of Department Plant Physiology, Bidhan Chandra Krishi Viswavidyalaya (BCKV), Mohanpur, Nadia and West Bengal in the year 2017-18 and 2018-19.
Result: Under moderate to high intensity of osmotic stresses the leaf proline content decreased. The mild and medium stress treatments induced much higher activity of GPOX and APX in the leaf which then decreased somewhat as the intensity of stress increased. The experiment showed that drought stress was found to produce more drastic effects on seedling growth in ricebean as compared to the salinity stress at iso-osmotic potentials.

Key words: Ascorbate peroxidase (APX), Catalase (CAT), Drought stress, Guaiacol peroxidase (GPOX), Proline, Ricebean, Salinity stress.

INTRODUCTION

Salinity stress can affect plants initially by creating an osmotic stress then it induces ion toxicity that lead to cyto-toxicity, metabolic impairment, nutrient imbalance and finally death of the plant. Initially, the presence of salts in high concentration makes very difficult for plants to withdraw water from soil due to very low osmotic potential. Salinity stresses during initial phases decreases water absorption capacity of root systems and loss of water from leaves is accelerated due to osmotic stress of high salt accumulation in soil and plants and therefore it is also considered as hyper-osmotic stress (Munns, 2005). In effect, the plants suffer from a sort of osmotic stress which causes yield reduction. At the later stages of stress, due to the absorption of sodium and chloride ions in high concentration plants suffer from cyto-toxicity which result in reduction of growth, leaf burn and plant death. The presence of high concentration of Na^+ and Cl^- ion also reduces the availability of other ions like K^+, Ca^{2+}, Mg^{2+}, thus, causing nutritional disorders (Marschner, 1995). Drought is a meteorological term and defined as a period without significant rainfall. It normally occurs under depleting soil moisture and the intensity of drought increases under atmospheric conditions conducive to increased water loss by transpiration and/ or evaporation. Crop growth and yield unfavourably affected by water deficit, which is one of the major abiotic stress. Drought stress is characterized by reduction of water content, diminished leaf water potential and turgor loss, closure of stomata and decrease in cell enlargement and growth. Severe water stress may result in the arrest of photosynthesis, disturbance of metabolism and finally the death of plant (Jaleel *et al*., 2008). The reduction in fresh and dry biomass (Farooq *et al*., 2009) is a major adverse effect of water stress on different crops.

Department of Plant Physiology, Bidhan Chandra Krishi Viswavidyalaya, Nadia-741 252, West Bengal, India.

Corresponding Author: Kousik Atta, Department of Plant Physiology, Bidhan Chandra Krishi Viswavidyalaya, Nadia-741 252, West Bengal, India. Email: kousikatta1995@gmail.com

How to cite this article: Atta, K., Sen, J., Chettri, P. and Pal, A.K. (2022). Antioxidant Responses of Ricebean [*Vigna umbellata* (Thunb.) Ohwi and Ohashi] Seedling under Iso-osmotic Potential of Salinity and Drought Stress. Legume Research. 45(4): 429-434. DOI: 10.18805/LR-4551.

Submitted: 25-11-2020 **Accepted:** 05-07-2021 **Online:** 06-08-2021

Plant produces reactive oxygen species (ROS) continuously as bi-products of various metabolic pathways in different cellular compartments like chloroplast, mitochondria and peroxisome. Reactive oxygen species (ROS) are routinely produced during various physiological processes of the plants and it causes oxidative damage to membrane lipids, proteins, DNA and RNA. To protect the cells from oxidative damage, cells have evolved antioxidant defence which neutralizes, scavenges or dismutase ROS

(Singh *et al.* 2015). The equilibrium between the production and the scavenging of ROS may be perturbed by various biotic and abiotic stress factors, such as salinity, drought, heavy metals, UV radiation, pathogen attacks *etc.* These disturbances in equilibrium lead to sudden increase in intracellular levels of ROS leading to oxidative stress which can cause significant damage to cell structure. To protect themselves against these toxic oxygen intermediates, plant cells contain both enzymatic and non-enzymatic components. Among the enzymatic antioxidants, superoxide dismutase (SOD), catalase (CAT), ascorbate peroxidase (APX), guaiacol peroxidase (GPOX) and non-enzymatic low molecular metabolites, such as ASH, GSH, a-tocopherol, carotenoids and flavonoids (Gill *et al*., 2011 and Mittler *et al*., 2004) have been found to be very important. In addition, proline can now be added to an elite list of non-enzymatic antioxidants that microbes, animals and plants need to counteract the inhibitory effects of ROS (Chen and Dickman 2005).

The present experiment has been designed to study the effects of different levels of salinity and drought stress on some biochemical frame work of ricebean during seedling stage.

MATERIALS AND METHODS

Plant material

Seeds of ricebean [*Vigna umbellata* (Thunb) Ohwi and Ohashi] variety Bidhan-1 were used in the experiment. The seeds were collected from AICRP on Forage Crops, Kalyani Centre.

Experimental site

The experiment was done in the laboratory of Department Plant Physiology, Bidhan Chandra Krishi Viswavidyalaya (BCKV), Mohanpur, Nadia and West Bengal in the year 2017-18 and 2018-19.

Plant culture

For studying the effect of salinity and drought stress on ricebean the experiment was conducted in sand culture using modified Hoagland solution (Epstein, 1972) under laboratory condition of diffused light, at around 80±1% relative humidity (R.H.) and at a temperature of 28±1°C. For this purpose, the seeds of ricebean cultivar Bidhan-1 were surface sterilized with 0.1% (w/v) $HgCl_2$ for 3 minutes. The pre-germinated seeds (five seeds) were then transferred to plastic beakers of capacity one litre containing neutral sand. The seedlings were grown with supplement of full strength Hoagland solution (Epstein, 1972) maintaining the pH 6.3 at 3 days interval for 14 days.

Treatment application

14 day old seedlings were subjected to salinity and drought treatments. For this purpose, the appropriate amounts of NaCl and PEG 6000 calculated as per Sosa *et al.* (2005) (Table 1) to obtain the osmotic potential (Ψ) of -0.2, -0.4 and -0.8 MPa were mixed with modified Hoagland nutrient solution and the pH was adjusted to 6.3. Thus, the drought and salinity stress with iso-osmotic potentials were created in the present experiment. A control set having Ψ_s equivalent to 0.0 Mpa osmotic potential without containing NaCl or PEG. Observations on different growth and biochemical parameters were recorded on 9 days after treatment application.

Estimation of biochemical characters

Proline content was estimated using the standard protocol (Mohanty and Sridhar, 1982). Proline was extracted from 0.25 g fresh leaf tissue of both stressed and non-stressed seedlings. The absorbance was measured at 520 nm and proline content was calculated from the standard curve of L- proline.

GPOX activity was calculated as per Siegel and Galston (1967). GPOX was extracted from 0.25 g fresh leaf tissue of both stressed and unstressed seedlings. The absorbance was measured at 470 nm and GPOX activity was calculated from the standard curve.

APX activity was calculated as per Nakano and Asada (1981). APX was extracted from 0.25 g fresh leaf tissue of both stressed and non-stressed seedlings. The absorbance was measured at 290 nm and APX activity was calculated from the standard curve of H_2O_2.

Catalase activity was calculated as per Cakmak *et al.* (1993). Catalase was extracted from 0.25 g fresh leaf tissue of both stressed and control seedlings. The absorbance was measured at 240 nm and Catalase activity was calculated from the standard curve of H_2O_2.

Statistical analysis

The mean data in all the cases were subjected to statistical analysis following completely randomised design using INDOSTAT version 7.1 Software. The mean values were statistically compared by least significant difference (LSD) at $P \leq 0.05$.

RESULTS AND DISCUSSION

Proline (Pro) is the most important and efficient compatible organic solute (Tang *et al.* 2015), which act as an osmo-protectant. Along with its antioxidant properties, proline can act as a molecular chaperone to protect the structure of biological macromolecules during dehydration, thus conferring plant tolerance to environmental stresses (Kundur *et al.* 2016; Tang *et al.* 2015; Ashraf and Fooland, 2007). Apart from being an osmolyte for osmotic adjustment, proline contributes to stabilizing sub-cellular structures (*e.g.*, membranes and proteins), scavenging free radicals and buffering cellular redox potential under stress conditions. It may also act as protein compatible hydrotrope, alleviating cytoplasmic acidosis and maintaining appropriate $NADP^+$/

Table 1: Concentrations of salts and PEG 6000 required to obtain the desired osmotic potentials.

Ψ_s (MPa)	NaCl (mol L^{-1})	PEG 6000 (% w/v)
-0.2	0.05	10
-0.4	0.1	12
-0.8	0.2	18

NADPH balances compatible with metabolism (Strizov *et al.,* 1997). The leaf proline content at 50 mM NaCl and PEG 10% (-2 MPa osmotic stress) solution increased significantly over that of control (Fig 1). This result was in accordance with the findings of Dar *et al.* (2007), El-Sayed (2011) and Aniat-ul-Haq *et al.,* (2012) under salinity stress and with study of Mohammadkhani and Heidari (2008) and Bhardwaj and Yadav (2012) under drought stress. The increased level of proline in leaf might attribute for osmotic adjustment under low levels of osmotic shock induced by NaCl and PEG. The level of proline content decreases as the osmotic potential of the growing medium decreased more. Earlier, Verma *et al.* (2012) also observed a decrease in leaf proline at high level of NaCl stress (200 mM). The leaf proline content registered almost similar adverse effects at iso-osmotic potentials of high intensity of salinity and drought stress. Here, ricebean seedling recorded 16.50% reduction over that of control for both in leaf proline content for both 200 mM NaCl and 18% PEG solution at osmotic potential of -0.8 MPa. The salinity and drought at high intensity was found to produce similar adverse effects on content of leaf proline in ricebean in the present experiment. So it cannot be concluded that increase proline content is indicative of resistance against osmotic stress but it indicates certainly that increment in proline content at moderate stress level promotes the defensive mechanism against drought and salinity stress.

The guaiacol peroxidase (GPOX) was also involved in the catabolism of H_2O_2 in the cytosol, vacuole and cell wall (Asada, 1992). Carrasco-Ríos and Pinto (2014) showed in maize seedlings that GPOX levels increased significantly under saline conditions at different osmotic potential. The leaf GPOX activity of ricebean seedling significantly increased under all the treatments of salinity and drought stress over that of unstressed control (Fig 2) although the level decreased with increased stress intensity in all the cases. The variety Bidhan 1 recorded 28.60% and 75.53% increase in content of leaf GPOX activity in 200 mM NaCl and 18% PEG solution producing an osmotic potential of -0.8 MPa, respectively, over that of control. The findings corroborated well the early reports of Livingstone *et al*. (1992) and Shi *et al*. (2006). Shi *et al*. (2006) showed in maize a linear and significant increase in GPOX activity under water stress, but PEG at highest osmotic potential the GPOX activity decreased which is similar to the present experiment. Finally, the leaf GPOX content registered more induction under drought stress in comparison with salinity stress at iso-osmotic potentials.

Ascorbate peroxidase (APX) is one of the fine regulators of intracellular ROS level and the enzyme detoxify H_2O_2 by using ascorbate for reduction in the ascorbate glutathione cycle (Noctor and Foyer, 1998). The APX has higher affinity for hydrogen peroxide and this enzyme is present in almost

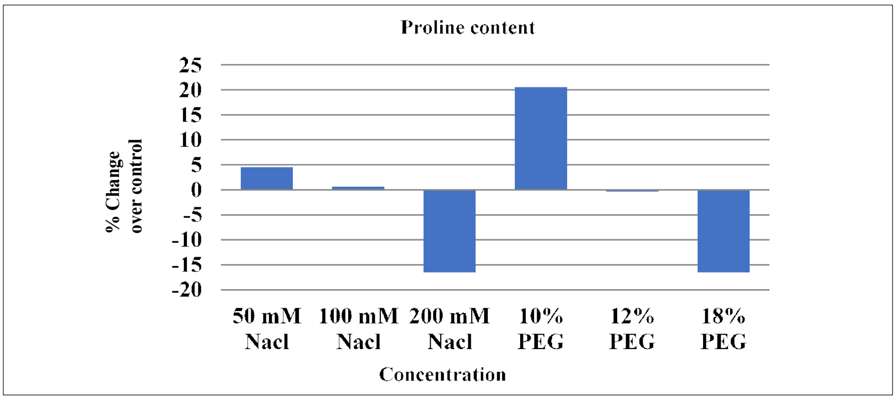

Fig 1: Changes in proline content in leaves of ricebean cv Bidhan 1 under abiotic stress.

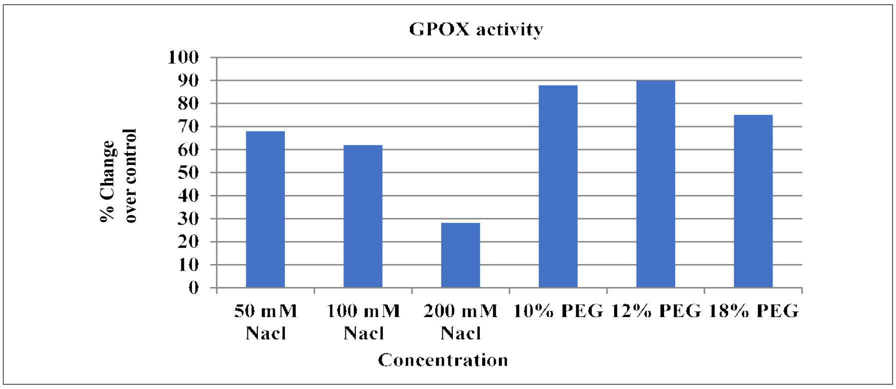

Fig 2: Changes in GPOX activity in leaves of ricebean cv Bidhan 1 under abiotic stress.

every ROS generating subcellular compartment. The activity of APX in leaf significantly increased over control at mild and moderate intensity of drought and salinity while it ultimately decreased under severe stress (Fig 3). Previously, the adverse effects of salinity stress (Dar *et al.* 2007) and drought stress (Bhardwaj and Yadav, 2012; Jiang-Jing Long *et al.* 2013) on APX in legumes were reported by different researchers. Jiang-Jing Long *et al.* (2013) showed in grass pea and garden pea seedlings stressed with 20% polyethylene glycol 6000 (PEG) that PEG caused a significantly greater increase of ascorbate peroxidase (APX). Here, the variety of ricebean recorded 65.30% and 57.14% reduction in APX activity for both 200 mM NaCl and 18% PEG solution producing an osmotic potential of -0.8 MPa, respectively, over that of control.

Catalase (CAT) is a main enzyme to eliminate H_2O_2 in the mitochondrion and microbody (Shigeoka *et al.* 2002) and thus help in ameliorating the detrimental effects of oxidative stress (Armand *et al.* 2016). It converts hydrogen peroxide to water and molecular oxygen (Willekens *et al.,* 1995). Catalase (CAT) activity increased gradually over control along with increase in NaCl concentration upto 100 mM followed by slight decline at the highest concentration, while in case of PEG, it only registered higher value over control at 10% PEG (Table 2), but then decreased substantially with increase in PEG concentration (Fig 4). Thus, it might be concluded that higher intensity of drought stress produced negative effects on CAT activity in this variety of ricebean. De Azevedo Neto *et al.* (2006) also found higher CAT activity in two maize cultivars differing in salt tolerance. There are other reports are available on the effect of salt stress on CAT activity in several plant species. CAT activity has been found to increase under salt stress in blackgram (Sivakumar and Jaya Priya, 2021), soybean (Comba *et al.* 1998), tobacco (Bueno et *al.,* 1998). Pratap and Sharma (2010) showed in the seedling of blackgram under drought stress using PEG-6000 at various osmotic potentials (-2, -5 and -10 bars) that the catalase (CAT) activity was increased at different osmotic gradients in comparison to control. This finding also conformed to early reports of Jiang-Jing Long *et al.* (2013). Bhardwaj and Yadav (2012) on an experiment of horsegram showed that antioxidant enzymes, likely Catalase (CAT) showed significant increase in the tolerant variety than the sensitive one under drought stress. PEG caused a significantly greater increase of

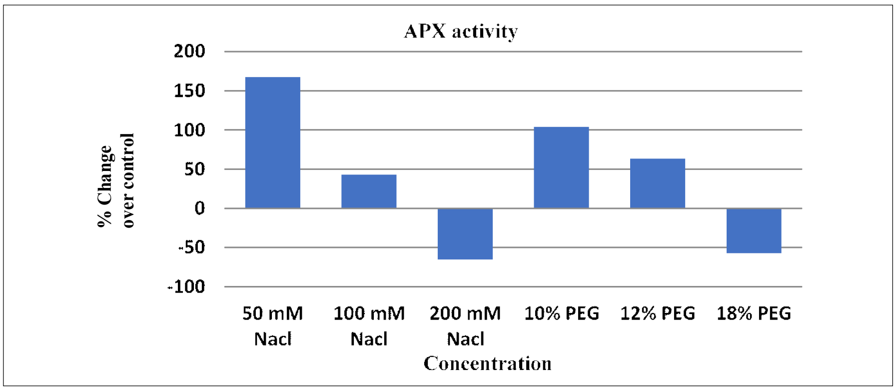

Fig 3: Changes in APX activity in leaves of ricebean cv Bidhan 1 under abiotic stress.

Table 2: Effect of salinity and drought stress on contents of proline and activities of guaiacol peroxidise (GPOX) ascorbate peroxidise (APX) and catalase (CAT) enzymes in the leaves of ricebean cv. Bidhan 1.

Treatments	Proline[a]	GPOX[b]	APX[c]	CAT[d]
Control	789.13	169.20	0.49	1.55
NaCl 50 mM	824.90 (4.53)	285.00 (67.94)	1.31 (167.34)	1.56 (0.64)
NaCl 100 mM	794.24 (0.65)	274.80 (61.93)	0.70 (42.85)	1.80 (16.12)
NaCl 200 mM	658.85 (-16.51)	217.60 (28.23)	0.17 (-65.30)	1.74 (12.25)
PEG 10%	951.35 (20.56)	318.80 (87.86)	1.00 (104.08)	1.90 (22.58)
PEG 12%	786.58 (-0.32)	322.20 (89.86)	0.80 (63.26)	1.50 (-3.22)
PEG 18%	658.85 (-16.51)	297.00 (75.01)	0.21 (-57.14)	1.30 (-16.12)
S.E. m(±)	6.55	4.19	0.04	0.04
C.D. (P=0.05)	19.88	12.71	0.13	0.13

[a]Data expressed as µM g^{-1} fresh weight.

[b]Data expressed as ΔA470 min^{-1} g^{-1} fresh weight.

[c]Data expressed as unit min^{-1} g^{-1} fresh weight.

[d]Data expressed as unit min^{-1} g^{-1} fresh weight.

Data in parentheses indicate percentage increase (+) or decrease (-) over control.

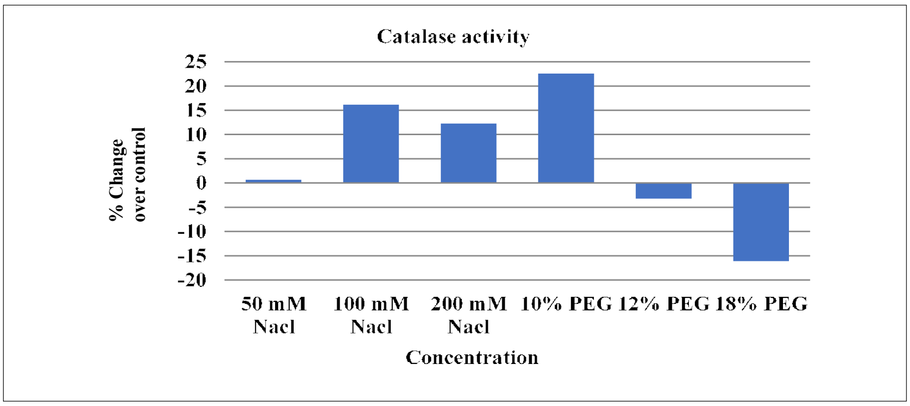

Fig 4: Changes in catalase activity in leaves of ricebean cv Bidhan 1 under abiotic stress.

catalase (CAT) showed by Jiang-Jing Long *et al.* (2013) in grass pea and garden pea seedlings stressed with 20% polyethylene glycol 6000 (PEG). In term of leaf catalase activity drought stress was found to produce more adverse effects than salinity stress.

CONCLUSION

From the present study, it might be concluded that drought and salinity are both important environmental challenges that reduces crop productivity by hampering balance between ROS generation and its safe detoxification by antioxidants. Though both the stresses ultimately induce oxidative stress to plants but drought stress was found to register more drastic effects on seedling growth as compared to iso-osmotic potential of salinity stress (200 mM NaCl and 18%PEG) in ricebean cultivar Bidhan 1. The drought stress also exhibited more negative effects on GPOX and CAT activity. While the APX activity was found to be more adversely affected by salinity stress. The leaf proline content at the highest intensity of both the salinity and drought stress have found similar effects. Differential sensitivity and affinity of catalase (CAT) and ascorbate peroxidase (APX) to ROS under drought and salinity stress is suggestive for their differential role in ROS scavenging.

ACKNOWLEDGEMENT

The authors acknowledge the assistance extended by AICRP on Forage Crops, Kalyani Centre, for supplying plant materials and Department of Plant Physiology, Faculty of Agriculture, BCKV for providing with all the facilities and support.

REFERENCES

Aniat-ul-Haq, Vamil, R., Agnihotri, R.K., Shakir, H.D. and Sharma, R. (2012). Growth performance of *lens culinaris* MEDIK under NaCl induced salt stress. International Journal of Current Research. 4(12): 12-15.

Armand, N., Amiri, H. and Ismaili, A. (2016). The effects of spraying methanol solution and drought stress on level of antioxidant enzyme activity, growth, biomass and yield of beans (*Phaseolus vulgaris* L.cv. COS16). Legume Research. 39(3): 385-395

Asada, K. (1992). Ascorbate peroxidase a hydrogen peroxide scavenging enzyme in plants. Physiologia Plantarum. 85: 235-241.

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

Bhardwaj, J. and Yadav, S.K. (2012). Comparative study on biochemical parameters and antioxidant enzymes in a drought tolerant and a sensitive variety of horsegram (*Macrotyloma uniflorum*) under drought stress. American Journal of Plant Physiology. 7: 17-29.

Bueno, P., Piqueras, A., Kurepa, J., Savoure, A., Verbruggen, N., Montagu, V.M. and Inze, D. (1998). Expression of antioxidant enzymes in response to abscisic acid and high osmoticum in tobacco BY-2 cell cultures. Plant Sci. 138: 27-34.

Cakmak, I., Strbac, D. and Marschner, H. (1993). Activities of hydro-gen peroxide scavenging enzymes in germinated wheat seeds. J. Exp. Bot. 44: 127-132.

Carrasco-Ríos, L. and Pinto, M. (2014). Effect of salt stress on antioxidant enzymes and lipid peroxidation in leaves in two contrasting corn, 'Lluteño' and 'Jubilee'. Chilean Journal of Agricultural Research. 74.

Chen, C. and Dickman, M.B. (2005). Proline Suppresses Apoptosis in the Fungal Pathogen *Colletotrichum trifolii*. Proc. Natl. Acad. Sci. U.S.A. 102(9): 3459-3464.

Comba, M.E., Benavides, M.P., Gallego, S.M. and Tomaro, M.L. (1998). Relationship between nitrogen fixation and oxidative stress induction in nodules of salt treated soybean plants. Phyton. 60: 115-126.

Dar, Z.M., Hemantaranjan, A. and Panday, S.K. (2007). Antioxidative response of mungbean (*Vigna radiata* L.) to salt stress. Legume Research. 30: 57-60.

De Azevedo Neto, A. D., Prisco, J. T., Enéas-Filho, J., Abreu, C. E. B. de and Gomes-Filho, E. (2006). Effect of salt stress on antioxidative enzymes and lipid peroxidation in leaves and roots of salt-tolerant and salt-sensitive maize genotypes. Environmental and Experimental Botany. 56(1):87-94.https://doi.org/10.1016 j.envexpbot. 2005.01.008.

El Sayed, H.E.A. (2011). Influence of salinity (NaCl and Na_2SO_4) treatments on growth development of broad bean (*Vicia faba* L.) plant. American Eurasian Journal of Agricultural and Environmental Sciences. 10: 600-610.

Epstein, E. (1972). Mineral Nutrition of Plants: Principles and Perspectives. John Wiley and Sons, New York.

Farooq, M., Wahid, A., Kobayashi, N., Fujita, D. and Basra, S.M.A. (2009). Plant drought stress: effects, mechanisms and management. Agron. Sustain. Dev. 29: 185-212.

Gill, S.S., Khan, N.A., Anjum, N.A. and Tuteja, N. (2011). Amelioration of cadmium stress in crop plants by nutrients management: Morphological, physiological and biochemical aspects. Plant Stress. 5: 1-23.

Jaleel, C.A., Manivannan, P., Lakshmanan, G.M.A., Gomathinayagam, M. *et al.* (2008). Alterations in morphological parameters and photosynthetic pigment responses of *Catharanthus roseus* under soil water deficits. Colloids Surf. B: Biointerfaces. 61: 298-303.

Jiang-Jing Long, Su-Miao, Chen-Yue Ru, Gao-Nan, Jiao-Cheng Jin, Sun-Zheng Xi, Li-Feng Min, Wang-Chong Ying (2013). Correlation of drought resistance in grass pea (*Lathyrus sativus*) with reactive oxygen species scavenging and osmotic adjustment. Biologia Bratislava. 68: 231-240.

Kundur, R., Reddy, P.T. and Rao, M.D. (2016). Effect of PEG mediated water stress on solute accumulation, relative water content, biomass and antioxidant enzymes in rice. Indian Journal of Agricultural Research. 50: 398-405.

Livingstone, D.R., Lips, F., Martinez, P.G. *et al.* (1992). Antioxidant enzymes in the digestive gland of the common mussel *Mytilus edulis*. Marine Biology. 112: 265-276. https://doi.org/10.1007/BF00702471.

Marschner, H. (1995). Mineral Nutrition of Higher Plants, 2nd ed.; Academic Press: London, UK.

Mittler, R., Vanderauwera, S., Gollery, M. and Van Breusegem, F. (2004). Reactive oxygen gene network of plants. Trends Plant Sci. 9: 490-498.

Mohammadkhani, N. and Heidari, R. (2008). Effects of Drought Stress on Soluble Proteins in two Maize Varities. Turkish Journal of Biology. 32: 23-30.

Mohanty, S.K. and Sridhar, R. (1982). Physiology of rice tungro virus disease: Proline accumulations due to infection. Physiol. Plant. 56: 89-93.

Munns, R. (2005). Genes and salt tolerance: Bringing them together. Plant Physiol. 167: 645-663.

Nakano, Y. and Asada, K. (1981). Hydrogen peroxide is scavenged by ascorbate specific peroxidase in spinach chloroplasts. Plant Cell Physiol. 22: 867-880.

Noctor, G. and Foyer, C. (1998). Ascorbate and glutathione: Keeping active oxygen under control. Annual Review of Plant Physiology and Plant Molecular Biology. 49: 249-279.

Pratap, V. and Sharma, Y.K. (2010). Impact of osmotic stress on seed germination and seedling growth in black gram (*Phaseolus mungo*). Journal of Environmental Biology. 31: 721-72.

Siegel, B.Z. and Galston, A.W. (1967). The iso-peroxidases of *Pisum sativum*. Physiol. Plant. 42: 212- 226.

Singh, R., Hemantaranjan, A. and Patel, P.K. (2015). Salicylic acid improves salinity tolerance in field pea (*Pisum sativum* L.) by intensifying antioxidant defense system and preventing salt-induced nitrate reductase (NR) activity loss. Legume Research. 38(2): 202-208.

Sivakumar, R. and Jaya Priya, S. (2021). PGRs and nutrient consortium effect on water relations, photosynthesis, catalase enzyme and yield of blackgram under salinity stress. Legume Research. 44(4): 413-418. DOI: 10.18805/LR-4118.

Shi, L.R., Lu, Z.G., Bai, L.R. and Cu, X.G. (2006). Studies on injury of maize seedlings under iso-osmotic NaCl and PEG (polyethylene glycol) stress. Journal of Maize Sciences. 14(4): 100-103.

Shigeoka, S., Ishikawa, T., Tamoi, M., Miyagawa, Y., Takeda, T., Yabuta, Y. and Yoshimura, K. (2002). Regulation and function of ascorbate peroxidase isoenzymes. Journal of Experimental Botany. 53(372): 1305-1319. https://doi.org/10.1093/jexbot/53.372.1305.

Sosa, L., Llanes, A., Reinoso, H., Reginato, M. and Luna, V. (2005). Osmotic and Specific Ion Effects on the Germination of *Prosopis strombulifera*. Annals of Botany. 96: 261-267.

Strizhov, N., Abrahám, E., Okrész, L., Blickling, S., Zilberstein, A., Schell, J. *et al.* (1997). Differential expression of two P5CS genes controlling proline accumulation during salt-stress requires ABA and is regulated by ABA1, ABI1 and AXR2 in Arabidopsis. The Plant Journal. 12(3): 571-581.

Tang, X., Mu, X., Shao, H., Wang, H. and Brestic, M. (2015). Global plant-responding mechanisms to salt stress: Physiological and molecular levels and implications in biotechnology. Critical Reviews in Biotechnology. 35: 425-437. https://doi.org/10.3109/07388551.2014.889080.

Verma, S.K., Chaudhary, M. and Prakash, V. (2012). Study of the alleviation of salinity effect due to enzymatic and non-enzymatic antioxidants in *Glycine max*. Research Journal of Pharmaceutical, Biological and Chemical Sciences. 3: 1177-1185.

Willekens, H., Inze, D., Van Montagu, M., *et al.* (1995). Catalases in plants. Mol. Breeding. 1: 207-228. https://doi.org/10.1007/BF02277422.