Australian Journal of Crop Science

AJCS 12(03):458-466 (2018) doi: 10.21475/ajcs.18.12.03.pne937 AJCS ISSN:1835-2707

Application of solid matrix priming to ameliorate salinity stress in mung bean (Vigna radiata)

Sujoy Kumar Sen¹ and Palash Mandal²*

¹Department of Botany, Siliguri College, Darjeeling, West Bengal, India-734 001

Plant Physiology and Pharmacognosy Research Laboratory, Department of Botany, University of North Bengal, Darjeeling, West Bengal, India-734 013

*Corresponding author: nbubotanypm@gmail.com

Abstract

Present study aimed to study the responses of mung bean seedlings under 0, 4 and 8dSm⁻¹ concentrations after Solid matrix priming (SMP), using celite matrix 10% moisture with 15 different elicitors such as kinetin, napthalene acetic acid (NAA), abscisic acid (ABA), salicylic acid, chitosan, calcium chloride (CaCl₂), gibberellic acid (GA₃), putrescine, gallic acid, Catechol, proline, lanthanum chloride (LaCl₃), ethylene glycol bis (2-aminoethyl ether) tetra acetic acid (EGTA), sodium nitroprusside (SNP) and water (hydropriming). The root length (RL), shoot length (SL), RL/SL ratio, average lateral root number, plant fresh weight (FW) and dry weight (DW) were measured after 6 days of germination. Indices like plant height stress tolerance index (PHSI), root length stress tolerance index (RLSI), shoot height stress tolerance index (SHSI), stress tolerance index (STI), root phytotoxicity, shoot phytotoxicity, fresh weight percentage reduction (FWPR), dry weight percentage reduction (DWPR), relative seedling water content (RSWC) were measured. The root length (RL), shoot length (SL), and biomass production along with dry weight percentage reduction (DWPR) were significantly decreased with increasing salt stress. However, priming treatments improved different morphological traits like SL, RL in almost all cases compared to unprimed seeds. Effect of SMP with salicylic acid and chitosan regarding RL and SL under high salinity is noteworthy. In case of FW, only chitosan at 4 dSm⁻¹ and catechol at 8dSm⁻¹ showed significant improvement i.e. 7.46% and 5.74%, respectively, compared to unprimed seeds. The DW at 4 dSm⁻¹ and significant enhanced biomass even better than unprimed seedling in control. Overall, among the elicitors studied chitosan followed by salicylic acid showed the best results under salinity stress.

Keywords: mung bean, salinity, seedling characters.

Introduction

Vigna radiata (L.) Wilkzek (mung bean) is a leguminous crop with a high nutritive value. It is consumed widely in the form of dry seeds or sprouts because of high protein content, vitamins and amino acids (Khattak et al., 2001). The seeds contain 22-28% protein, 60-65% carbohydrates, 1.0-1.5% fat, 3.5-4.5% fibres and 4.5-5.5% ash. Because of the production of huge biomass and capability to recover after grazing to yield plentiful seeds, this crop can be utilized for both seeds and forage (Iqbal and Ashraf, 2010). On the other hand, salinity is considered as a vital problem in agriculture as it causes significant reduction in plant growth of economic crops through both ionic toxicity and osmotic stress (Upadhyay and Panda, 2005).

Salinity is a severe abiotic stress which leads to decrease in crop production. Usually, with higher salt concentration in irrigation water the salinity problems are amplified. Nearly 6% of the world's total land area is salt affected (FAO, 2009). Salinity severely affects crop production (Maibody and Feizi, 2005; Demicral and Turkan, 2005). Under salinity stress the mungbean production may be reduced up to 50% (Singh et al., 1989). Such a reduction in mung bean growth and production under salinity stress may occur due to a combination of ions toxicity and altered water relations

which also results in large accumulation of Na^{+} and Mg^{+} ions. This also results in the reduction of the concentration of calcium and potassium in plant body. Moreover, with increasing salt stress, osmotic potential, water potential, stomatal conductance, transpiration etc are decreased (Raptan et al., 2001). Besides, during the life cycle of crop plants salinity tolerance is crucial. It is already confirmed that higher yields and early vigor in mung bean are highly correlated (Kumar et al., 2002) and that those plants which show tolerance at seedling stage also exhibit greater salt tolerance at adult stage (Shannon et al., 1998; Rao and McNeilly, 1999; Soloviev et al., 2003; Khan et al., 2003). So there is urgent need to develop strategies to improve crop yield under salt stress. Salinity prevents water uptake by plants due to influence of ion toxicity and high osmotic stress (Hasanuzzaman et al., 2013). Salt stress sensitivity was assessed among five most commonly used cultivars in West Bengal, India and results arranged as follows: Samrat < Sonali < Panna < Sukumar < Bireshwar, in which Samrat was the most salt tolerant, whereas Bireshwar proved to be the most sensitive to salinity (Sen et. al., 2016). Hence, Samrat was chosen for Solid Matrix Priming (SMP) treatments with different elicitors for biochemical analyses after priming.

Heydecker (1973) defined seed priming as a technique, in which seeds absorb water slowly and post dehydration occurrence. Seed priming technique has been proved to be a practicable technology to improve speedy and consistent emergence, high vigor, and enhanced yields for plants (Janmohammadi et al., 2009; Rouhi et al., 2011). It can be done by various methods like solid matrix priming (SMP), hydro priming, osmopriming and using plant growth regulators (Harris et al., 1999). After priming, seeds show enhanced emergence rate, increased resistance to different types of stresses like drought (Wang et al., 2004), cold (Li and Fu, 1990) and salinity (Ruan et al., 2003). Germination and stress tolerance were studied in legumes by Gu et al. (2000). Priming helps to enhance resistance to abiotic stresses by operating different pathways involved in various metabolic processes (Jisha et al., 2013). Seed priming with plant growth hormones like auxin (IAA), gibberellins (GA), abscisic acid and ethylene in optimum doses has considerably improved the germination performance and yield of many crops under both normal and stress conditions (Hurly et al., 1991, Anosheh et al., 2014).

Though extensive works have been done on liquid priming in different crop species, the Solid Matrix Priming (SMP) is comparatively a new and efficient approach of seed invigoration. It is much advantageous than liquid priming because it needs a small amount of liquid per unit of seed and solid particles, compared to liquid priming. In case of SMP, slow or controlled imbibition allows repair mechanisms to work. Liquid priming may be injurious to the cotyledons due to fast liquid uptake, causing cell death (Orphanos and Heydecker, 1968). It is easy to handle and very convenient to any size of seeds. Moreover, aeration is hindered due to large volume of osmotic solutions and enriched air is frequently needed in liquid priming (Pill, 1995). SMP enhances adventitious roots, compared to the control (Beckman et al., 1993). Besides, very meagre work has been done on the SMP. The SMP on mung bean is a totally new approach. SMP utilizes carriers having properties like low osmotic potentials, high water holding capacities, and low bulk density (Khan et al., 1990). This study aimed to evaluate the potential of seed priming in improving early seedling growth in mung bean by some selected elicitors at 10% matrix moisture (using Celite as matrix) in alleviating the detrimental effects of salt stress with an emphasis on several growth parameters and indices.

Results and Discussion

Effects of SMP with different elicitors

Results revealed that priming treatments improved different morphological traits like SL, RL etc. In comparison to the unprimed seeds, overall maximum improvement in RL was observed using chitosan treatment at 8 dSm⁻¹ salinity. Furthermore, it was noticed that induction of root growth was more prominent at higher salinity in the primed seeds compared to unprimed seeds, especially in presence of gallic acid, LaCl₃, putrescine and SNP. Overall, the maximum improvement in SL was observed with application of LaCl₃ and chitosan followed by catechol, putrescine, CaCl₂, EGTA and kinetin at 4 dSm⁻¹ salinity. Conversely, at 8 dSm⁻¹ salinity, catechol, putrescine and gallic acid showed better results. In case of FW, only chitosan at 4 dSm⁻¹ and catechol at 8 dSm⁻¹ showed significant improvement i.e. 7.46% and 5.74%, respectively, compared to unprimed seeds. Regarding DW, GA₃ followed by SNP showed significantly enhanced biomass at 4 dSm⁻¹, even 23% and 13% better than control, respectively. GA₃ and SNP showed 96.55% and 78.33% improvement at 4 dSm⁻¹ salinity, compared to unprimed seeds. The result revealed progressive decrease in seedling dry weight with higher salt concentrations, which corroborates with the previous work done by Jeannette et al. (2002). The DWPR showed similar trends as above. It is noted that, although SNP showed better results at low salinity level but it was not so effective at higher salinity levels. On the other hand, GA₃ was found to be effective at both 4 and 8 dSm⁻¹ salinity levels. In case of fresh weight increase of the plants, although GA₃ and SNP showed comparatively better results than other priming agents, no other attributes was found to be better than control (i.e. without salinity). It also should be noted that, except CaCl₂, plant biomass was increased in all cases compared to unprimed seeds at 8 dSm⁻¹ salinity. Among these, GA₃ and proline showed the maximum improvement followed by NAA, ABA, kinetin and gallic acid. Better results were obtained for chitosan and GA3 primed seeds for Fresh Weight Percentage Reduction (FWPR) at 4 dSm⁻¹ salinity, only compared to unprimed seeds. Regarding RLSI, more pronounced and effective results were obtained at higher salinities with gallic acid, catechol, chitosan, LaCl₃, CaCl₂, sodium nitroprusside, proline, and 1-naphthaleneacetic acid (NAA). However, the GA₃ showed better results only at low salinity levels. The shoot height stress tolerance index (SHSI) revealed similar results compared to RLSI. At lower salinity, only gallic acid showed better result than unprimed seeds. But at 8 dSm⁻¹ salinity, catechol and chitosan along with gallic acid showed improved results than unprimed seeds. In all cases, kinetin and EGTA failed to invigorate (for PHSI, RLSI, and SHSI). Present work is in agreement with the effect of gallic acid and catechol at its optimum dose of 50 µm as indicated by previous workers (Muzaffar et al., 2012). Higher values of water uptake percent were obtained using CaCl₂, catechol and chitosan treatment at 4 dSm⁻¹ salinity, whereas only CaCl₂ showed higher values at 8 dSm⁻¹.

Four types of phytohormones were used in this experiment to ameliorate the detrimental effects of salt stress during early stage of seedling development. Among them, best results were obtained in case of GA₃. This is also in agreement with the previous results in wheat (Parasher and Varma, 1988) and rice (Prakash and Prathapasenan, 1990). Similarly, enhancement of plant water use efficiency along with reduced stomatal resistance has been demonstrated in tomato under low saline condition (Maggio et al., 2010). Therefore, it is obvious that GA₃ could induce salt tolerance in plants under salinity. Furthermore, GA₃ also interacts with other hormones regulating different metabolic processes. However, salt tolerance induction by GA₃ priming is not very clear in crops. But according to Iqbal and Ashraf (2010), the possible mechanism of inducing salt tolerance by GA₃ priming is hormonal homeostatsis as hormonal balance is grossly disturbed by salinity in plants. It is already established that under salinity, auxin concentration is reduced considerably. Dunlap and Binzel (1996) reported 75% reduction of indole-3-acetic acid in tomato. Similar type of reduction in auxin was also noticed in wheat root system by Sakhabutdinova et al. (2003). Thus,



Fig 1. Principal component analysis between different treatments and morphological attributes of mung bean at 0 dSm⁻¹ salinity. [Abbreviations used: RtPh_root phytotoxicity, StPh_shoot phytotoxicity, DWPR_dry weight percentage reduction, FWPR_fresh weight percentage reduction, RL/SL_ Root length/shoot length ratio, ALRN_Average lateral root number, Dw_Dry weight, STI_salt tolerance index, PHSI_ plant height stress tolerance index, WATUP_water uptake %, rswc_ relative water content, RL_root length, RLSI_ root length stress tolerance index, FW_Fresh weight, SHSI_shoot height stress tolerance index, SL_shoot length; in all attributes "zero" indicates 0 dSm⁻¹ salinity].



Fig 2. Principal component analysis between different treatments and morphological attributes of mung bean at 4 dSm⁻¹ salinity. [Abbreviations used: RtPh_root phytotoxicity, StPh_shoot phytotoxicity, DWPR_dry weight percentage reduction, FWPR_fresh weight percentage reduction, RL/SL_ Root length/shoot length ratio, ALRN_Average lateral root number, Dw_Dry weight, STI_salt tolerance index, PHSI_ plant height stress tolerance index, WATUP_water uptake %, rswc_ relative water content, RL_root length, RLSI_ root length stress tolerance index, FW_Fresh weight, SHSI_ shoot height stress tolerance index, SL_shoot length; in all attributes "4" indicates 4 dSm⁻¹ salinity].



Fig 3. Principal component analysis between different treatments and morphological attributes of mung bean at 8 dSm⁻¹salinity. [Abbreviations used: RtPh_root phytotoxicity, StPh_shoot phytotoxicity, DWPR_dry weight percentage reduction, FWPR_fresh weight percentage reduction, RL/SL_ Root length/shoot length ratio, ALRN_Average lateral root number, Dw_Dry weight, STI_salt tolerance index, PHSI_ plant height stress tolerance index, WATUP_water uptake %, rswc_ relative water content, RL_root length, RLSI_ root length stress tolerance index, FW_Fresh weight, SHSI_ shoot height stress tolerance index, SL_shoot length; in all attributes "8" indicates 8 dSm⁻¹ salinity].



Fig 4. Heat map dendrogram of the morphological parameters and stress indices with different elicitor treatments at 0 dSm⁻¹ salinity. Treatments were separated into two main clusters based on nature of priming agent, whereas the attributes were also separated in two main clusters confirming their relative association. [Abbreviations used: RtPh_root phytotoxicity, StPh_shoot phytotoxicity, DWPR_dry weight percentage reduction, FWPR_fresh weight percentage reduction, RL/SL_ Root length/shoot length ratio, ALRN_Average lateral root number, Dw_Dry weight, STI_salt tolerance index, PHSI_ plant height stress tolerance index, WATUP_water uptake %, rswc_ relative water content, RL_root length, RLSI_ root length stress tolerance index, FW_Fresh weight, SHSI_ shoot height stress tolerance index, SL shoot length; in all attributes "zero" indicates 0 dSm⁻¹ salinity]



Fig 5. Heat map dendrogram of the morphological parameters and stress indices with different elicitor treatments at 4 dSm⁻¹ salinity. Treatments were separated into four main clusters based on nature of priming agent, whereas the attributes were separated in two main clusters confirming their relative association [Abbreviations used: RtPh_root phytotoxicity, StPh_shoot phytotoxicity, DWPR_dry weight percentage reduction, FWPR_fresh weight percentage reduction, RL/SL_ Root length/shoot length ratio, ALRN_Average lateral root number, Dw_Dry weight, STI_salt tolerance index, PHSI_ plant height stress tolerance index, WATUP_water uptake %, rswc_ relative water content, RL_root length, RLSI_ root length stress tolerance index, FW_Fresh weight, SHSI_ shoot height stress tolerance index, SL_shoot length; in all attributes "4" indicates 4 dSm⁻¹ salinity]



Fig 6. Heat map dendrogram of the morphological parameters and stress indices with different elicitor treatments at 8 dSm⁻¹ salinity. Treatments were separated into three main clusters based on nature of priming agent, whereas the attributes were separated in four main clusters confirming their relative association. [Abbreviations used: RtPh_root phytotoxicity, StPh_shoot phytotoxicity, DWPR_dry weight percentage reduction, FWPR_fresh weight percentage reduction, RL/SL_ Root length/shoot length ratio, ALRN_Average lateral root number, Dw_Dry weight, STI_salt tolerance index, PHSI_ plant height stress tolerance index, WATUP_water uptake %, rswc_ relative water content, RL_root length, RLSI_ root length stress tolerance index, FW_Fresh weight, SHSI_ shoot height stress tolerance index, SL_shoot length; in all attributes "8" indicates 8 dSm⁻¹ salinity]

it appears that under stress conditions the decrease in plant growth could occur due to altered hormonal balance and the stress conditions can be overcome by their exogenous application. There are many reports of alleviating the adverse effect of salinity in wheat by the application of synthetic or natural auxin like NAA (Gulnaz et al., 1999). Similar results of increased vigour with enhanced FW and DW in wheat seedlings were reported by the application of auxin by Akbari et al. (2007). However, our study contradicts the earlier findings as significant improvement of seedling vigour in *Vigna radiata* was not observed by exogenous application of NAA through SMP. ABA (Keskin et al., 2010) and kinetin (Boucaud and Ungar, 1976) are also proposed to be a major mediator in plant responses under salt stress condition enabling plants to live in adverse situations. Under salinity, such hormonal content is decreased (Walker and Dumbroff, 1981) and; hence, their application might overcome the inhibiting effects of salt stress as shown in wheat (Naqvi et al., 1982) and potato plants (Abdullah and Ahmad, 1990). In our experiment, GA₃ showed the best results among all hormonal treatments in all attributes. Previous works revealed that SMP by GA₃ can break seed dormancy and enhance seed germination in eastern gama grass (*Tripsacum dactyloides* L.) (Rogis et al., 2004). Khan et al. (2004) also reported similar trends showing alleviative effect of GA₃ over salt stress on seed germination. Phenyl propanoid elicitors could also play a vital role in alleviating the harmful effects of salinity in plants. For instance, chitosan treated rice seeds showed better seed germination and seedling growth withstanding the stress condition (Ruan and Xue, 2002). Improved rate of germination and growth were exhibited by chitosan treatment in ajowan (Mahdavi and Rahimi, 2013) and sunflower (Cho et al., 2008), overcoming the negative result of salinity. Improvement in germination attributes and stress tolerance indices were also noticed in mung bean in Sonali B1 cultivar of mung bean by SMP with chitosan under 3 different (5%, 10% and 20%) moisture levels (Sen and Mandal, 2016). In this study, chitosan priming significantly improved the detrimental effect of salinity (up to 8 dSm⁻¹), particularly in case of SL, RL, RL/SL ratio, SHSI, RLSI, PHSI and STI. Positive effects of simple phenylpropanoids like catechol and gallic acid were described by Muzaffar et al. (2012). In our study, significant improvement was noticed for PHSI, RLSI, SHSI and STI by catechol and gallic acid treatment. Priming with polyamines also was found useful to alleviate salinity in some plants (Mansour et al., 2002). In the present study, we observed that putrescine could induce shoot growth at high salinity to an extent. Under salinity stress, the mineral nutrient availability to the plants is hampered (Knight and Knight, 2001) but gaining Ca^{2+} beneficial to the plants, as it regulates many physiological and cellular events providing stress protection (Tang et al., 2006). The negative effects of Na⁺ and Mg²⁺ can be alleviated by Ca²⁺ in *Hordeum vulgare* (Bliss et al., 1986). Our results regarding the rate of water uptake showed best action in case of CaCl₂ treatment at both high and low salinity levels.

Principal Component Analysis (PCA)

PCA statistically explains the covariance organization of a set of variables and is used mainly to dimensionally condense the multiple features to two or three dimensions. In general, it indicates the main directions, in which the data varies and summarizes the variation of correlated multi-attributes with respect to uncorrelated components set. PCA in present study (Fig 1, 2, 3) revealed that with increasing salt stress, all the treatments were scattered; although in 0 dSm⁻¹ salinity all the treatments were clustered towards the centre of the graph and all the attributes form 3 distinct clusters showed their correlations. In higher salinity levels, chitosan enhanced shoot phytotoxicity level, inducing tolerance to the plant. Similarly SA improved plants against root phytotoxicity. In 0 dSm⁻¹ salinity, GA₃, SA, Kinetin, Proline showed effective results compared to untrimmed seeds. In 4 dSm⁻¹ salinity, LaCl₃, Kinetin, GA₃, Proline, ABA and SA showed effective results compared to unprimmed seeds. In 8 dSm⁻¹ salinity, chitosan, SNP, catechol, NAA, CaCl₂, GA₃, SA, Kinetin, showed effective results compared to the untrimmed seeds. Shoot phytotoxicity, DWPR, FWPR, RSWC and Water uptake % formed the 1st cluster, which was significantly affected by putrescine, catechol and CaCl₂. The 2nd cluster comprised of RLSL, RLSI, RL and SHSI, which were significantly affected by LaCl₃, SNP and NAA. The 3rd cluster contained STI, DW, FW, SL, PHSI and average lateral root number, which were significantly affected by proline, kinetin, ABA and EGTA. So, these attributes were seemed to be correlated in this experiment.

Heat map analysis

The cluster heat map is a graphical illustration of data. In heat map the individual values are represented as colours. In this case, the rows (columns) of the tiling are ordered in such a way that the similar rows (columns) are nearer to each other. On the other hand, hierarchical cluster trees were on the vertical and horizontal margins of the tiling. In the present study (Fig 4, 5, 6) overall chitosan, SA and GA_3 showed significant improvement in almost all morphological parameters. At higher salinity levels, significant enhancement in PHSI, SL, DWPR, etc. using chitosan priming is noteworthy.

Materials and Methods

Plant material

Seeds of mung bean cultivar "Samrat" was collected from the Pulse and oilseed Research station, Berhampur, West Bengal, India.

Treatments

Our interest in selection of suitable elicitors in SMP system in mung bean seeds came from several research workers (Akbari et al., 2010, Muzaffar et al., 2012, Zavariyan et al., 2015, Gahtyari et al., 2017, Mahdavi and Rahimi 2013). The priming agents were selected under five categories viz., hormonal priming (Kinetin, GA₃, NAA and ABA), phenyl propanoids (gallic acid, catechol, and chitosan), Amine/amino acid (putrescine and proline), components related with calcium ion as a mediator of cell signaling (CaCl₂ as exogenous source of calcium ion, LaCl₃ as blocker and EGTA as chelator of calcium), and NO generating agent (SNP).

Experimental design and conduction

After surface sterilization, mung bean seeds (Samrat) were mixed with celite (1:1 proportion) with the solutions of some selected elicitors in selected dose viz., kinetin, gallic acid (50 μ m), catechol (50 μ m), chitosan (0.2% dissolved in 1% acetic acid solution, whose pH was adjusted to 6.00 with the help of 1% sodium hydroxide solution), LaCl₃ (1mM), EGTA(1mM), putrescine (10 µm), CaCl₂(2 mM), SNP (2 mM), proline (500 μ m), GA₃(10⁻⁴ M), NAA (7.5 μ m) and ABA (10⁻⁴ M). Seeds were kept in plastic air tight zip packet at 10% moisture level for priming for 24 hours at room temperature. After the treatment, the celite was sieved. Then, the seeds were placed in three different saline solutions including control *i.e.*, 0, 4 and 8 dS/m. The electrical conductivity of saline solution was measured with a conductivity meter (dS m⁻¹ = deci Siemen per meter). Finally seeds were germinated in seed germinator (REMI) adjusted to 25±2°C. Data were recorded after 6 days of germination. RL and SL were measured with the help of table scale.

Traits measured

Weight reduction percentage

Fresh weight of each seedling was measured in an analytical balance. Each seedling was then dried to constant weight (80°C) for 48 hours and dry weights were immediately taken. According to each salt treatment, the fresh and dry weights (referred to the control), were calculated in percent, by the following equations:

1. Fresh weight (FW) percentage reduction: FWPR % = 100 ×(1- Dry weight $_{Salt stress}$ / Dry weight $_{control}$)

(1- Fresh weight $_{Salt stress}$ / Fresh weight $_{Control}$)

Salt Tolerance Index (STI)

It is calculated by the following standard formula: STI % = 100 × (Total DW _{Salt stress} /Total DW _{control}) (Ashraf et al., 2006)

Stress tolerance and Phytotoxicity analysis

These are measured by using following standard formulas:

1. Plant height stress tolerance index (PHSI) = (Plant height of stressed plant / Plant height of control plants) \times 100 (Ashraf et al., 2006)

2. Root length stress tolerance index (RLSI) = (RL of stressed plant / RL of control plants) \times 100 (Ashraf et al., 2006)

3. Shoot height stress tolerance index (SHSI) = Shoot height of stressed plant / Shoot height of control plants) \times 100 (Ashraf et al., 2006) Where, W₁ = Initial weight of seed; W₂ = Weight of seed after absorbing water in a particular time.

4. Root phytotoxicity (%) = (RL of Control – RL of treatment) / RL of control ×100 (Asmare, 2013)

5. Shoot phytotoxicity (%) = (SL of Control – SL of treatment) / SL of control × 100 (Asmare, 2013)

Relative water content (RSWC)

The relative water content of the seedling with respect to the FW was calculated by the following formula as described by (Asmare, 2013) RSWC $\% = 100 \times (FW-DW)/FW$

Water uptake percentage

It is calculated by the following formula as described by Gairola et al., 2011:

Water uptake % = $100 \times (W_2-W_1) / W_1$ where, W_1 = Seed fresh weight and W_2 = Seed dry weight

Statistical analysis

Principal component analysis (PCA) was used to determine the importance of different elicitors applied and their contribution in the improvement of different attributes of mung bean at different concentration of salinity using XLSTAT 2017 software. Heat maps were made with the help of R software.

Conclusion

The results recorded in this study recommended that application of salicylic acid, chitosan, $CaCl_2$, putrescine and GA_3 led to significant improvement for different attributes in mung bean seedlings enhancing their phytotoxicity level and; thus, inducing tolerance against salt stress.

Acknowledgements

This research was supported by UGC minor research project.

References

- Akbari GA, Hojati M, Modarres-Sanavy SAM, Ghanati F (2011) Aminolevulinic acid improves photosynthetic gas exchange capacity and ion uptake under salinity stress in oilseed rape (*Brassica napus* L.) plants. Pestic Biochem Phys. 100: 244-250.
- Akbari N, Barani M, Drikvand E, Ahmadi H (2010) The effect of gibberellic acid (GA₃) on minerals of Mungbean (*Vigna radiata* L. Wilczek) irrigated with different levels of saline water. Afr J Agric Res. 5(4): 275-279.
- Akbari G, Sanavy SA, Yousefzadeh S, (2007) Effect of auxin and salt stress (NaCl) on seed germination of wheat cultivars (*Triticum aestivum*L.). Pakistan J Biol Sci. 10: 2557-2561.
- Anosheh HP, Emam Y, Ashraf M (2014) Impact of cycocel on seed germination and growth in some commercial crops under osmotic stress conditions. Arch Agron Soil Sci. 60(9): 1277-1289.
- Ashraf MY, Akhter K, Hussain F, Iqbal J (2006) Screening of different accessions of three potential grass species from Cholistan desert for salt tolerance. Pakistan J Bot. 38 (5): 1589-1597.
- Asmare HA (2013) Impact of salinity on tolerance, vigor and seedling relative water content of haricot bean (*Phaseolus vulgaris* L.) cultivars. J Plant Sc. 1 (3): 22-27.
- Beckman DW, Wilson CW, Stanley AL (1989) Age and growth of red drum, Sciaenopsocellatus, from offshore waters of the northern Gulf of Mexico. US Fish Bull. 87: 17-28.
- Bliss RD, Platt-Aloia KA, Thomson WW (1986) The inhibitory effect of NaCl on barley germination. J Plant Cell Environ. 9: 727-733.
- Boucaud J, Ungar IA (1976) Hormonal control of germination under saline conditions of three halophyte taxa in genus *Suaeda*. Physiol Plant. 36: 197-200.
- Cho MH, No HK, Prinyawiwatkul W (2008) Chitosan treatments affect growth and selected quality of sunflower sprouts. J Food Sci. 73(1): 570-577. http://dx.doi.org/10.1111/j.1750-3841.2007.00607
- Demicral T, Turkan I (2005) Comparative lipid peroxidation, antioxidant defense systems and proline content in roots of two rice cultivars differing in salt tolerance. Environ Exp Bot. 53: 247-257.
- Dunlap JR, Binzel ML (1996) NaCl reduces indole-3-acetic acid levels in the roots of tomato plants independent of stress induced abscisic acid. Plant Physiol. 112: 379-384.
- FAO (2009) Food Security and Agricultural Mitigation in Developing Countries: Options for Capturing Synergies. Rome, Italy.

www.fao.org/docrep/012/i1318e/i1318e00.pdf.

- Gahtyari NC, Jaiswal JP, Talha M, Choudhary R, Uniyal M, Kumar N (2017) Effect of osmotic stress and seed priming on wheat seed germination traits. Int J Curr Microbiol App Sci. 6(4): 2799-2809.
- Gairola KC, Nautiyal AR, Dwivedi AK (2011) Effect of temperatures and germination media on seed germination of *Jatropha curcas* L. Adv Biores. 2: 68-71.
- Gill PK, Sharma AD, Singh P, Bhullar SS (2003) Changes in germination, growth and soluble sugar contents of *Sorghum bicolor* (L.) Moench seeds under various abiotic stresses. Plant Grow Reg. 40: 157–62.
- Gu GP, Wu GR, Lu CM, Zhou CF, Zuo JJ, Wei JC (2000) Effect of PEG on vigor index and active oxygen metabolism in soybean seeds. Chin J Oil Crop Sci. 22(2): 26-30.
- Gulnaz AJ, Iqbal J, Azam F (1999) Seed treatment with growth regulators and crop productivity. II. Response of critical growth stages of wheat (*Triticum aestivum* L.) under salinity stress. Cereal Res. 27: 419-426.
- Harris D, Joshi A, Khan PA, Gothkar P, Sodhi PS (1999) Onfarm seed priming in semiarid agriculture: Development and evaluation in maize, rice and chickpea in India using participatory methods. Exp Agr. 35: 15–29.
- Hasanuzzaman M, Nahar K, Fujita M (2013) Plant response to salt stress and role of exogenous protectants to mitigate salt-induced damages. In: Ahmad P, Azooz MM, Prasad MNV, (eds) Ecophysiology and responses of plants under salt stress. Springer, New York, USA. 25-87.
- Heydecker W (1973) Germination of an idea: the priming of seeds. University of Notingham School of Agriculture Report. 50-67.
- Hurly RF, Staden JV, Smith MT (1991) Improved germination in seeds of guayule (*Parthenium argentatum* Gray) following polyethylene glycol and gibberellic acid pretreatments. Ann Appl Biol. 118: 175-184.
- Iqbal M, Ashraf M (2010) Gibberellic acid mediated induction of salt tolerance in wheat plants: Growth, ionic partitioning, photosynthesis, yield and hormonal homeostasis. Environ Exp Bot. doi:10.1016/j.envexpbot. 2010.06.002.
- Janmohammadi M, Moradi PD, Dezfuli P, Sharifzadeh F (2009) Seed invigouration techniques to improve germination and early growth of inbred lines of maize under salinity and drought stress. Gen. App Pl Physiol. 343(4): 215-226.
- Jeannette S, Craig R, Lynch JP (2002) Salinity tolerance of *Phaseolus* species during germination and early seedling Growth. Crop Sci. 42: 1584-1594.
- Jisha KC, Vijayakumari K, Puthur JT (2013) Seed priming for abiotic stress tolerance: an overview. Acta Physiol Plant. 35: 1381-1396.
- Keskin BC, Sarikaya AT, Yuksel B, Memon AR (2010). Abscisic acid regulated gene expression in bread wheat. Aust J Crop Sci. 4: 617-625.
- Khan MA, Gul B, Weber DJ (2004) Action of plant growth regulators and salinity on the seed germination of *Ceratoides lanata*. Can J Bot. 82: 37-42.
- Khan AS, Asad MA, Ali Z (2003) Assessment of genetic variability for NaCl tolerance in wheat. Pak J Agr Sci. 40: 33-36.
- Khan AA, Mura H, Prusinki J, Ilyas S (1990) Matriconditioning of seeds to improve emergence. Minneapolis, MN: Proceedings of the National Symposium on Stand Establishment of Horticultural Crops. 19-40.

- Khattak GSS, Haq MA, Rana SA, Elahi T, Srinives P (1998) An efficient technique for crossing mung bean (*Vigna radiata* (L.) Wilczek). Thai J Agric Sci. 31:577-582.
- Kumar R, Tyagi CS, Ram C (2002). Association of laboratory seed parameters with field performance in mung bean. Seeds Farms. 15: 33-36.
- Li XH, Fu JR (1990) Effects of PEG pretreatment in vigor and hardiness on seed vigor of peanut. Acta Sci Nat Univ Sunyaatseni. 29(1): 63–70. Lust JB. 1974. The herb book. Bantam Books Inc. New York.
- Maggio A, Barbieri G, Raimondi G, De Pascale S (2010) Contrasting Effects of GA3 Treatments on Tomato Plants Exposed to Increasing Salinity. J Plant Growth Regul. 29:63-72.
- Mahdavi B, Rahimi A (2013) Seed priming with chitosan improves the germination and growth performance of ajowan (*Carum copticum*) under salt stress. Eurasia J Biosci. 7:69-76.
- Maibody SAM, Feizi M (2005) Evaluation of salt-tolerant genotypes of durum wheat derived from in vitro and field experiments. Field Crops Res. 91: 345-354.
- Mansour MMF, Salama KHA, Al-Mutawa MM Abou Hadid AF (2002) Effect of NaCl and polyamines on plasma membrane lipids of wheat roots. Biol Plant. 45: 235–239.
- Muzaffar S, Ali B, Wani NA (2012) Effect of Catechol, Gallic acid & Pyrogallic acid on the germination, seedling growth & the level of endogenous phenolics in cucumber (*Cucumis sativus* L.); Int J Life Sc Bt and Pharm Res. 1(3): 51-59.
- Naqvi SSM, Ansari R, Kuawada AN (1982) Responses of salt stressed wheat seedlings to kinetin. Plant Sci Lett. 26: 279-283.
- Orphanos PI, Heydecker W (1968) On the nature of soaking injury to *Phaseolus vulgaris*. J Exp Bot. 19: 770-784.
- Parasher A, Varma SK (1988) Effect of pre-sowing seed soaking in gibberellic acid on growth of wheat (*Triticum aestivum* L.) under different saline conditions. Indian J Biol Sci. 26: 473-475.
- Pill WG (1995) Low water potential and pre-sowing germination treatments to improve seeds quality. In: Basra AS (ed) Seed Quality Basic Mechanisms and Agricultural Implications. The Haworth Press, New York, USA. 319-359.
- Prakash L, Prathapasenan G (1990) NaCl and gibberellic acid induced changes in the content of auxin, the activity of cellulose and pectin lyase during leaf growth in rice (*Oryza sativa* L.). Ann Bot. 365: 25-257.
- Rao SA, McNeilly T (1999) Genetic basis of variation for salt tolerance in maize (*Zea mays* L). Euphytica. 108: 145-150.
- Raptan PK, Hamid A, Khaliq QA, Solaiman ARM, Ahmed JU, Karim MA (2001) Salinity tolerance of black gram and mung bean: II- Mineral ions accumulation in different plant parts. Korean J Crop Sci. 46(5): 387-394.
- Rogis C, Gibson LR, Knapp AD, Horton R (2004) Can Solid Matrix Priming with GA_3 Break Seed Dormancy in Eastern Gama grass? J Range Manage. 57: 656-660.
- Rouhi HR, Abbasi AS, SharifZadeh F, Tavakkol RA, Aboutalebian MA, Vand GA (2011) Study of different priming treatments on germination traits of soybean seed lots. Not Sci Biol. 31: 101-108.
- Ruan SL, Xue QZ, Wang QH (2003) Physiological effects of seed priming on salt-tolerance of seedlings in hybrid rice (*Oryza sativa* L.). Sci Agric Sin. 36: 463–468.

- Ruan SL, Xue QZ (2002) Effects of chitosan coating on seed germination and salt tolerance of seedlings in hybrid rice (*Oryza sativa* L.). Acta Agron Sin. 28(6): 803-808.
- Sakhabutdinova AR, Fatkhutdinova DR, Bezrukova MV, Shakirova FM (2003) Salicylic acid prevents the damaging action of stress factors on wheat plants. Bulg J Plant Physiol. Special issue 314-319.
- Sen SK, Jha S, Mandal P (2016) Evaluation of Phytotoxicity of Mung Bean genotypes against Salinity Stress. Int J Sci Rep Pub. 6(12): 333-340.
- Sen SK, Mandal P (2016) Solid matrix priming with chitosan enhances seed germination and seedling invigoration in mung bean under salinity stress. JCentrl Europ Agric. 17(3): 749-762.
- Shannon MC, Rhoades JD, Draper JH, Scardaci SC, Spyres MD (1998) Assessment of salt tolerance in rice cultivars in response to salinity problems in California. Crop Sci. 38: 394-398.
- Singh H, Malik BP, Sharma HC, (1989) Relative performance of mungbean (*Vigna radiata* L. Wilczek) cultivars under varying levels of soil salinity. Haryana J Agron. 5(2): 171-73.

- Soloviev AA, Kuklev MY, Karnauakhova T (2003) Functional status of some genotypes of tomato under salinity conditions in green house. Acta Hortic. 609: 47-50.
- Tang D, Dean WL, Borchman D, Paterson CA (2006) The influence of membrane lipid structure on plasma membrane Ca2+-ATPase activity. Cell Calcium. 39: 209-216.
- Upadhyay RK, Panda SK (2005) Salt tolerance of two aquatic macrophytes, *Pistia stratiotes* and *Salvinia molesta*. Biol Plantarum. 49(1): 157-159.
- Walker MA, Dumbroff EB (1981) Effects of salt stress on abscisic acid and cytokinin levels in tomato. Z Pflanzenphysiol. 101: 46-470.
- Walker MA, Dumbroff EB (1981) Effects of salt stress on abscisic acid and cytokinin levels in tomato. Z Pflanzenphysiol. 101: 46-470.
- Wang YR, Zhang JQ, Liu HX (2004) Physiological and ecolpogical responses of alfalfa and milk vetch to PEG priming. Acta Ecol Sin. 24: 402-408.
- Zavariyan AM, Rad MY, Asghari M (2015) Effect of seed priming with kinetin on germination indices and proline activity of *Silybum marianum* L. under drought stress conditions. Int J Life Sci. 9(1): 6-11.