SCREENING OF PLANT GROWTH PROMOTING ENDOPHYTIC BACTERIA FOR SALINITY STRESS TOLERANCE

Sheheryar [1,] Muhammad Ali[1*,] Ikramullah[2], Roohul Amin[2], Khan Niaz Khan[3], Taimur Nawaz[1] and Shahzeb Khan[1]

1. Institute of Biotechnology & Microbiology, Bacha Khan University Charsadda, Pakistan
2. Department of Agriculture, Bacha Khan University Charsadda, Pakistan
3. Department of Biology Edwards College Peshawar, Pakistan

Corresponding Author
Muhammad Ali

Abstract
A variety of endophytic bacteria thrive inside plants, enhancing their growth under a variety of conditions. By modulating growth and stress hormones directly, they improve nutrient uptake and nutrient utilization. There are a number of ways in which endophytic bacteria can improve plant health indirectly, including antibiotics, hydrolytic enzymes, nutrient limitations, and priming plant defenses. Maize roots were used in the present study to isolate salt-tolerant bacteria. Bacteria were exposed to salinity stresses of 50mM, 100mM, 150mM, and 200mM and showed a tolerance of 200mM of NaCl. The LB media were used to produce indole-3-acetic acid (IAA). Ammonia was also produced by isolates, as confirmed by Nessler's reagent. It was found that the isolates tested positive for catalase. Based on all these findings, the endophytes might be useful in agriculture.
Keyword: endophytes, salinity, phytohormone, abiotic stress

INTRODUCTION

Endophytic bacteria have been known as microorganisms that could be extract from the sterilize surface of the plant tissues and do not visibly damage host plants (Petrini, 1991; Hallmann et al., 1997; Schulz and Boyle, 2006). But it is now considered that endophytism is a universal phenomenon (Kobayashi and Palumbo, 2000) and is similar that all plants harbor endophytic bacteria (Rosenblueth and Martínez-Romero, 2006; Ryan et al., 2008; Compant et al., 2010; Dudeja and Giri, 2014).

Endophytic bacteria is present in rhizosphere in a diverse range of organs (e.g., roots, stems, leaves, seeds, and fruits) of plants (Rayan et al ,2008). Beneficial endophytic bacteria linked with the plants which play an important role in the health and growth of plants and low yield in a different environmental stresses (H. Etesami, and D.K. Maheshwari,2018). Bacteria colonizing the roots of plants and have advantageous effects on plant growth and development which is known to be as plant growth promoting–rhizobacteria (PGPR) (J.W. Kloepper et al 1989). These bacteria have benefit on plant growth by various mechanisms (H. Etesami, and D.K. Maheshwari,2018). Endophyte bacteria, which colonize the internal tissue (the intercellular spaces) of plants rather shows any negative effects on their host, constitute a great reservoir of bacterial diversity with a striking biotechnological potential (Rayan et al ,2008). The interest in endophytic bacteria has more, as they colonize the internal tissues of their host plants and improve plant tolerance to various abiotic stress factors and can save plants from different pathogenic microbes (Malfanova et al., 2011; Hashem et al., 2016). Endophytic bacteria were found in a diverse range of plants containing crops, aromatic and medicinal plants, halophytes etc. (AzariasGuimarães et al., 2012; Sharma et al., 2012; Egamberdieva et al., 2017). Plant growth promoting endophytic (PGPE) bacteria are a group of microorganisms linked ecofriendly with the terrestrial plants which to be known for their advantagous effects on growth,yield and health of the host (Qin et al. 2011b; Reinhold-Hurek and Hurek 2011; Grönemeyer et al. 2012). The well-known mechanisms is referred to be a PGPE can be either direct or indirect such as nitrogen fixation for plant use, ammonia production, solubilization of mineral phosphate, sequestration of iron for plants by siderophores, production of plant hormones like auxins, cytokinins and gibberellins, and biological control of plant pathogens and deleterious microbes through the production of antibiotics and cell wall degrading enzymes (Lucy et al. 2004; Patel et al. 2012).

Endogenous bacterial endophytes is referred which is capable of breaking hydrocarbons are likely to be widespread, to date only a few studies have assessed the degradation potential inherent to endophytic associated of common plant are used in phytoremediation studies. Pseudomonas, Arthrobacter, Enterobacter, and Bacillus strains which extract from the root and stem tissues of a diverse range of popular cultivars growing at a car manufacturing plant were capable to break BTEX compounds (Porteous-Moore et al. (2006).

The distribution of these bacteria was spatially limited both within and between individual poplar cultivars. The bacterial populations were assessed which are linked with tall fescue and red clover growing at an aged-hydrocarbon contaminated site (Siciliano et al. (2001). These researchers found from 0.1% to 4% of culturable heterotrophic endophytic bacteria possessed genes involved in hydrocarbon degradation, but that there were significant plant specific differences in the prevalence of degrader genotypes. In some cases, as with alkB genotypes in tall fescue endophytic communities, there was increased degrader prevalence in endophytic communities relative to rhizosphere communities.

Endophytic bacteria exhibit complex interactions with their hosts which involves mutualism and antagonism. Their association can be obligate or facultative. Plants strictly limit the growth of endophytes, and these endophytes use many mechanisms to gradually adapt to their living environments. In order to maintain stable symbiosis, endophytes produce several compounds that promote growth of plants and help them adapt better to the environment (Ulrich K.et .al, (2008). Endophytes are capable to enter inside the plant hosts colonizing the intercellular spaces and also the xylem vessels.

Objectives: The objectives of the present study were to screen weather plant endophytic bacteria have a role in salinity stress and to identify the threshold level of salinity by the endophytic bacteria.

MATERIAL AND METHODS

1. Isolation of endophytic bacteria

A total of four soil samples were collected from the roots of the maize plants including the 1cm two samples from the surface and 5 cm two samples from under the surface. All soil samples were inoculated in double distilled water for a short period of time using four falcon tubes for each sample. After inoculation with loop few drops of inoculated soil samples were steaked on four L.B media plates.

2. Preparation of bacterial inocula:

To perform each of the tests in this study, the fresh inoculum of each isolate was prepared in the following way. Initially, endophytic bacterial isolates were re–cultured on LB medium. The 100 m L flask containing 20 mL of sterilized LB culture medium were inoculated with bacterial isolates. The cultures were incubated at 37 C for 48 h (for fast– growing bacteria) and 72 h (for slow–growing bacteria) on a shaker with a rotation speed of 120 rpm until (turbid) logarithmic phase was attained. The bacterial cells were harvested through centrifugation (7000 g for 10 min) after growth in LB culture medium and then washed twice with phosphate buffer and then suspended in the same buffer.

3. Characterization of bacteria on the basis of morphology

Bacteria was grown but changes in color, diameter, status and appearance of colonies were investigated after 24 hours of incubation. The tolerance of isolates to different levels of salinity was evaluated by observing the quality of the colonies grown in the control plates.

4. Salt tolerance test of isolates

For this purpose, NA medium containing different salinity percentages (50 to 1700 mM) NaCl salt) was used. At first, culture media including different concentrations of NaCl were distributed in sterile conditions in Petri dishes and fresh culture of each isolate with a uniform population was spotted onto the agar plates. For each isolate, three replicates were taken and the petri dishes were kept in the incubator at 37̊ C after sealing.

5. Biochemical Characterization

Catalase Test: Catalase activity of the isolates was estimated by using H2O2 solution onto the microscopic slides containing the culture of the isolates separately. Observations were taken for immediate bubble formation (O2 + water = bubbles). Release of O2 bubbles indicated positive catalase activity. It was used to differentiate staphylococci (positive) from streptococci (negative) the enzyme Catalase was produced by bacteria that respired using oxygen and protect them from toxic byproduct of oxygen metabolism. We took six plates of samples and dropped few drops of H2O2 on each plates by using dropper. As a result, bubbles like liquid were formed which showed positive catalase activity.

6. Nessler's reagent test

In an aqueous solution of potassium iodide (7.0gm), mercuric chloride (10.0gm) and sodium hydroxide which ass used to determine the presence of ammonia in specific material when this reagent detect ammonia in a particular substance, the color of the solution will change to yellow. First nester reagent was formed and then poured in six falcon tubes each with five micro liter then colonies of endophytic bacteria were picked by using toothpick and dropped in each of the six falcon tubes. After that we kept those falcon tubes in continuous shaker at 37̊ C for 12 hours the next day the solution of colour was changed which showed positive results.

RESULTS

Isolated endophytic bacteria were tolerant to Salinity stress (50mM to 200mM):

Endophytic bacteria were isolated from maize crops roots. First its plant growth promoting activities were analyzed by checking its tests for IAA production in order to know whether those bacteria has a role in salinity stress. The endophytic bacteria were exposed todifferent concentrations of salinity stresses 50mM,100mM,150 mM,and 200 mM stress of NaCl on LB respectively each; experiment was continue after successful growth of these bacteria on LB and salt stress media after growth on 1 plate of salt stress the level for salinity were evaluated gradually it was noted that the salt stress up to 200mM didn't inhibit the growth of these endophytic bacteria suggesting that these endophytic bacteria have a role of salinity stress. Isolate were known to have different level of maximum salt concentration stress tolerance for the growth and yield. But some bacteria were growing upto 200mM stress of Nacl but showed the same results and grow on plates as same as 50mM stress of Nacl. The temperature and various morphology of bacteria were recorded for each isolate. Morphology was helpful in identification of streptococcus and staphylococcus. For example, pseudomonas family. It confirmed that the bacteria were capable of growing to tolerate the salinity stress.

Isolated endophytic bacteria were tolerant to Salinity stress (400mM to 1000mM)

To analyze that the level of the endophytic bacteria can cope with the salinity stress a second round of salinity stress were given. The endophytic bacteria were exposed to different stresses after growing in one stress and the level of stress were elevated gradually. On exposure to 400mM to 1000mM it was observed that the endophytic bacteria were capable to grow under applied stresses without inhibiting their normal routine growth at 37̊ C, suggested that these bacteria's were capable of handling salinity stress regardless of its growth promoting activities as shown in fig 4.1.2 surprisingly there was no reduction at the growth rate of these bacteria.

Isolated endophytic bacteria screening for maximum salinity stress threshold level.

In order to characterize the maximum threshold level for salinity stress. Isolated endophytic bacteria were exposed to 1200mM, followed by 1500mM, 1650mM and 1800mM. The data suggested that these bacteria were resistant to 1200mM, 1500m, and 1650mM. However, at 1800mM these bacteria did not show any growth hence no colonies were seen on the plates as shown in figure 4.1.3. These results confirmed that these bacteria can handle salinity stress up to 1700mM which is the maximum capacity of these endophytic bacteria to cope with salinity.

Endophytic bacteria responded positively for ammonia production.

Nessler's reagent test was performed to determine the presence of ammonia in specific material when this reagent detect ammonia in a particular substance, the color of the solution changed to yellow which confirms that endophytic bacteria responded positively for ammonia production.

Selected endophytic bacterias were positive for Catalase test.

To differentiate staphylococci (positive) from streptococci (negative) by using H2O2 solution onto the microscopic slides containing the culture of the isolates separately. Observations were taken for immediate bubble formation as shown in fig. 4.3.

DISCUSSION

A specific amount of endophytic bacteria were isolated first and then grown on the agar media as it indicated endophytic isolates. The diversity and beneficial characteristics of endophytic microorganisms have been studied in Plectranthustenuiflorus medicinal plant. Endophyte as microorganism resist inside the plant without any regards to specific tissues, interactions between plants and bacteria help plants to settle in ecosystem restoration processes (Glick et al. 1995).

These interactions may increase the ability of plants to utilize nutrients from the soil by increasing root development, nitrate uptake or solubilizing phosphorus, and to control soil-borne pathogens. Recently, many known as well as new endophytic bioactive metabolites, possessing a wide variety of biological activities as antibiotic, antiviral, anticancer, anti-inflammatory, antioxidant, etc., have been identified .Therefore, it has a high potential to screen novel, highly active, and low toxicity antimicrobial substances from endophytes.(Strobel and Daisy 2003).

Lately there were some reports shown that endophytic isolates also behaves as bio-medicine. Characterization of endophytic bacteria was done on the basis of colony morphology, colony color, cell shape, motility, growth rate, and Gram reaction. The interaction of endophytic bacteria or isolates with microorganism contains nutrients and growth-influencing substances. The 51% of biologically active substances were isolated from the endophytic fungi (Strobel 2003).

Endophytic bacteria are non-pathogenic microorganism that lives or are present in the internal tissues of plant. In most of the research the characterization of the endophytic bacteria is done on the bases of the shape, size and color. As mention in this research the tests that were done on the endophytic bacteria some were positive and some have different results then our research. It also plays an important role in metabolism and physiology of plant.

By using media like nutrient agar media the endophytic bacteria are grown and on the basis of the morphological and biochemical characterization of the endophytes. Root and leaf tissue were selected and it was found that the population density and type of the Endophyte was more in root than in leaves. Many of the bacterial genera encountered in this work were previously reported by Lopez-Lopez *et al.*, and many species of genera *Bacillus* were found by Walker *et al.* in bean seeds. Endophytic population according to cultivar or clone has differences, some specific endophytic bacteria were only isolated from a single studied cultivar.

Endophytic bacteria are bacteria that live in plant tissues without doing substantive harm or gaining benefit other than residency. Both gram-positive and gram-negative bacterial endophytes have been isolated from several tissue types in numerous plant species. These variations are attributed to plant source, plant age, tissue type, time of sampling, and environment. Generally, bacterial populations are larger in roots and decrease in the stems and leaves. All of this research has done for the determination of the prevalence, properties, persistence, and types of endophytic bacteria in agronomic and native plants.

Modification of plants to obtain organisms with improved genetic capabilities and tolerance to different environmental conditions is generally carried out by plant breeding and by integrating foreign DNA into plant genomes to produce transgenic plants. The successful colonization of several crops with such microbes suggests that they can be utilized in future applications, such as delivery of degradative enzymes for controlling certain plant diseases or other useful products.

In our research we have taken samples of endophytic bacteria which were specifically taken from the maize plant and then grown in vitro in agar nutrient media, after the growth different stress tolerant tests were performed to check how much endophytic bacteria from maize have tolerance resistance (salt stress, nessler reagent test and others) with the help of these tests we examine different shape, size and colored bacteria and also the stress level ranks from 50mM to 1650mM.

CONCLUSION

Changes to the environment drastically damage plants, resulting in low yield and growth. In plants with ecofriendly approaches, plant stress tolerance is correlated with the associated microbes. Whereas endophytes are beneficial over rhizosphere protected from stress conditions. However, it was concluded that endophytes are potential tools for the improvement of plant growth in term of high yield and exploring the diversity of endophytes in many stages of plants and their role in growth. endophytic bacteria associated with halophytic bacteria are capable to tolerate high salt stress conditions.

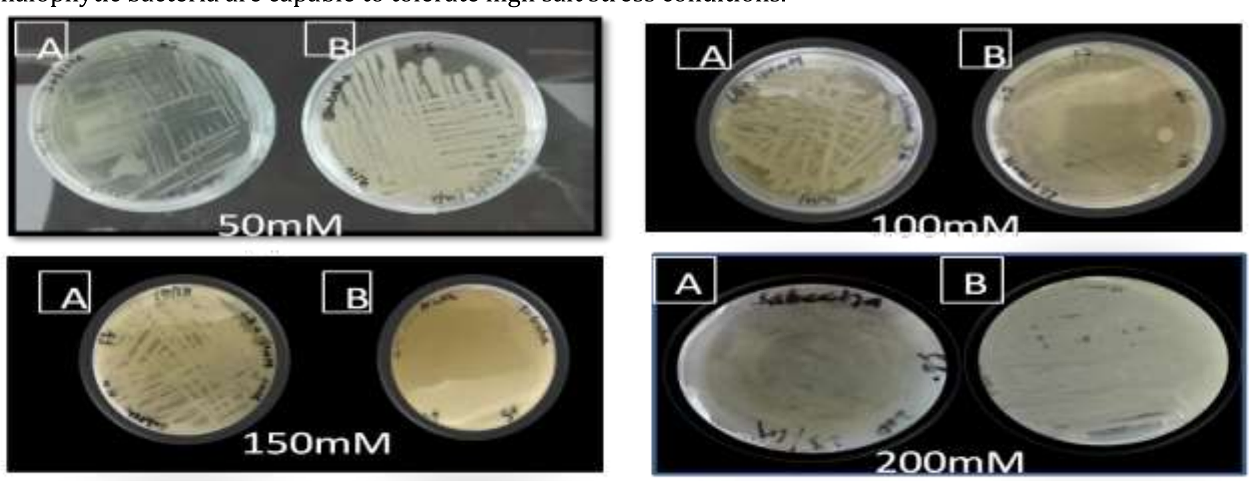

Fig. 4.1.1: Growth of endophytic bacteria under 50mM to 200 Nacl stress

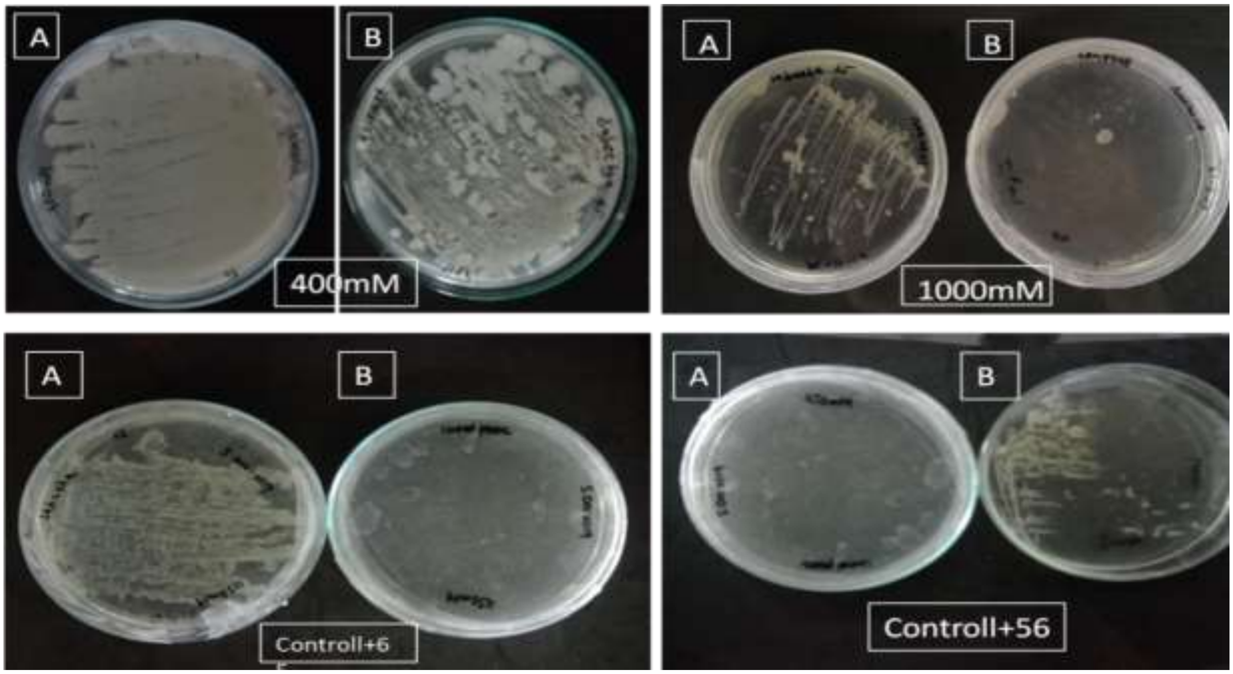

Fig. 4.1.2: Growth of endophytic bacteria under 400mM to 1000mM NaCl stress

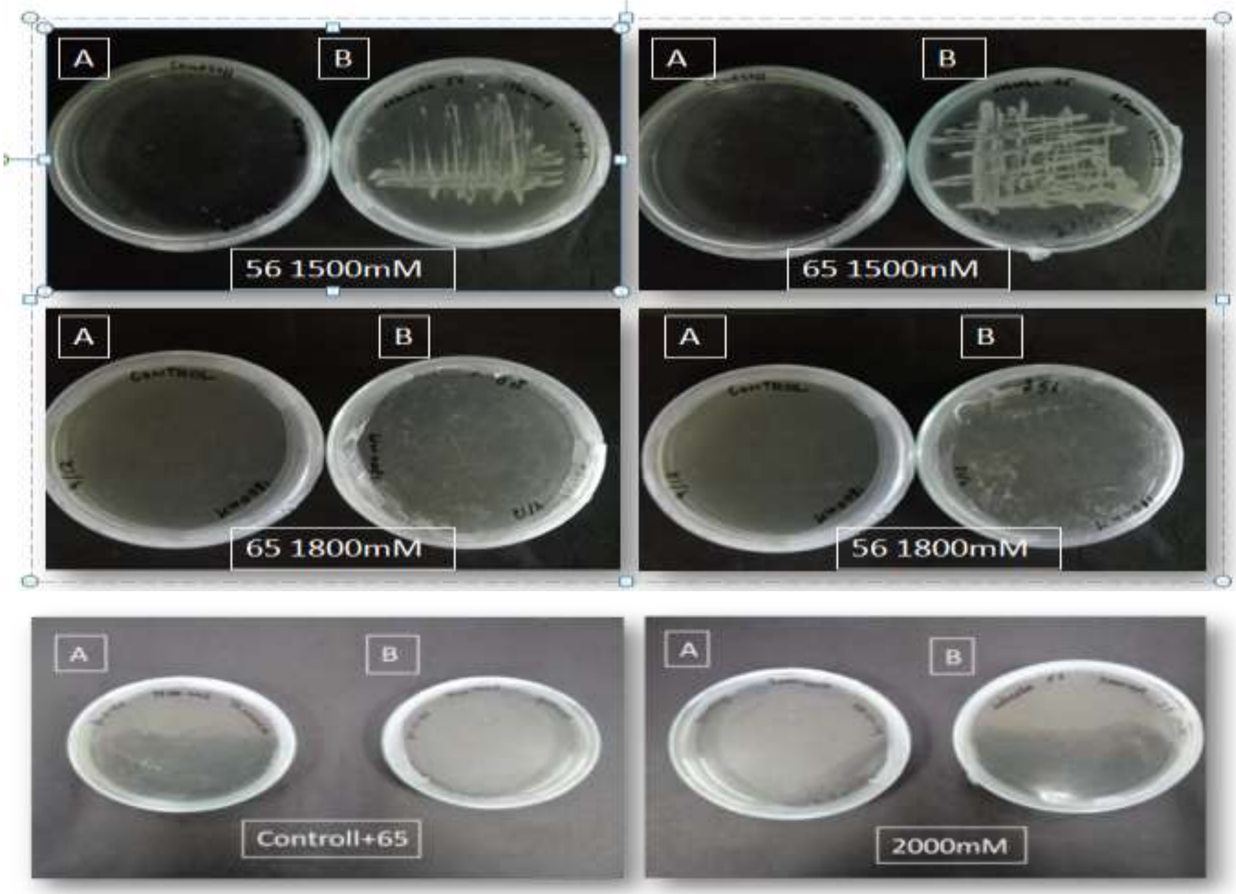

Fig. 4.1.3: Growth of endophytic bacteria at 2000mM Nacl stress

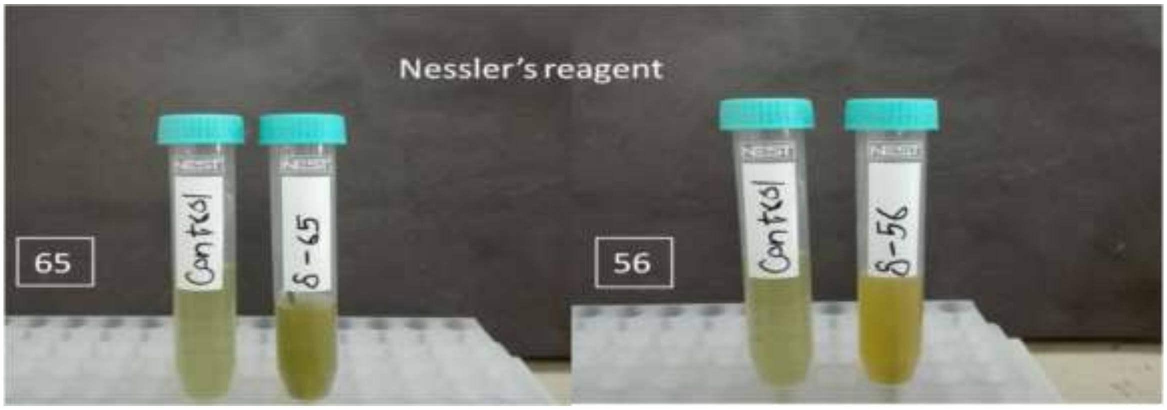

Fig. 4.2: Shows that the solution of colour was changed which shows positive results.

Fig. 4.3: Formation of O_2 bubbles which indicated positive catalyse activity.

Reference

1. Barnawal, D., Bharti, N., Pandey, S. S., Pandey, A., Chanotiya, C. S., and Kalra, A. (2017). Plant growthpromoting rhizobacteria enhance wheat salt and drought stress tolerance by altering endogenous phytohormone levels and TaCTR1/TaDREB2 expression. Physiol. Plant. 161, 502–514. doi: 10.1111/ppl.12614
2. Bassin, J. P., Dezotti, M., and Sant'Anna, G. L. Jr. (2011). Nitrification of industrial and domestic saline wastewaters in moving bed biofilm reactor and sequencing batch reactor. J. Hazard. Mater. 185, 242–248. doi: 10.1016/j.jhazmat.2010.09.024
3. Bharti, N., Pandey, S. S., Barnawal, D., Patel, V. K., and Kalra, A. (2016). Plant growth promoting rhizobacteria Dietzia natronolimnaea modulates the expression of stress responsive genes providing protection of wheat from salinity stress. Sci. Rep. 6:34768. doi: 10.1038/srep34768
4. Bhise, K. K., Bhagwat, P. K., and Dandge, P. B. (2017). Synergistic effect of Chryseobacterium gleum sp. SUK with ACC deaminase activity in alleviation of salt stress and plant growth promotion in Triticum aestivum L. 3 Biotech. 7:105. doi: 10.1007/s13205-017-0739-0
5. Bottini, R., Fulchieri, M., Pearce, D., and Pharis, R. P. (1989). Identification of gibberellins A1, A3, and iso-A3 in cultures of Azospirillum lipoferum. Plant Physiol. 90, 45–47. doi: 10.1104/pp.90.1.45
6. Carter, D. L. (1975). "Problems of salinity in agriculture," in Plants in Saline Environments, eds A. Poljakoff-Mayber and J. Gale (Berlin: Springer), 25–35. doi: 10.1007/978-3-642-80929-3_3

7. Chauhan, P. S., Lata, C., Tiwari, S., Chauhan, A. S., Mishra, S. K., Agrawal, L., et al. (2019). Transcriptional alterations reveal Bacillus amyloliquefaciens-rice cooperation under salt stress. Sci. Rep. 9, 1–13. doi: 10.1038/s41598-019-48309-8
8. Chen, H., and Jiang, J. G. (2010). Osmotic adjustment and plant adaptation to environmental changes related to drought and salinity. Environ. Rev. 18, 309–319. doi: 10.1139/A10-014
9. Cheng, Z., Park, E., and Glick, B. R. (2007). 1-Aminocyclopropane-1-carboxylate deaminase from Pseudomonas putida UW4 facilitates the growth of canola in the presence of salt. Can. J. Microbiol. 53, 912–918. doi: 10.1139/W07-050
10. Ciulla, R. A., Diaz, M. R., Taylor, B. F., and Roberts, M. F. (1997). Organic osmolytes in aerobic bacteria from mono lake, an alkaline, moderately hypersaline environment. Appl. Environ. Microbiol. 63, 220–226. doi: 10.1128/aem.63.1.220-226.1997
11. Daliakopoulos, I. N., Tsanis, I. K., Koutroulis, A., Kourgialas, N. N., Varouchakis, A. E., Karatzas, G. P., et al. (2016). The threat of soil salinity: a European scale review. Sci. Total Environ. 573, 727–739. doi: 10.1016/j.scitotenv.2016.08.177
12. Das, P., Behera, B. K., Meena, D. K., Azmi, S. A., Chatterjee, S., Meena, K., et al. (2015). Salt stress tolerant genes in halophilic and halotolerant bacteria: paradigm for salt stress adaptation and osmoprotection. Int. J. Curr. Microbiol. App. Sci. 4, 642–658.
13. Deinlein, U., Stephan, A. B., Horie, T., Luo, W., Xu, G., and Schroeder, J. I. (2014). Plant salt-tolerance mechanisms. Trends Plant Sci. 19, 371–379. doi: 10.1016/j.tplants.2014.02.001
14. Dietz, K. J., Vogel, M. O., and Viehhauser, A. (2010). AP2/EREBP transcription factors are part of gene regulatory networks and integrate metabolic, hormonal and environmental signals in stress acclimation and retrograde signalling. Protoplasma 245, 3–14. doi: 10.1007/s00709-010-0142-8
15. Eakins, B. W., and Sharman, G. F. (2010). Volumes of the World's Oceans from ETOPO1. Boulder, CO: NOAA National Geophysical Data Center.
16. Egamberdieva, D., Jabborova, D., and Hashem, A. (2015). Pseudomonas induces salinity tolerance in cotton (Gossypium hirsutum) and resistance to Fusarium root rot through the modulation of indole-3-acetic acid. Saudi J. Biol. Sci. 22, 773–779. doi: 10.1016/j.sjbs.2015.04.019
17. Erice, G., Ruíz-Lozano, J. M., Zamarreño, ÁM., García-Mina, J. M., and Aroca, R. (2017). Transcriptomic analysis reveals the importance of JA-Ile turnover in the response of Arabidopsis plants to plant growth promoting rhizobacteria and salinity. Environ. Exp. Bot. 143, 10–19. doi: 10.1016/j.envexpbot.2017.08.006
18. Etesami, H., Emami, S., and Alikhani, H. A. (2017). Potassium solubilizing bacteria (KSB): mechanisms, promotion of plant growth, and future prospects A review. J. Soil Sci. Plant Nutr. 17, 897–911. doi: 10.4067/S0718-95162017000400005
19. Etesami, H., Mirseyed Hosseini, H., and Alikhani, H. A. (2014). In planta selection of plant growth promoting endophytic bacteria for rice (Oryza sativa L.). J. Soil Sci. Plant Nutr. 14, 491–503. doi: 10.4067/S0718-95162014005000039
20. Fasciglione, G., Casanovas, E. M., Yommi, A., Sueldo, R. J., and Barassi, C. A. (2012). Azospirillum improves lettuce growth and transplant under saline conditions. J. Sci. Food Agr. 92,
21. Ferreira, M. J., Silva, H., and Cunha, A. (2019). Siderophore-producing rhizobacteria as a promising tool for empowering plants to cope with iron limitation in saline soils: a review. Pedosphere 29, 409–420. doi: 10.1016/S1002-0160(19)60810-6
22. Gill, S. S., and Tuteja, N. (2010). Reactive oxygen species and antioxidant machinery in abiotic stress tolerance in crop plants. Plant Physiol. Biochem. 48, 909–930. doi: 10.1016/j.plaphy.2010.08.016
23. Goel, D., Singh, A. K., Yadav, V., Babbar, S. B., Murata, N., and Bansal, K. C. (2011). Transformation of tomato with a bacterial codA gene enhances tolerance to salt and water stresses. J. Plant Physiol. 168, 1286–1294. doi: 10.1016/j.jplph.2011.01.010

24. Golldack, D., Lüking, I., and Yang, O. (2011). Plant tolerance to drought and salinity: stress regulating transcription factors and their functional significance in the cellular transcriptional network. Plant Cell Rep. 30, 1383–1391. doi: 10.1007/s00299-011-1068-0
25. Gupta, B., and Huang, B. (2014). Mechanism of salinity tolerance in plants: physiological, biochemical, and molecular characterization. Int. J. Genomics. 2014:701596. doi: 10.1155/2014/701596
26. Gupta, J., Rathour, R., Singh, R., and Thakur, I. S. (2019). Production and characterization of extracellular polymeric substances (EPS) generated by a carbofuran degrading strain Cupriavidus sp. ISTL7. Bioresour. Technol. 282, 417–424. doi: 10.1016/j.biortech.2019.03.054
27. Haggag, W. M. (2010). The role of biofilm exopolysaccharides on biocontrol of plant diseases. Biopolymers 14, 271–284. doi: 10.5772/10266
28. Halo, B. A., Khan, A. L., Waqas, M., Al-Harrasi, A., Hussain, J., Ali, L., et al. (2015). Endophytic bacteria (Sphingomonas sp. LK11) and gibberellin can improve Solanum lycopersicum growth and oxidative stress under salinity. J. Plant Interact. 10, 117–125. doi: 10.1080/17429145.2015.1033659
29. Hardy, N., Shainberg, I., Gal, M., and Keren, R. (1983). The effect of water quality and storm sequence upon infiltration rate and crust formation. J. Soil Sci. 34, 665–676. doi: 10.1111/j.13652389.1983.tb01063.x
30. Howell, S. H., Lall, S., and Che, P. (2003). Cytokinins and shoot development. Trends Plant Sci. 8, 453–459. doi: 10.1016/S1360-1385(03)00191-2
31. Hunte, C., Screpanti, E., Venturi, M., Rimon, A., Padan, E., and Michel, H. (2005). Structure of a Na+/H+ antiporter and insights into mechanism of action and regulation by pH. Nature 435, 1197–1202. doi: 10.1038/nature03692
32. Jaiswal, D. K., Verma, J. P., Prakash, S., Meena, V. S., and Meena, R. S. (2016). "Potassium as an important plant nutrient in sustainable agriculture: a state of the art," in Potassium Solubilizing Microorganisms for Sustainable Agriculture, eds V. S. Meena, B. R. Maurya, J. P. Verma, and R. S. Meena (New Delhi: Springer), 21–29. doi: 10.1007/978-81-322-2776-2_2
33. Jha, Y., and Subramanian, R. B. (2014). PGPR regulate caspase-like activity, programmed cell death, and antioxidant enzyme activity in paddy under salinity. Physiol. Mol. Biol. Plants 20, 201–207. doi: 10.1007/s12298-014-0224-8
34. Jha, Y., Subramanian, R. B., and Patel, S. (2011). Combination of endophytic and rhizospheric plant growth promoting rhizobacteria in Oryza sativa shows higher accumulation of osmoprotectant against saline stress. Acta Physiol. Plant 33, 797–802. doi: 10.1007/s11738-010-0604-9
35. Joshi, R., Paul, M., Kumar, A., and Pandey, D. (2019). Role of calreticulin in biotic and abiotic stress signalling and tolerance mechanisms in plants. Gene 714:144004. doi: 10.1016/j.gene.2019.144004
36. Kalaji, H. M., Bosa, K., Kościelniak, J., and Żuk-Gołaszewska, K. (2011). Effects of salt stress on photosystem II efficiency and CO2 assimilation of two Syrian barley landraces. Environ. Exp. Bot. 73, 64–72. doi: 10.1016/j.envexpbot.2010.10.009