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Effect of PGPR isolates on Plant growth promotion in relation to salinity stress

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ABSTRACT
PGPRs are directly or indirectly involved in promoting growth and development of the plant by producing certain phytohormones through different mechanisms. Various studies revealed that application of plant growth promoting microbes can increase the crop production and yield even under various biotic and abiotic stress conditions. Different direct and indirect mechanisms are involved in the plant growth promotion of plant.Direct mechanism include nitrogen fixation, phytohormone production (Indole-3-acetic acid, cytokinins, gibberellins, abscisic acid), phosphate solubilization, ACC deaminase production whereas, indirect mechanism includes antibiotics production, HCN production, siderophore formation, induced systemic resistance, systemic acquired resistance, polysaccharide and antioxidant production. Role of plant growth promoting microbes under salinity stress condition is being discussed.
Keywords: *Rhizosphere, PGPRs, Salinity stress, Phytohormones, Phosphate solubilisation, Nitrogen fixation and ACC deaminase.*

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INTRODUCTION

Agriculture is the largest financial source of our country. About 1.3 billion people of the world are directly dependent on agriculture. For sustainable agriculture there is a need to maintain the soil dynamic. In India, 60.6% of land is used for the purposes of agricultural by half of the population of the country inorder to grow vegetables, pulses and cereals. There are various factors that are responsible for the change in climate, water quality and ultimately agricultural productivity [1]. The two major threats for the agricultural sustainability are increasing human population and decreasing availability of land for cultivation. Depletion of field nutrients combined with the effects of poor land fertility is one of the major cause of reduction in crop yield. Cultivation of agricultural crops and it productivity is affected by various environmental stresses. There are two major types of environmental stresses biotic and abiotic stresses. Abiotic stresses include high and low temperature, soil salinity, drought, flood, wind etc. And among these abiotic stresses, salinity stress is one of the most destructive environmental stresses which cause reduction in crop productivity and its quality. Drought and salinity are the major abiotic stresses which affect the productivity of the crop and reduce the average yield up to 50% or more [2]. Salinity stress is defined as the presence of excess amounts of soluble salts in the soil which affects or hinders the normal growth and functioning of the plant. Worldwide, there is an increase in the area of salinity and hence there is a need to develop such crop varieties that can better perform under the stress condition i.e. developing salinity resistant crop varieties. The problem of soil salinity is a naturally occurring issue of arid and semiarid regions and is increasing day by day due to the extensive use of chemical fertilizers and improper irrigation system [3]. According to the report of United Nations, upto 50% of the world's arable lands are subjected to salinity stress [4]. There is an estimate that about 70% of the crop yield loss can be attributed to abiotic stresses. In the modern era agriculture land is reducing therefore abiotic stress is a problem of great concern for the growth and productivity of crop. So, in order to overcome from this problem use of plant growth promoting microbes can be a better and ecofriendly method.Rhizosphere is enriched with microbial diversity and the bacteria residing in the rhizhospheric region are called rhizobacteria[5]. Bacteria linked with plants are categorized into different groups such as beneficial, deleterious and neutral according to their effects on plants. Beneficial bacteria living in the rhizosphere are known as plant growth promoting rhizobacteria. In 1970s Kloepper introduced the term PGPR and described PGPR as bacteria living in soil that colonize plants roots and improves and enhance the growth of plant [6]. The microbes colonizing rhizosphere are bacteria, fungi, acticomycetes, protozoa, and algae and among them bacteria is the most abundant microbe present in the rhizosphere. PGPR can also be termed as plant health promoting rhizobacteria or nodule promoting rhizobacteria [7].PGPR can also be divided into two groups according to their residing sites: iPGPR, symbiotic bacteria that lives inside the plant cells, produce nodules and ePGPR, a free-living rhizobacteria that lives outside the plant cells, do not produce nodules but still promotes plant growth. PGPRs play an important role in conferring plant tolerance to abiotic stresses such as drought and salinity. *Bacillus* strains such *as B. subtilis, B. cereus, B. licheniformis, B. pumilus, B. amyloliquefaciens, B. polymyxa,* and *B. Megaterium* have been reported to successfully colonize the roots and rhizosphere of several plants such as tomato, banana, canola, wheat, apple, red pepper, and Arabidopsis resulting in promotion of plant growth and yield, disease resistance, drought tolerance, and heavy metal remediation [8]. Varieties of PGPR such as *Pseudomonas, Bacillus, Enterobacter, Klebsiella, Azobacter, Variovorax, Azosprillum,* and *Serratia* have been commercialized used.The working mechanisms of PGPR can be separate either direct or indirect ones. The mechanisms of PGPR include biofertilization, Nitrogen fixation, and production of phytohormones, minerals solubilization, siderophore activity, antibiotics production, and antagonistic activity such as competition, parasitism ACC deaminase activity, and induction of plant systemic resistance.

ABIOTIC AND BIOTIC STRESSES

Stress can be defined as any factor which has a negative impact on plant growth and development. Plant growth and development is influenced by a variety of stresses. These stresses are classified into two major groupsi.e., biotic and abiotic. Biotic stresses are developed by interaction between organisms. Biotic stresses are result of competition between the organisms for the resources. It can be also developed by parasitism and predation due to the actions of allelopathic chemicals released by one organism and inhibiting the growth of the other. Diseases are also a kind of biotic stresses. Biotic stress is caused by certain biotic agents such as bacteria, fungi, virus, insects, viroids and nematodes which results in significant reduction in the crop yield. Abiotic stress is the primary cause of crop loss, worldwide by more than 30%. Abiotic stresses such as salinity, drought, alkalinity, flood, cold, heat, radiation, heavy metal, air pollution and several others are the major causes restricting agriculture produce and adversely influencing plant growth processes. Aridity stress imparted by drought, salinity, and high temperature is the most dominant abiotic stress limiting plant growth and productivity. Among several abiotic stresses salinity stress is responsible for huge crop losses. In the modern era agriculture land is reducing therefore abiotic stress is a problem of great concern for the growth and productivity of crop.

SALINITY STRESS

Salinity stress is one of the most major abiotic stress factors in modern agriculture which effects the growth and development of the plant. Salinity stress is defined as the presence of excess amounts of soluble salts in the soil which hinders the normal growth and functioning of the plant. It is measured in terms of electrical conductivity (ECe). Soil salinization is caused either by natural activities or human activities which leads to the increase in the concentration of dissolved salts in the soil. Primary salinity in the soil is caused by natural processes which lead to the accumulation of salt in soil and ground water. Primary source of salinity may arise from weathering of rocks. Excessive irrigation, inadequate drainage and deforestation are the cause of secondary salinity. Salt is deposited on the top soil as the water evaporates which results in dry land salinity and forms a salt scald. Salinity effect is more destructive when ground water table is high, as in arid and coastal areas where only salt tolerant plants are able to grow. Irrigated lands are more prone to salinity than dry lands due to salt deposition because of irrigated water. Salt from the soil is also discharged by surface runoff and leached down into soil profile by rainfall and then move to water bodies. Salinity is a major problem in arid and semiarid regions of the world where there is an insufficient rain to leach away the soluble salts from the root zone. Salt stress affects the plants by reducing the plants ability to take up water which leads to slower growth and this effect is known as osmotic or water-deficit effect of salinity. Most of the agricultural lands are highly affected by salinity due to certain reasons. Salinity stress results in poor microbial activity because of toxic effect of ions and osmotic stress which leads to reduction in plant growth and development. Salinity is a form of physiologically dry habitat under which plants are unable to uptake substantial amount of water which results in disturbed physiological and metabolic activity of the plant. The salinity in soil is caused by anions such as chloride and nitrate and cations such as sodium, calcium, potassium.

SALINITY STRESS: CHALLENGE FOR AGRICULTURE

Increased accumulation of Na^+ and Cl^-ions have a detrimental impact on plant and leads to stunted plant growth, effects seed germination, ultimately reduces the crop yield and is toxic to plant cell as it affects the enzyme activity. The presence of soluble salts in the root zone of plant decreases water uptake through root from soil and accumulation of higher amount of salt water within cell are toxic to plant hence suppressing the plant growth. High amount of essential minerals such as calcium, sodium chloride, sodium carbonate and calcium chloride and also disturbs physicochemical and ecological balance. Salinity stress affects the plants in certain ways such as alteration in metabolic pathways, water stress, ion toxicity, nutritional disorders, oxidative stress, reduction in cell division, expansion, genotoxicity and membrane disorganization. It also causes adverse effect on nodulation process, reducing nitrogen fixation and crop yield. Nitrogen fixation is one on the most important process that is being highly affected under salt stress condition. The Nitrogenase enzyme responsible for nitrogen fixation is being reduced during salinity stress condition. In the recent time soil salinity is one of the challenging tasks for the farmers as well as the agricultural scientist. Accumulation of toxic ions such as Na^+ and Cl^- ions and nutrient imbalance in the soil results a negative impact on microbial activities as well as on plant growth. It has been reported that inoculation with plant growth promoting microbes and endophytic microbes reduces the negative impact of salt on different plants. Plant growth promoting microbes can promote plant growth under salinity stress condition through various direct and indirect mechanisms. In saline condition, Lettuce seed inoculated with *Azospirillum* showed better germination and vegetative growth in comparison to the control [9].Inoculation of plant with growth-promoting bacteria *Pseudomonas stutzeri* to salt tolerant and salt susceptible chilli pepper reduce negative effect on soil salinity [10]. Co-inoculation of AM fungi along with salt tolerant bacteria significantly improves the salinity tolerance in certain plants. This fungal and bacterial association exhibit significant impact on salinity tolerance in maize plant [11].

DIRECT MECHANISM

Nitrogen Fixation:

Nitrogen (N) is one of the chief plant nutrients for agricultural ecosystems, which is lost due to heavy rainfall and mineral leaching. Biological Nitrogen fixation can occur in bulge in rhizospheric soil. Fixed nitrogen can then be utilised and contributed by the crop through root uptake. Varieties of PGPR strains including *Klebsiella pneumoniae, Pantoea agglomerans, Azoarcus* sp., *Beijerinckia* sp., and *Rhizobium* sp.have the ability to fixed the atmospheric N_2 in soil [12] and make it available to plants. The variety of nitrogen fixing bacteria associated with the rhizosphere have been increasingly used in non-leguminous crops such as Sugar beet, Sugarcane, Rice, Jatropha, Maize, and Wheat. The Nitrogen fixing *Bacillus* strains and *A. Brasilense*sp.246 have the ability to promote the plant growth activity of spring wheat and barley agronomy in organic and low-N input agriculture [13]. In the recent years many bacterial isolates such as *Klebsiella, Burkholderia, Enterobacter,* and *Stenotrophomonas,* have been gaining attention because of their potential to enhance the plant growth and their association with many important crops [14]. Many N-fixing bacterial strains such as *Bacillus* OSU-142, *Pseudomonas putida* RC06, *Paenibacillus polymyxa* RC05 and RC14 have great potential to be used as a bio fertilizer for maintaining the better yield and the quality of spinach, wheat and sugar beet growth [15]. Recently, the PGPR strain, *Pseudomonas fluorescens* B16, displaying vigorous colonization in the tomatoes roots, increasing the plant height, flower number and total fruit weight. In the present day of agriculture the consortia of rhizobacterial species also improve the quality of soil and are also seemed to be a potent area of research. For example, the consortia of *Bradyrhizobium* sp., with *Pseudomonas striata* have established improved nodule occupancy in soya bean and increasing the biological N2 fixation. There are two mechanism by which PGPR can fix atmospheric nitrogen such as: symbiotic and non-symbiotic.

(a) Symbiotic nitrogen fixation is a mutualistic interaction between the plant and a microbe. The microbe (Rhizobia) first enters the root colonizing there and form root nodules with leguminous plants where nitrogen is fixed to ammonia and make it available for the plant. Many PGPR such as are *Rhizobium, Bradyrhizobium, Sinorhizobium,* and *Mesorhizobium*s showed the symbiotic association with leguminous plants, while *Frankia* with non-leguminous trees and shrubs [16].

(b) Non-symbiotic nitrogen fixation is carried out by free living diazotrophs and nitrogen fixing bacteria such as *Acetobacter, Azospirillum, Pseudomonas, cyanobacteria (Anabaena, Nostoc), Azoarcus, Azotobacter, Enterobacter, Gluconacetobacter* and *Burkholderia* which stimulate the growth of non-leguminous plants such as radish and rice [17]. Nitrogenase (*nif*) genes are found in both symbiotic and free living systems which help in nitrogen fixation [18]. *Nif* genes include structural and regulatory genes, structural genes play an important role in the iron molybdenum cofactor biosynthesis, activation of the Fe protein, and electron donation and the regulatory genes required for the synthesis and function of the enzyme. Inoculation by biological nitrogen fixing PGPR on crop provides an integrated approach for growth promotion activity, maintaining the nitrogen level in soil and for disease management.

Phytohormone production: Phytohormones such as indole-3-acetic (IAA) acid (auxin), gibberellins, cytokinins, abscisic acid and Ethylene are those substances which at lower/equal to micromolar concentrations stimulate the plant growth and development.

Indole-3-acetic acid (IAA)

Indole acetic acid is the primary phytohormone that improves root, shoot growth and seedling vigour while accelerating the plant growth and development as they involve in cell division, cell differentiation, nodule formation and vascular bundle formation. Patten and Glick [19] assessed that 80 % of bacteria isolated from the rhizosphere can produce IAA (including *A. caulinodans, B.japonicum, B. elkanii, M. loti, R. japonicum, R. leguminosarum, R. lupine, R. meliloti, R. phaseoli, R. trifolii* and *Sinorhizobium* spp. Indole-3-pyruvic acid and Indole-3-acetic aldehyde pathway are the precursors for IAA production in rhizobium. It has been reported that 60-fold IAA was increased on inoculation of *R. Leguminosarum* bv. viciae in the nodules of vetch roots while the inoculation with *B. japonicum*-SB1 with *B. Thuringiensis*KR1 reported the highest productions of Indole-3-acetic acid [20]. Co-inoculating *Pseudomonas* with *R. galegae* bv. Orientalis that produce IAA had shown to increases in shoot and root growth, nodule number and nitrogen content. There are various factors that influence the biosynthesis of IAA such as environmental stress factors (acidic pH, matrix stress, osmotic and carbon limitation) and genetic factors (auxin biosynthesis genes and their mode of expression) [21].

Cytokinins

Cytokinin stimulates plant cell division and also helps in the development of root and root hair formation. It has been documented by Nieto and Frankenberger [22] that 90 % of rhizospheric microorganisms have the ability to release cytokinins and about 30 growth-promoting cytokinin group compounds has been identified from microbial origin. Rhizobium strains are reported as the potent producers of cytokinins [23].

Gibberellins

Gibberellins, the phytohormones denoted as GA1 to GA89 depending on the approximate order of their discovery and are responsible for stem elongation and leaf expansion. It is also supposed that gibberellin has no effect on roots but it causes certain types of dwarfness due to gibberellin deficiency. Gibberellins have many applications such as it promote bolting of the plants, increase fruit size and number of buds, parthenocarpy in fruits and break down the tuber dormancy. They also control the flowering, sex expression of flowers and help in seed germination as in the case of lettuce. Plant growth promoting microbes including *Rhizobium, S. meliloti* are described to produce gibberellins [24].

Abscisic acid

Abscisic acid partially synthesized in the chloroplasts and the whole biosynthesis is primarily occurs in the plant leaves. The abscisic acid is produced when plants are exposed to stresses such as water deficit and freezing temperatures and their biosynthesis occurs indirectly through carotenoids production. Their transportation occurs primarily in both xylem and phloem tissues and also can be translocated through parenchyma cells. Like Auxin, abscisic acids do not show the polarity movement [25]. It was reported to inhibit shoot growth while not affecting or promoting root growth, stimulate the stomatal closure, induce seeds to deposit proteins and in dormancy induce gene transcription and there by provide pathogen defence [26]. It has been reported that *Rhizobium* sp. and *B. Japonicum* have the ability to produce abscisic acid [24].

Mineral phosphate solubilization

Several soil microorganisms (PGPB) have the ability to solubilize insoluble phosphorous into soluble formand make it possible for plants use [27]. Their availability ranges from 1 μmol l^{-1} in soil, while plants require approx 30 μmol l^{-1} to reach their maximum productivity. Most of the phosphatic fertilizers are not efficiently taken up by the plants due to re-precipitated into insoluble mineral complexes. Many higher plants group evolved highly systemic mechanisms for phosphate absorption even from very dilute solutions and attains the maximum growth rates even with soil solution phosphate levels of 2 μmol l-or less. Gaur [28] reported that inorganic phosphates compounds solubilize by microbes have great economic importance in plant nutrition. Microbes such as *Achromobacter, Agrobacterium, Erwinia, Escherichia, Bacillus, Pseudomonas, Enterobacter, Flavobacterium, Mycobacterium* and *Serratia* are highly systemic in solubilizing unavailable phosphate into available inorganic phosphate ion. Soil also contains a variety of organic substrates, which can be a source of P for plant growth and development but this P is need to be hydrolysed to inorganic P to make available for plant nutrition. Many enzymes like phosphonoacetate hydrolase, phosphatase, D-α-glycerophosphatase, phytase, and C-P lyase help in the mineralization of most organic phosphorous compounds [7]. Many phosphatases activity was significant in the inner rhizosphere at acidic and neutral soil pH of barley, wheat and maize. The bacterial strains such as *Enterobacter, Serratia, Pseudomonas, Citrobacter, Proteus, Rhizobium, Klebsiella* and*Bacillus* expressing a significant level of acid phosphatases. Phosphate-solubilizing bacteria (PSB) viz. *Arthrobacter ureafaciens, Rhodococcus erythropolis, Phyllobacterium myrsinacearum,* and *Delftia* sp. have the ability to solubilize extensive amounts of tricalcium phosphate (TCP) in the medium by secreting organic acids.

Mechanism for phosphate solubilization

Many Phosphate-solubilizing microorganisms (PSM) solubilize the insoluble forms of the phosphate to soluble forms via different mechanisms. They release the organic acids which act as good chelators of Ca^{2+} ions accompanying discharge of phosphates from insoluble phosphatic compounds. Organic acids may form soluble complexes with metal ions associated with insoluble Phosphorous, thus releasing the phosphate [29]. Many of the Phosphate Solubilizing Microorganisms secrete the organic acids such as lactic, malic, acetic, succinic, tartaric, oxalic, citric and gluconic acids or H+ extrusion which lowers the pH of the medium [29].Inorganic phosphorous in acidic soils is associated with Aluminium and Iron compounds, whereas Ca^{2+} phosphates are the major form of inorganic phosphates in calcareous soils. The major form of phosphorous is Phytate (a hexaphosphate salt of inositol) in organic matter, according between 50 and 80% of the total organic Phosphorus [30]. Although phytases are produced by microorganisms which hydrolyzethe phytate, that tends to accumulate in virgin soils due to complex molecules formation with Fe, Al and Ca [30]. Nucleic acids and phospholipids are another pool of imbalanced phosphorous in soil which is easily available to most of the present organisms. The maintenance is necessary for Phosphorous in the labile pool and how readily it is released from the solid phase into the soil is depends on the Phosphorous buffering capacity or the soil. Holford described three important soil components which control the P supply from the labile pool to refill crop extraction. These include the concentration of Phosphorous in the soil solution; the amount of phosphorous in the replacement source that enters into equilibrium with the soil solution phase and the phosphorous buffering capacity of the soil.

1-aminocyclopropane-1-carboxylic acid (ACC) Deaminase

1-aminocyclopropane-1-carboxylic acid deaminase is considered to be under tryptophan synthase family and a member of a large group of enzyme that utilizes vitamin B6. PGPR has the ability to metabolise the ACC and convert it into α-ketobutyrate and NH_3 which is used as a source of nitrogen. Inoculation of plant with PGPR producing ACC deaminase enzyme, ethylene levels are lowered in plants and result in longer roots providing relief from various stresses like drought, radiation, salinity, heavy metals, pathogens etc. Bacterial strains such as *R. leguminosarumbv. viciae, R. hedysari, R. japonicum, R. gallicum, B. japonicum, B. elkani, M. loti* and *S. meliloti* had been known to produce ACC deaminase. Glick [31] reported that those bacteria which are IAA producing they produce high levels of ACC deaminase and known to inhibit 2nd peak of ethylene levels during salinity stress. Hence, inoculation with these bacteria had shown beneficiary effects on root elongation, shoot growth, enhanced rhizobial nodulation and minerals uptake such as N_2, P, K, Fe, Ca, etc. The structural acds gene of (ACC deaminase) in *Mesorhizobium* sp. is under the control of nif promoter which controls the nitrogen fixing gene.

INDIRECT MECHANISM

Antibiotics

Plant growth promoting microbes have the ability to produce a variety of antibiotics such as 2,4-diacyetyl phloroglucinol (DAPG), kanosamine, phenazine-1-carboxylic acid, pyoluteorin, neomcycin A, pyrrolnitrin, pyocyanin, amphisin, tensin, tropolone, oligomycin A, zwittermicin A and viscosinamide. And among these antibiotics2,4-diacyetyl phloroglucinol (DAPG) has a broad spectrum antifungal, antibacterial and antihelminthic activity.*Pseudomonas* sp. producing antibiotic 2, 4-diacetylphloroglucinol (2, 4- DAPG) was reported for the biocontrol of wheat disease caused by the fungus *Gaeumanomyces graminis var. Tritici.* Production of lipopeptide and polyketideby *Bacillus amyloliquefaciens*helps in plant growth promotion and biological control activity towards soil borne pathogens. One issue with depending too much on these antibiotics producing plant growth promoting rhizobacteria as biocontrol agents is it develops resistant in plant pathogens to specific antibiotics due to increased use of these PGPR strains. And to overcome from this researchers are using those biocontrol strains that are capable to synthesize more than one antibiotic.

Hydrogen Cyanide (HCN)

Hydrogen Cyanide is a secondary metabolite commonly produced by rhizosphere *Pseudomonads.* Hydrogen Cyanide gas is potential and environmentally compatible mechanisms for biological control of weeds biologically as HCN gas negatively affect root growth and its metabolism. *Pseudomonas* and *Bacillus* sp. have the capability of producing hydrogen cyanide gas. HCN producing rhizospheric isolates have the ability to promote plant growth. Chickpea rhizospheric soil isolates exhibits certain PGPR traits including HCN production, which directly, indirectly or synergistically promotes plant growth. *Bacillus* and *Pseudomonas* sp. isolated from rhizospheric soils of mustard produces HCN gas and application of herbicides (quizalafop-p-ethyl &clodinafop) doesn't show any significant change in HCN production. *Pseudomonas entomophila* an entomopathogenic bacterium producing HCN as a secondary metabolite that can be used a biocontrol property.

Siderophore formation

Iron (Fe) is an essential plant micronutrient present in soil. Siderophores are defined as a small high-affinity iron chelating compounds secreted by certain microbes such as bacteria and fungi. Siderophores are known as iron carriers. Bacteria having the ability to synthesis low molecular weight compounds are known as siderophores capable of sequestering Fe^{3+}. Siderophores are known to have high affinity for Fe^{3+}, and thus making the iron available for plants. Rhizobacteria reduces Fe^{3+} ions are to Fe^{2+}. The siderophores are water soluble and can be extracellular and intracellular. Siderophores form stable complex with heavy metals such as Al, Cd, Cu etc. and radionucleides [32].Siderophore producing rhizobacteria can relieve plants from heavy metal stress and helps in iron uptake. *R. meliloti, R. tropici, R. leguminosarumbv.viciae, R. leguminosarumbv. trifolii, R. leguminosarumbv. phaseoli, S. meliloti* and *Bradyrhizobium* sp. are siderophores producing rhizobial species.

Osmoprotectants or Compatible Solutes

Salt tolerant bacteria under saline conditions to defend the osmotic upshift and efflux of K^+ ions, accumulate intra cytoplasmic soluble sugars like maltose, sucrose, trehalose, cellobiose, turanose, gentiobiose, palatinose and solutes like amino acids such as glutamate, proline, alanine, serine, threonine and aspartic acid; quaternary amines such as glycine, betaine and carnitine; K^+; and tetrahydropyrimidines (ectoines). And these solutes are known as compatible solutes. These compatible solutes help in maintaining high turgor pressure, equilibrium across the membranes, reducing cell osmotic potential, stabilizing proteins, ensuring for the correct folding of polypeptides under denaturing conditions at high salt concentration, and alleviating toxicity of NaCl [33]. Bacteria either synthesize these solutes or uptake it from the surrounding environment. Under stress environment it is preferable to uptake solutes rather than synthesizing because synthesis of solutes requires more energy. Proline is significantly produced at 4% NaCl by *Azospirillum brasilense, Bacillus subtilis*, and *Pseudomonas fluorescens*. Osmolyte production by salt-tolerant PGPR under stress conditions increasing their survival chances. Studies conducted by Lopez-Leal et al. [34] suggested that *Rhizobium etli* uses treYZ pathway to synthesize trehalose and sucrose sugars during osmotic stress. Fan et al. [35] detected plant growth promoting related gene cod A in *Arthrobacter strain* TF4 and TF7, which was responsible for synthesis of glycine betaine under saline conditions, resisting the stress in tomato plant. Singh and Jha [36] study showed that under saline conditions, inoculation of wheat plant with *Serratia marcescens* CDP-13 balanced the concentration of proline and soluble sugars in comparison to uninoculated control plants. Maintaining the osmotic balance is the primary defense mechanism for microbes and plants surviving under saline conditions. Therefore, compatible solutes or osmolytes act as first-line immunity in protecting from the toxicity of NaCl.

Polysaccharides:

Bacteria secrete polysaccharides under saline conditions to promote adherence to environmental surfaces and helps in the formation of organo-mineral sheath i.e. biofilm, providing physical and functional protection against desiccating and high salinity situation. Bacterial polysaccharides have been characterized as capsular polysaccharides (CPS), exopolysaccharides (EPS), lipopolysaccharides (LPS), and β-1,2-glycans. Extracellular polysaccharides are an important component in biofilm formation and most promising in reducing salinity stress. Exopolysaccharides producing plant growth promoting rhizobacteria helps in formation of rhizosheath which acts as protective and active site for nutrient recycling, ion balancing, availing water to plants, monitoring cationic intake, maintaining the symbiotic relation between the bacteria and the plant, and help in nodulation [37]. Extracellular polysaccharides prevent the contact between rhizobia and saline environment thus protecting the nodules from oxygen toxicity and therefore maintaining activity of nitrogenase in nodules, increasing phosphate solubilization by legumes from organic and inorganic sources, protecting bacteria against plant antimicrobial compounds and also neutralizing the harmful effects of ROS (reactive oxygen species) produced under salinity stress. Immobilization of Na^+ ions is also facilitated by EPS to reduce salt toxicity and osmotic unbalance. Ashraf et al. [38] explained alleviation of salinity in plants by exopolysaccharides producing PGPR inoculation rendering reduced Na^+ uptake in roots and restricted transfer to leaves.

Antioxidant System

Partial reduction of oxygen due to increased salinity leads to production of reactive oxygen species (ROS) such as superoxide radical, hydroxyl radical, and hydrogen peroxide. The reason behind the ROS production is over reduction of photosynthetic electrons by reduced photosynthetic activity [39]. ROS increase oxidative damage in plant by slowing down nitrogenase activity, decreasing nodule protein content and leghemoglobin [40]. To combat the detrimental effects of ROS, salt-tolerant PGPR can be used to scavenge the oxidative radicals. PGPR enhance the production of antioxidant enzymes in plants to higher levels in comparison to control plants [41]. Antioxidant enzymes include superoxide dismutase, guaiacol peroxidase, ascorbate peroxidase, catalase, polyphenol oxidase, and glutathione reductase which are generated in secondary metabolic pathways [42]. Ascorbate peroxidase and catalase enzymes are important for ROS detoxification. These enzymes detoxify the effects of hydrogen peroxidase by converting it into H_2O and O_2.

Induced systemic resistance

Infection by microbes such as bacteria, fungi, virus can induce the plant to develop resistance to a future attack called induced systemic resistance. Induced systemic resistance induced by phytopathogens, immunizes plant against broad spectrum pathogens. Induced systemic resistance accompanied by PGPM through the production of the allopathic compound, competition for ecotype and nutrient. Allelochemicals such as siderophores, antibiotics, act effectively against pathogens and inhibit their growth. PGPM induced defence mechanisms first reported in response to pathogen *Fusarium* sp. causes wilt disease in carnation (Dianthuscaryophillus) and cucumber (Cucumissativus) in response to pathogen *Colletotrichum orbiculare* caused foliar disease [43]. Lee et al. [44] has reported that root associated *B. amyloliquefaciens* strain HK34 effectively induced resistance against *P. cactorum.* In addition, *Pseudomonas* and *Bacillus* strains manage plant disease in many crops through induced systemic resistance. *Paenibacillus* P16 showed an effective biological control agent (BCA) in cabbage for black rot (*Xanthomonas campestris*) disease and has potential ability of induced systemic resistance. PGPB species such as *Bacillus* strains induced systemic resistance in rice against bacterial leaf blight caused by *Xanthomonas oryza* epv. *Oryzae*.

Systemic acquired resistance (SAR)

The SAR develops in plant as fully active defence mechanism in response to primary infection. The host plant can recognize nature of pathogen based on molecular pattern and detoxify its effects by changing gene expression, production of hormones and metabolites [45]. According to Banerjee et al. [46]*Arthrobacter* sp. and *Bacillus* sp. isolated from the rhizosphere of tomato showed plant growth promoting potential such as phosphate solubilization, IAA production and biocontrol properties. Some bacterial species acts against fungicides produced by fungi. For instance, *P. aeruginosa* strain PS1, an efficient PGPB applied in soil against fungicides to ameliorate their effects. Siderophores, phytohormones, hydrogen cyanide and ammonia were produced under stress condition.

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