Alleviating Salt Stress in Crop Plants through Salt Tolerant Microbes

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Abstract: ***Soil salinity is regarded as the major hurdle for the cultivation of crops especially in the arid and semi-arid regions of the world causing drastic reduction in the yield of crops plants, threatening the global food production potentials. Inoculating the crop plants with the efficient salt tolerant plant growth promoting rhizobacteria will certainly helps the plant to overcome the adverse effect of salt stress and make the plants to survive better under salt stress condition and to produce healthy yield.***

Keywords: Salinity, salt stress, yield and arid and semi-arid

1. Introduction

Soils containing considerable amount of soluble salts such as sodium, potassium, magnesium, calcium, chloride, sulphate, carbonate and bicarbonate are called saline soils. Primary salinisation is caused by accumulation of salt through natural process such as underground water and salt content in parent material. Whereas, secondary salinisation is caused by various human interventions such as indiscriminate use of chemical fertilizers and saline water for irrigation coupled with inefficient drainage. The United States salinity laboratory (1954) defined saline soil as soil whose electrical conductivity of the saturation extract (EC) is more than 4 dS/m at 25°C and exchangeable sodium percentage (ESP) is less than 15. The pH of such saline soil is usually below 8.5.

Soil salinity is widely emphasized as the main agricultural problem owing to its ill effect on the growth and development of the plant, especially in the irrigated agriculture. Approximately 20 per cent of cultivable lands in the world are under salt stress. According to UNESCO and FAO, the area of saline soils in the world is of 9.5 x 107 km^2 dimention. In India, out of total geographical area of 329 M ha. nearly 8 M ha. is affected by soil salinity. At present, of the 1.5 billion ha. of total cultivated land around the world, an about 77 million ha. (5 per cent) is affected by higher salt content (1).

Uttar Pradesh alone accounts for 1.37 million hectares of sodic and saline soils, followed by Rajasthan has 3.75 lakh hectares, Haryana 2.32 lakh hectares and Punjab 1.5 lakh hectares of land severely affected by salt accumulation. The coastal, deltaic plains and mud flats/mangrove swamps of Orissa and Kerala state are reported to be saline. Theses salt affected soils posing serious threat for the cultivation of crops (2).

Soil salinity particularly more severe in an arid and semi-arid region of India, because of insufficient rainfall to leach down the salt to deeper soil layers. High rate of evaporation leads to accumulation of more amount of salt in the root zone and the upper soil horizon. Salt development in command area is mainly attributed to seepage, excessive irrigation with the poor quality water or saline water in particular for irrigation coupled with inadequate drainage (2) and (3) and excessive use of chemical fertilizers render the soil more unsuitable for the cultivation of crops due to the unsuitable soil chemical reaction, runoff from the cultivated fields has added to the severity of salinity, alkalinity and water logging problems in the command areas which is common to witness in the downstream river basins.

2. Effect of Salt stress on plant growth

Plant growth and development and final yield performance of a crop is decided by several environmental factors such as prevailing temperature, moisture, nutrient content, accumulation of toxic ions, salinity, alkalinity and water logging. Soil salinity is the major abiotic stresses to the growth and development of crop plants affecting an area with the dimension of 950 X 10^6 ha of land worldwide (4). There is a limit or threshold level up to which the crops can tolerate salt stress, beyond which the yield decreases with the increase in the salt content of the soil (5) and (6). The presence of higher level of soluble salt in the soil affects the various stages of plant growth and development and creates yield differences by altering the nutrient status and maturity of the plant (7). All the plants exhibit differential ability towards salt stress tolerance.

Presence of salts in the soil hinders the plant growth (Fig 1) mainly by two reasons. First, by osmotic or water-deficit effect of salinity i.e. reduction of water uptake by the plant. Second, by ion-excess effect of salinity excessive accumulation of salts in the plants system causes injury to the plant cells (8).

Salinity is also similar extreme environment as that of drought bears adverse affect on crop growth and development by hindering the germination of seeds, growth of seedling, flowering, fruiting *etc.* NaCl, a salt which is most abundantly available in the soil which competes with the various nutrients at various levels of crop growth and development resulting in nutrient deficiency and ion toxicity by specific elements (9).

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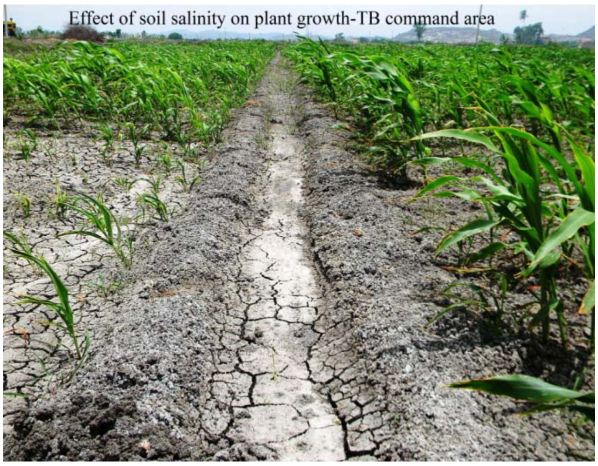

Figure 1: Effect of soil salinity on plant growth-TB command area (Karnataka)

It has long been recognized that a crop's sensitivity to salinity varies from crop to crop and one developmental stage to another stage (10). Most annual crops are tolerant during the germination stage but are sensitive during emergence and early vegetative development. As plants mature, they become progressively more tolerant to salinity, particularly at later stages of development (11) and (12).

Most of the plants are tolerant during germination but the presence of salt stress considerably delays the germination process and percentage of germination in different treatments (13). Presence of higher salt concentrations in the soil eventually reduces the percentage of seed germination in cotton, tomato (14), (15) and (16). The rate of germination and germination percentage of chickpea were significantly reduced with the increase in the soil salinity (6).

Salinity affects both vegetative and reproductive developmental stages of the plant. As compared to germination plants are more susceptible to salinity during the seedling and early vegetative growth stage. Crop tolerance during this sensitive growth-stage differs considerably among crop plants. Sorghum (5), Wheat (13) and cowpea (17) crops are most sensitive during vegetative and early reproductive stages, less sensitive during flowering and least sensitive during the seed filling stage. Salinity oftenly reduces shoot growth more than root growth (11) and can reduce the number of florets per ear, increase sterility and affect the time of flowering and maturity in both wheat (13) and rice (18).

The yield of peanuts growth in artificially salinised plots was reduced to 20 per cent at EC of 3.8 dS m^{-1} and by 50 per cent at EC of 4.7 dS m^{-1} (19). The fresh pod yield of okra was reduced by 90, 75 and 50 per cent, respectively at ECiw (irrigation water) 6.7, 3.9 and 2.1 dS m^{-1} (20). Bresler *et al.*, (21) reported 50 per cent reduction in yield of rice at EC of 7.0 dS m^{-1}. Ayers and Westcot (22) reported 50 per cent yield reduction in wheat at ECe of 13.0 dS m^{-1}. Shalhevet and Yaron (23) reported 10 per cent yield reduction in tomatoes for every 1.5 dS M^{-1} increase in EC.

Mehta and Desai (24) found delayed germination and decreased emergence percentage under increased salinity created by either NaCl or $CaCl_2$ and gave order of tolerance for salt as tobacco > bajra > cotton > bean > tomato > pea.

Bhumbla *et al.* (25) noticed 50 per cent reduction in germination of sesbania, cotton and rice at EC of 4, 8 and 12 mM cm^{-1}, respectively.

Lashin and Atanasiu (26) found that growth of the cotton plant was decreased when grown in sand culture with nutrient solution and different levels of NaCl and $CaCl_2$. Growth further decreased with the increase in the salinity concentration at four and eight weeks after imposing salinity treatments and held that early vegetative growth stage is more sensitive to salinity.

Janardhan *et al.* (27) found that germination per cent on 30^{th} day of three cotton cultivars Varalakshmi and Bhagya and Hampi at salinity level of 12 m mhos cm^{-1} was 40, 25 and 15 per cent and further stated that these difference further varies if they grown under saline water irrigation on saline soil condition.

Ashraf *et al.* (28) investigated the salt tolerance of two cultivars of mungbean, AuMg 588 and Mg 6601 at germination and at the seedling stage. There was a significant reduction in the germination percentage, fresh and dry weights, protein and carbohydrate contents of all plant parts, leaf area, shoot and root lengths, shoot/ root ratio, chlorophyll a, and b and total chlorophyll in both the cultivars. At high salinities, cv AuMg 588 had greater fresh and dry weights of all plant parts than cv Mg 6601. They finally reported that mungbean as a very salt sensitive crop.

Nautiyal *et al.* (29) opined that germination, growth of the seedlings, root length and hypocotyls length of groundnut cultivar was suppressed at different salt concentration and held that sodium carbonate was found to be more toxic for germination than sodium sulphate and magnesium sulphate. Hampson and Simpson (30) reported reduction in germination, root and shoot lengths and growth in wheat. Seedlings when grown with magnesium salt treatments produced shorter shoots and roots than seedlings in other Na and K salts and osmotic stress resulted in reduced hypocotyl elongation. Hypocotyl elongation was noticed with the increase in the temperature.

Subbarao *et al.* (31) observed that germination per cent of salt tolerant wild pigeon pea genotype was 80 to 100 per cent at 6 dS m^{-1} EC, while 70 per cent of mortality was observed in the susceptible genotype.

Zidan and Elewa (32) observed that germination was unaffected at 160 mM Nacl, but at higher salinity level (280 mM) germination was completely inhibited of anise and coriander and remain unaffected in cumin and caraway with 35 and 60 per cent germination respectively, root length and shoot length gradually decreased with the increase in the salt concentration, above 80 mM. Growth of cumin and caraway was stimulated at the lower salinity level (40-80 mM NaCl).

The growth of sunflower seedling in terms of root and hypocotyl length was high at the salinity concentration of 5 m mhos cm^{-1} but greatly reduced at 10 m mhos cm^{-1} and

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inhibited at higher salinity concentration of 15, 20 and 25 m mhos cm^{-1} (33).
Ghoulam *et al.* (34) observed reduction of leaf area, root and shoot length, germination, fresh weight and dry weight and more of solute leakage in case of sugar beet at higher NaCl concentration.

Saghir Ahmed *et al.* (35) attributed the reduction in cotton yield to soil salinity. A threshold salinity level at which initial cotton decline was 7.7 dS m^{-1} with a 50 per cent reduction in yield at 17.0 dS m^{-1}. Anuradha and Seetaram Rao (36) observed 50 per cent reduction in germination, root length; shoot length and seedling dry weight in rice at 150 mM NaCl when compared to the control. Neera Garg and Jasleen (37) reported reduction in root and shoot length, dry weight and gradual accumulation of total soluble sugars in eleven cultivars of chickpea when they were tested for their relative salt tolerance at different level of salt concentration 2, 4, 6 and 8 dS m^{-1}.

3. Causes for the reduction in plant growth under soil salinity stress

Soil salinity affects various physiological and biochemical process within the plant which ultimately results in the reduced plant growth and development. Salt stress, affects the plant water relation at cellular and whole plant level causing specific as well as unspecific reactions and damages. Photosynthetic capacity of different species is reduced due to salinity (38), reduction in leaf growth, root growth, and decrease stomatal conductance and thereby photosynthesis (39). Salt stress causes reduction in plant growth because plant may suffer four types of stresses (8).

Soil salinity bears adverse on physiological and biochemical process and ultimately reduces the growth and development of the plant. It affects plant water uptake mechanism and nutrient status of the plant and induces the series of specific and unspecific ion effect. The presence of salt stress in the root regime of the plant drastically hinders the photosynthetic ability of the plant.

Plant experiences different types of stresses mainly, Osmotically induced water stress, Specific ion toxicity due to higher concentration of Na^+ and $C1^-$, Nutrient ion imbalance, due to high level of Na^+ and $C1^-$ which reduce the uptake of K^+ , NO_3^-, PO_4^{2-} *etc and* Increased production of reactive oxygen species which damage the macromolecules. Salt stress reduces ability of the plants to take up water, and this leads to reduction in growth. This is the osmotic or water-deficit effect of salt stress. So, the salt taken up by the plant does not directly inhibit the growth of new leaves (40).

Salinity stress in plants can up-regulate the production of reactive oxygen species (ROS) such as H_2O_2, O_2^- , $1O_2$ and OH^- resulting in oxidative damage to cells. Excess ROS causes phytotoxic reactions such as lipid peroxidation, protein degradation and DNA mutation (41), (42), (43) and (44).

4. Role of Salt tolerant bacteria in mitigating salt stress

Microorganisms that are able to grow in the absence as well as in the presence of salt are regarded as salt tolerant and those that are able to grow above approximately 15 per cent (w/v) NaC1 (2.5 M) are considered extremely halotolerant (45) and (46).

4.1 Osmoadaptive mechanisms in salt tolerant bacteria

Microbes respond to increase in osmolarity by accumulating osmotica in their cytosol, which protects them from cytoplasmic dehydration and salt stress (47). Majority of bacterium accumulates sugars (e.g. trehalose), polyols (e.g. proline, betaine and ectoine. *Rhizobium* spp. accumulate K^+ when exposed to osmotic shock (48). *Pseudomonas pseudoalcaligenes*, *de-novo* synthesize certain amino acids for salt tolerance (Paul *et al.*, 2005). Salt tolerant *Rhizobium* spp. was reported to accumulate glutamate when cells were grown at elevated salt stress (49).

4.2 Characterization and identification of salt tolerant bacteria

Many studies have been carried out on the isolation and characterization of P-solubilizing, nitrogen fixing, HCN producing, indole acetic acid producing and ACC deaminase producing bacteria from salt affected areas. Novel P-solubilizing salt tolerant bacteria have been isolated from saline soils of China, that could grow at 20 per cent NaCl (50). Phosphate solubilizing salt tolerant bacterium, identified as *Burkholderia vietnamiensis* by 16S rRNA gene sequence has been isolated from South Korea (51). P-solubilizing salt tolerant bacteria have also been isolated from Khewra salt range of Pakistan, could also produce the stress hormone abscisic acid (52). Previously, P-solubilizing bacteria isolated from tomato rhizosphere were reported to produce IAA (53).

4.3 Mechanism of salt stress alleviation by salt tolerant bacteria

A variety of mechanisms have been identified behind microbial elicited salt stress tolerance in plants. Egamberdieva and Kucharova, (54) reported that, the production of indole acetic acid, gibberellins and many unknown determinants by plant growth promoting rhizobacteria result in increased root length, root surface area and number of root tips. Nitrogenase activity in *Azospirillum* and *Klebsiella pneumoniae* was affected by salt stress (55) and (56). *Azospirillum brasilense* accumulated glycine betaine to restore growth and acetylene reduction activity under salt stress (57). Production of phytohormone indole acetic acid was also affected at above 100 mM NaCl concentration (58).

Inoculation of the salt tolerant bacterial strains belonging to genera *Brevibacterium epidermidis, Micrococcus yunnanensis* and *Bacillus aryabhattai* to mitigate salt stress of (150 mM NaCl) in canola plants produced more than 40 per cent increase in root length and dry weight when compared to uninoculated salt stressed canola seedlings

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(59). *Azospirillum brasilense* significantly ameliorated the adverse effect of salinity on growth and yield of barley (60). There was increase in photosynthetic pigments content, photosynthetic capacity and accumulation of K, P, Mg, Ca and Fe in the shoots and roots.

The term 'induced systemic tolerance' (IST) has been proposed for PGPR induced physical and chemical changes in plants that result in enhanced tolerance to abiotic stresses (61). Some of these kinds of bacteria include *Pseudomonas*, *Azospirillum*, *Azotobacter*, *Klebsiella*, *Enterobacter*, *Alcaligens*, *Arthobacter*, *Burkholderia*, *Bacillus* and *Serratia* have reported to enhance plant growth (62). Certain bacteria help the plants to overcome abiotic stresses. Yildirim *et al* (63) found that salt stress was mitigated by plant growth promoting rhizobacteria like *Staphylococcus kloosii* and *Kocuria erythromyxa* in *Raphanus sativus.*

5. Conclusion

Salt stress is believed to be the prime reason for the decreased yield of crops plants in the irrigated arid and semi- arid regions of the world, threatening the global food production and food security potentials of the world. Soils contains versatile group of microorganisms, even the soil may be problematic would inhabit a diverse group of microbes adoptable to that particular climate. Salt affected soils, thus inhabit salt tolerant bacteria depicting wide range of diversity among themselves. Identification, isolation and utilization ofsuch efficient salt tolerant plant growth promoting bacteria as seed inoculants would nourish the crop plants under the salt stress and helps to reap the good harvest in the sustainable way.

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