

Remediation of saline soils using halo-tolerant plant growth promoting rhizobacteria

ABSTRACT:

Soil salinization poses a significant threat to global agriculture, affecting approximately 6.73m Ha land area in India. Salinity stress impacts plant growth and soil health negatively, leading to reduced crop yields and soil degradation. This review examines the sources and effects of soil salinity, highlighting the intricate interplay between salinity and soil nutrients and its remediation. Traditional methods for soil remediation often have detrimental long-term effects, prompting the exploration of alternative strategies such as the use of halo-tolerant plant growth-promoting rhizobacteria. HT-PGPR offer a promising solution for sustainable agriculture by enhancing soil fertility and plant resilience to salinity stress through various mechanisms. Furthermore, this review identifies research gaps in understanding the metabolic pathways and strain selection of HT-PGPR, as well as their interactions with soil microbiota. Future research directions include field-scale experiments to validate the effectiveness and economic viability of HT-PGPR inoculation for large-scale application in saline soils. Overall, leveraging the potential of HT-PGPR represents a critical step towards mitigating the global challenge of soil salinity and ensuring food security in the face of climate change.

Keywords: Soil salinity, plant growth, HT-PGPR, field application and soil nutrients

1.1 INTRODUCTION:

Globally, 424 million hectare (Mha) surface (0-30 cm) and 833 Mha sub soil (30-100 cm) which is about 73% of the land area is salinity affected (FAO, 2021). Agricultural practices rely significantly on the state of land and water use, both of which are progressively influenced by climate change (Hertel *et. al.*, 2014). Saline soil is identified by having an EC_e of about 4 dS/m or more in the root zone with 15% ESP and a pH below 8.5 (Jamil *et. al.*, 2011). Although multiple salt ions contribute to salinity, sodium (Na^+), calcium (Ca^{2+}) magnesium (Mg^{2+}), chloride (Cl^-), sulphate (SO_4^{2-}) and carbonate (CO_3^{2-}) are the most prevalent and display the greatest levels of phytotoxicity (Mahmood *et. al.*, 2024). Soil alkalinity, a variant of soil salinity, arises when the soil is primarily saturated with sodium carbonate, resulting in a higher soil pH. The effects of soil alkalinity can be more severe than those of normal salinized soils due to its impact on nutrient availability and soil microbial activity (Rengasamy *et. al.*, 2022). Salinization causes nutritional disorders in plants by modifying nutrient availability and disrupting processes like competitive absorption, transport and distribution within the plant. The interplay between salinity and primary mineral nutrition is intricate and can negatively impact essential soil nutrients such as NPK (Ehtaiwesh, 2022). Cultivating crops under saline soils is a major problem that farmers face today as salinity in the soil results in yield losses of the crop. To counter the negative effect of salinity, farmers use different physical and chemical techniques

including excess use of fertilizers and the use of chemicals. These methods can have negative effects on plant and soil health. An alternative to this is the use of halo tolerant plant growth promoting rhizobacteria (HT-PGPR) which help the soil to regain its fertility and do not have any ill effects on soil and plants. Inoculation of HT-PGPR can alleviate plant salt stress by triggering various physiological and molecular mechanisms. This includes modifying root systems, activating antioxidant mechanisms, producing siderophores and exopolysaccharides (EPS), regulating phytohormones, synthesizing osmolytes, absorbing minerals and controlling phytopathogens (AbuQamar *et. al.*, 2024). It has been observed that several halotolerant soil bacterial species, including *Arthrobacter*, *Azospirillum*, *Alcaligenes Bacillus*, *Burkholderia*, *Enterobacter*, *Flavobacterium*, *Pseudomonas* and *Rhizobium*, can help crops under salinity stress (Bhadrecha *et. al.*, 2023). Overall, the use of HT-PGPR bioinoculants represents a promising strategy for sustainable agriculture, offering multiple benefits including improved soil health, enhanced water retention and increased productivity in saline environments while minimizing environmental impact.

1.2 SOURCES OF SOIL SALINIZATION:

According to international standards, saline and alkaline soils can be classified into three categories based on specific salinity and alkalinity parameters. These classifications utilize soil reaction pH, electrical conductivity (EC) and either the sodium adsorption ratio (SAR) or the exchangeable sodium percentage (ESP) (Table 1). Salinization is governed by various factors, including environmental conditions, water supply and management systems (irrigation and drainage), and agricultural practices (such as plant cover type, density and rooting characteristics) (Singh *et. al.*, 2022). These factors influence the soil-water balance, thereby affecting the movement and accumulation of salts within the soil. Salinization typically occurs under conditions where soluble salts are present, the water table is high, evaporation rates are high and annual rainfall is low. Soil salinization can result from various natural and human-induced factors such as weathering of rocks and improper agricultural practices etc. (Okur and Örçen, 2020).

Table 1: Classification of salt-affected soils

Classification of salt-affected soil	E_{c_e} (dS m^{-1})	pH	SAR	Soil physical condition
Saline	> 4.0	< 8.5	< 13	Normal
Saline-sodic	> 4.0	< 8.5	> 13	Normal
Sodic	< 4.0	> 8.5	> 13	Poor

Natural processes

The weathering of the parent rocks and soil, atmospheric deposition and groundwater are the sources of ions, including carbonate and sulphate. Minerals undergo weathering as a result of water action and dissolved CO₂ (Vinnarasi *et. al.*, 2021). Salinity has an impact on coastal ecosystems, including salt marshes. There are usually three types of marsh zones in tidal salt marshes: high, middle and low. Near the high marsh zone, the salinity of the soil reaches a maximum. The shortened tidal inundation period, which permits evapotranspiration to concentrate pore water salinity and salt to build, is one of the primary causes of this salinity increase (Visser *et. al.*, 2019). The capillary rise in groundwater also results in soil salinization as in areas with high groundwater tables, capillary action can draw saline water up to the soil surface, where it evaporates and leaves salts behind (Yu *et. al.*, 2021)

Human-induced processes

First and foremost, salinization is a naturally occurring process brought about by the movement and dispersal of water-soluble salts from a salt source location to an area that was initially salt-free (Gupta and Gupta, 2019). Salinization is also a human-induced process, known as secondary salinization, that either causes salt-free soils to become contaminated due to poor water and land management practices or increases the concentration of salt in soils that are already affected by salt.

Salt concentrations in the root zone can result from the use of saline groundwater for irrigation, particularly if internal drainage is impeded and leaching is inadequate (Singh, 2021). Small areas are affected by this salinization process, which happens in arid conditions. A build-up of salt concentration in the soil can occur from the prolonged use of irrigation water, even in cases when the amount of dissolved salt is low, in regions with significant evapotranspiration (Hopmans *et. al.*, 2021). The use of excess fertilizers also results in soil salinization as some fertilizers contain salt ions that can accumulate in the soil due to excess and prolonged use (Mohanavelu *et. al.*, 2021). High crop demands and continuous cropping can deplete soil nutrients and contribute to salinity. Discharge of industrial wastewater can also introduce salts into the soil (Ondrasek and Rengel, 2021).

1.3 EFFECT OF SALINITY ON SOIL PROFILE:

Crops exhibit various responses to salinity stress, which not only affects their productivity but also induces changes in soil physicochemical characteristics and ecological balance. Salinity exerts detrimental effects on agricultural yield by reducing plant water uptake capacity, resulting in soil erosion and diminished economic returns. It represents a substantial challenge in various regions worldwide (Hailu and Mehari, 2021).

1.3a Plant available water:

Soil salinity presents a challenge when an accumulation of soluble salt ions in the root zone reaches levels that harm plant growth. Elevated levels of salt ions in the root zone hinder a plant's capacity to absorb water from the surrounding soil, reducing available water for plants (Stavi *et. al.*, 2021). In saline environments, water retention in the soil is enhanced, but its accessibility for plant uptake is reduced due to osmotic stress. Consequently, this is likely to result in reduced growth and manifestations similar to drought stress, such as wilting or leaf abscission (Kazemi ArPanahi *et. al.*, 2023). Following irrigation, soil salinity is minimal and plant-available water reaches its peak. The water is transpired by plants or evaporates into the atmosphere, the remaining soil water becomes more saline. This is attributed to the retention of salts while water diminishes in volume due to transpiration and evaporation. This phenomenon is particularly significant during high temperatures and the peak of the growing season which increases the evapotranspiration rate (Abdalla *et. al.*, 2022).

1.3b Flocculation under salinity:

Increased salinity in soil can induce flocculation, a process where fine soil particles clump together to form aggregates. Higher salt levels in the soil solution encourage the aggregation of clay particles which increases the flocculation rate in the soil. On the other hand, the spaces between soil aggregates are comparatively larger than in non-flocculated soil, enhancing soil permeability and reducing the likelihood of waterlogging upon wetting (Le Bissonnais, 2023; Chi *et. al.*, 2024).

1.3c Infiltration rates:

The ratio of salinity to sodicity is indicated by the SAR value which is crucial in determining the impact of salts and sodium on the soil while infiltration rates determine the effect of saline and sodicity on the soil physical health (Singh *et. al.*, 2023). The overall salinity level of the soil solution impacts certain physicochemical properties of the soil. Conversely, the presence of Na⁺ in the soil solution and its absorption into the soil exchange complex can induce distinct effects on soil physicochemical traits (Wang *et. al.*, 2023). Salt ions *viz.* Na⁺ are dissolved in the soil water solution and are thus transported downward along with soil water, primarily through drainage within the soil profile. However, sodium being attached to soil particles, remains immobile and does not migrate downward with soil water (Sharma, 2021). Elevated sodium concentration induces soil dispersion, while higher salinity levels promote soil flocculation. Rainfall can enhance the possibility of sodium-induced dispersion and decrease the possibility of soil aggregations caused by salts.

The physicochemical attributes of salt-affected soils exhibit seasonal fluctuations. These variations manifest in the range of salinity from 0.76 to 1.24 dSm⁻¹, with salinity levels typically escalating in the surface layers during summer due to elevated evaporation rates and decreasing during the monsoon period owing to leaching processes (Mali *et. al.*, 2012). Salt-affected soils exhibit increased salinity and dominance of Na⁺ and Cl⁻ ions, leading to higher SAR values in the summer compared to other seasons. Consequently, optimal conditions for seed sowing or cultivation in such soils are likely during periods of minimal salinity, notably in the monsoon season (Hiranmai and Neeraj, 2024).

1.4 EFFECT OF SALINIZATION ON PLANTS:

Salinity has a major impact on nearly every facet of agricultural development, including decreased yield potential and productivity and the onset of soil contamination and degradation. Around the world, salinized soils are known for causing salinity stress, which inhibits the growth of salt-intolerant plants (Raza *et. al.*, 2023). The negative impacts of salinity typically occur in two stages. Initially, the altered osmotic pressure due to excess salt ions in the root zone disrupts regular water uptake, known as the physiological drought conditions. This disruption impairs cell division and differentiation, resulting in stunted growth and development. Furthermore, excessive salt ion accumulation in the cell walls offers osmotic stress and ionic toxicity to plants and sometimes can also result in cell death (Lu and Fricke, 2023) (Figure 1). Salinity results in membrane injury of the cell and lowers the total carotenoid and chlorophyll content thus affecting photosynthesis in the plants (Hnilickova *et. al.*, 2021). Certain plants modify their photosynthetic pathway in response to salinity stress. For instance, *Mesembryanthemum crystallinum* and *Atriplex lentiformis* respond to water scarcity in saline conditions by switching from the C₃ to CAM and C₃ to C₄ pathways respectively (Wungrampha *et. al.*, 2020). Increased soil salinity offers ionic toxicity to plants which results in the formation of ROS in the plant cells (Hasanuzzaman and Fujita, 2022). These ROS alter membrane permeability and protein structures results in resulting in disruption of plant growth and metabolism (Sachdev *et. al.*, 2021). Salinity results in deteriorative plant growth as under such conditions, plant cells become more permeable to Na⁺ as compared to K⁺ which changes membrane potential and also causes K⁺ deficiency in the plants. The absorption of K⁺, NO₃⁻, PO₄²⁻, Ca²⁺ and Mg²⁺ reduce under salinity and as a result, the ratios of K⁺/Na⁺, Ca²⁺/Na⁺, Mg²⁺/Na⁺ and PO₄²⁻/Cl⁻ also decreases which causes various macro and micronutrient deficiencies in the plants. High sodium and chloride concentrations can also cause ion imbalance and toxicity to specific ions (Mahmood *et. al.*, 2024).

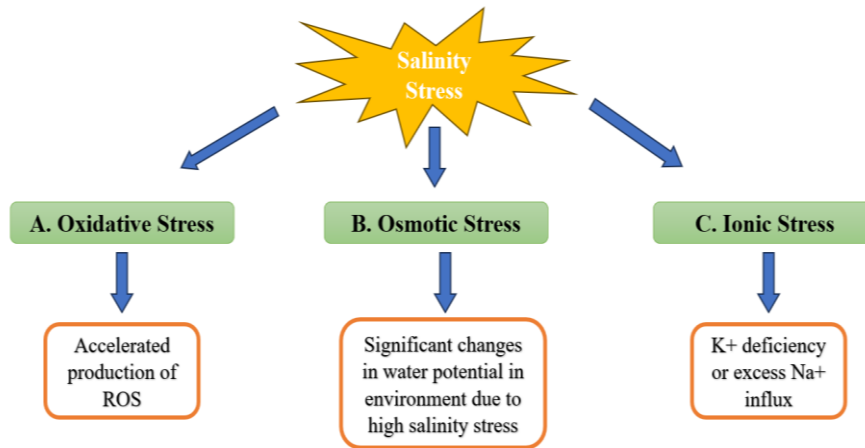


Figure 1: Impact of salinity stress on plants

Excess salinity results in various deteriorative effects on plants which makes plants more prone to various diseases and biotic stresses (Chourasia *et. al.*, 2021). However, some plants become tolerant to salinity by improving growth and survival through various metabolism and adaptations which include maintaining ionic balance, enhanced anti-oxidative enzymatic activity and production of osmolytes to maintain osmotic balance in the plants and sometimes plants try to avoid the adverse effect of salinity by shortening their life cycle.

1.5 ALTERNATIVE AND TRADITIONAL METHODS FOR REMEDIATION OF SALINE SOILS:

Efforts have been made from time to time to address salinization issues and the urgent need for fertile agricultural lands, including soil reclamations and crop modifications. Standard methods for soil reclamation include physical techniques (salt scrapping, flushing and soil washing), chemical amendments (gypsum) and various agricultural management practices (irrigation management, crop rotation and intercropping). But in practice, budgetary constraints and the quality of the water supply constantly place restrictions on such projects (Farooqi *et. al.*, 2023). Furthermore, overuse of pesticides is typically involved by farmers since plants under salinity stress are more susceptible to disease and pests (Shahid *et. al.*, 2023). These short-term fixes will ultimately cause even more soluble salt accumulations from the chemical additives, which will exacerbate the salinity issues (Zhang *et. al.*, 2023). Different molecular approaches have also been discussed for the reclamation of saline soil which include the use of modified crops using genetic engineering, induced mutant breeding, priming agent treatments, conventional selective breeding and other methods to give crops new potential traits like increased salt tolerance. Saline soil reclamation require a sustainable and reliable approach with no long-term ill effect on soil health and improved plant growth under

saline conditions (Singh *et. al.*, 2022). The application of HT-PGPR inoculation is an effective alternate strategy for mitigating salinity hazards and enhancing crop performance. Naturally, the plant tolerance to salt and biotic stresses is increased by these advantageous microorganisms significant growth-promoting and salinity-defensive characteristics. HT-PGPR alleviate salinity hazards through various pathways that activate complex physiological, biochemical and molecular responses in plants to support their growth (Peng *et. al.*, 2024).

1.6 HALOTOLERANT BACTERIA AS SOIL SALINITY REMEDIATORS:

Using halotolerant bacteria for soil remediation offers a sustainable approach to address salinity issues, especially in agricultural areas where saline soils can severely impact crop productivity (Figure 2). Research findings have validated that inoculation with HT-PGPR positively influences nutrient mineral uptake, soil physicochemical properties, organic carbon (OC) content, pH, EC and the accumulation of ionic salts in the soil (AbuQamar *et. al.*, 2024; Arora *et. al.*, 2020). Numerous studies have shown that applying HT-PGPR can improve the salt index of saline soil. Additionally, HT-PGPR addresses nutrient deficiencies by increasing levels of nitrogen (N), phosphorus (P), potassium (K), iron (Fe) and zinc (Zn) in saline soils, thereby restoring vegetation and promoting agricultural sustainability (Channab *et. al.*, 2024).

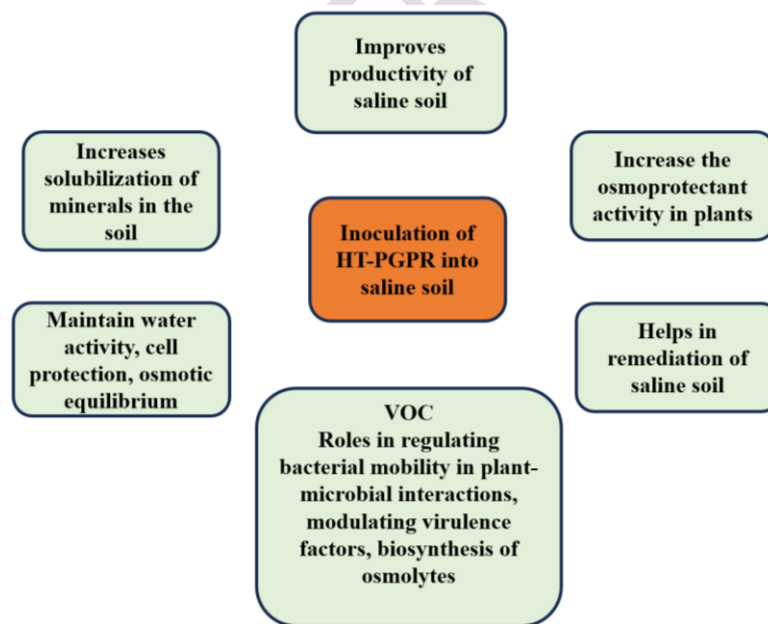


Figure 2: Remediation of salt-affected soil with inoculation of HT-PGPR

Halotolerant bacteria can actively participate in the remediation of salt-affected soil involving enzymatic pathways which are capable of degrading specific salt compounds, converting them into less harmful substances (Kumawat *et. al.*, 2022). Some bacteria can also bind or sequester salt ions, reducing their availability in the soil and mitigating their negative

effects on plant growth known as salt immobilization (Arora, 2021). Halotolerant bacteria may contribute to the improvement of soil structure and fertility in saline soils by producing EPS which act as soil binders, improving soil aggregation and water retention (Agbodjato and Babalola, 2024). HT-PGPR in the soil helps in enhancing organic matter decomposition which contributes to soil nutrient cycling (Peng *et. al.*, 2024). Beyond nutritional aspects, soil quality is greatly influenced by aggregation, which enhances the infiltration rate, root aeration and penetration and the development of micropores (Sharma and Kumar, 2023). HT-PGPR enhance soil physical quality through the synthesis of EPS, particularly under stress conditions (Kumar *et. al.*, 2023). This process promotes the formation of microaggregates of size <250 μm and macroaggregates of size >250 μm , which effectively sequester nutrients and water molecules (Abdullatif *et. al.*, 2024). The establishment of a cohesive EPS-soil matrix offers a protective microenvironment, particularly against salinity stress, thereby safeguarding both plant health and the HT-PGPR population within the rhizosphere. Functioning as a defensive barrier, the EPS within the biofilm complex serves to enhance soil water retention capacity and ameliorate water activity in plants during salinity and drought stress conditions (Santra and Banerjee, 2024). Research has shown that *Pseudomonas putida* strain GAP-P45 EPS improved soil water retention capacity and aggregation under saline conditions (Sandhya, 2015). Biofilm formation within soil particles and on root surfaces is marked by high concentrations of root exudates, signalling molecules, water content and OC content (Guhra *et. al.*, 2022). This complex matrix plays a crucial role in the establishment and selection of different microbial populations. The predominant constituent of EPS regulates organic matter dynamics by serving as a carbon source and facilitating soil particle coagulation, thus promoting the formation of stable organic carbon compounds known as humic substances (Ali *et. al.*, 2024). An increase in salinity diminishes the bacterial populations. The exogenous application of HT-PGPR enhances soil structure and promotes the assimilation of organic matter, thereby fostering microbial interactions. An increase in microbial population and dehydrogenase activity was observed in saline soil after introducing *Bacillus cereus* strain Pb25, which belongs to the category of HT-PGPR (Islam *et. al.*, 2015). This research provides compelling evidence of the role and potential application of HT-PGPR in enhancing soil quality under abiotic stress conditions such as salinity. Table 2 provides an insight into previous research revolving around how inoculation of HT-PGPR in different crops helps mitigate salinity stress through different mechanisms resulting in improved growth and metabolism of the crop plants:

Table 2: HT-PGPR mediated tolerance in different crops under salinity stress

HT-PGPR	Crop Evaluated	Mechanism That Promotes Plant Growth	Effect on the plant	Reference
<i>Gracilbacillus saliphilus</i> , <i>Staphylococcus petrasii</i> , <i>Bacillus licheniformis</i> , <i>Brevibacterium halotolerans</i>	<i>Zea mays</i>	IAA production increased ACC deaminase activity and EPS formation	Increased growth by higher root- to-shoot ratio and increase in fresh and dry biomass	(Aslam <i>et. al.</i> , 2018)
<i>Bacillus safensis</i> , <i>Bacillus haynesii</i>	<i>Amaranthus viridis</i>	GA, IAA, exopolysaccharide s, defence-related enzymatic activities	Growth promotion	(Patel <i>et. al.</i> , 2023)
<i>Hartmannibacter diazotrophicus</i>	<i>Hordeum vulgare</i> L.	Reduced Na ⁺ uptake by the roots, increased phytohormone production	Growth promotion	(Saurez <i>et. al.</i> , 2015)
<i>Pseudomonas pseudoalcaligenes</i> , <i>Bacillus subtilis</i>	<i>Glycine max</i> L.	ACC-deaminase activity, production of siderophores and IAA	Increase in shoot and root dry weight, higher K ⁺ /Na ⁺ in shoot and root and increased proline concentration	(Yasmin <i>et. al.</i> , 2020)
<i>Pantoea ananatis</i>	<i>Oryza sativa</i>	ACC deaminase, P-solubilization, IAA, Siderophore	Higher K ⁺ /Na ⁺ ratio in the roots,	(Lu <i>et. al.</i> , 2021)

			increased chlorophyll content	
<i>Sinorhizobium mellilote</i> and <i>Rhizobium leguminosarum</i> b.v phase <i>oli</i>	<i>Brassica napus</i> L.	IAA, ACC deaminase activity and phosphate solubilizing	Improved growth, higher nutrient uptake and lower Na ⁺ /K ⁺ ratio in the roots	(Saghafi <i>et. al.</i> , 2018)
<i>Bacillus</i> sp. (EN1), <i>Zhihengliuella halotolerans</i> (EN3), <i>Bacillus</i> sp. (EN5), <i>Bacillus gibsonii</i> (EN6 and EN10), <i>Oceanobacillus oncorhynchi</i> (EN8), <i>Zhihengliuella</i> sp. (EN12), and <i>Halomonas</i> sp. (IA)	<i>Triticum aestivum</i> L.	Ammonia, IAA and ACC deaminase production, phosphate solubilizing and N ₂ fixation	Increased the root and shoot length and total biomass of the plants	(Orhan, 2016)
<i>Bacillus</i> sp. and <i>Pseudomonas</i> sp.	<i>Musa acuminata</i>	higher levels of K ⁺ , Ca ²⁺ , and Mg ²⁺ and lower levels of Cl ⁻ and Na ⁺ , defence-related enzyme activities	Improved growth parameters and development	(Kaleh <i>et. al.</i> , 2023)
<i>Bacillus licheniformis</i> NJ04 strain	<i>Solanum lycopersicum</i> L.	Lower Na ⁺ /K ⁺ ratio in the roots	Increased root and shoot length of the plant	(James <i>et. al.</i> , 2023)

1.7 RESEARCH GAPS:

Rapid salinization of agricultural land adversely affected our crop yields and posed a serious threat to food security. Global climate change results in the conversion of fertile agricultural land to saline soils every year. Different physical, and chemical amendments and agricultural techniques have been used by farmers from time to time for the reclamation of

saline soils but these techniques negatively affect the soil health and also yield potential of the crops. HT-PGPR has proven to be an effective and sustainable solution from time to time for the reclamation of saline soils for crop production (Arora *et. al.*, 2020). Several PGPR metabolites and genes that respond to salinity stress have been found as a result of approach and method advancements. Still, more research is needed to understand the HT-PGPR's metabolome in both physiologically stressed and unstressed plant interactions (Al-Turki *et. al.*, 2023). Furthermore, optimal strain selection is important for the proper reclamation of saline soils and investigation is required for the identification of the most effective HT-PGPR for various saline soil types and conditions (Teo *et. al.*, 2022). A better understanding of the mechanism of tolerance for mitigating stress mediated by halotolerant bacteria and interacting with plants and soil in saline environments to promote growth (Hernández-Canseco *et. al.*, 2022). Investigating the interactions between introduced halotolerant bacteria and the indigenous soil microbiome is crucial to understand their impact on soil ecology and microbial diversity (Ma *et. al.*, 2024). Research should focus on assessing the long-term effects of halotolerant bacterial inoculation on soil salinity levels, soil health, and crop productivity to determine the sustainability of this approach (Phour and Sindhu, 2023). More field-scale experiments are necessary to validate the effectiveness of halotolerant bacterial inoculation under field conditions and to evaluate its practical feasibility and economic viability (Malik and Garg, 2024). Developing scalable and cost-effective strategies for the large-scale application of halotolerant plant growth-promoting bacteria in saline soils is essential for practical implementation and widespread adoption. These research gaps will contribute to the development of more effective and sustainable strategies for remediating saline soils using halotolerant plant growth-promoting bacteria.

1.8 CONCLUSION:

Changing climate increases the threats of abiotic stresses such as soil salinity which negatively affects the agriculture system and food security. Salinity negatively affects the plant and soil health posing ionic and osmotic stress to the plants and thereby reducing yields of the crops. Various unsustainable traditional methods have been used by farmers to increase yield under saline soils. Their unsustainable approaches had long-term effects on soil microorganisms and their health. Inoculation of HTPGPR can be a sustainable and reliable approach for the reclamation of saline soils as it improves soil and plant health. Furthermore, areas such as the interaction of HT-PGPR with other soil microbes and the mechanism of salt tolerance to salinity and be explored and more research on this innovative microbial technology is required to control the pandemic of soil salinity and increase crop yield which will help better understanding of HT-PGPR mediated salinity tolerance and its effect on soil health.

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