Int.J.Curr.Microbiol.App.Sci (2019) *8*(4): 2847-2858

International Journal of Current Microbiology and Applied Sciences
***ISSN: 2319-7706* Volume 8 Number 04 (2019)**
Journal homepage: http://www.ijcmas.com

Original Research Article https://doi.org/10.20546/ijcmas.2019.804.332

Plant Growth Promoting *Bacillus* spp. and *Paenibacillus alvei* on the Growth of *Sesuvium portulacastrum* for Phytoremediation of Salt Affected Soils

P. Kalaiselvi*, R. Jayashree and R. Poornima

Department of Environmental Sciences, Tamil Nadu Agricultural University, Coimbatore-641003, India

**Corresponding author*

Keywords

Textile effluent, Salt, Remediation, Rhizobacteria, PGPR

Article Info

Accepted:
20 March 2019
Available Online:
10 April 2019

A B S T R A C T

The aim of the study was to isolate and identify the salt tolerant growth promoting bacteria from rhizosphere soil of *Sesuvium portulacastrum* and also from the soils of dye and textile effluent contaminated sites (Andipalayam, Orathupalayam, Mangalam and Palayakottai villages of Tirupur District, Tamil Nadu) to remediate the salt contaminated soil. On total twenty five strains were selected based on the distinct morphological characters on R2A agar medium supplemented with 3 % NaCl. These strains were further screened for salt tolerance potential and growth with various concentrations of NaCl (0.5%, 1%, 2% and 3%). Only 4 strains (OPS2, OPS4, APS1 and APS3) showed the highest salt tolerance potential. The bacterial strain OPS2 has shown the highest removal of salt from the medium. The phylogenetic analysis revealed that 3 strains belonged to *Bacillus* sp. and a single strain was within *Paenibacillus* sp. Further these four strains were characterised for plant growth promotion activities. A pot culture experiment was conducted to assess the role of bioamendments and bioinoculants in enhancing salt removal capacity of *S. portulacastrum*. The maximum EC reduction (72.27%) and sodium removal (80.29%) was observed in the treatment Soil+ *Sesuvium portulacastrum* applied with Vermicompost (5tha^{-1}) and Salt tolerant growth promoting rhizobacteria (ST-PGPR).

Introduction

Textile industries, being a diverse sector, hold almost 14% of the total industrial production in India. Nearly, 10,000 garment manufacturers and 2100 bleaching and dyeing industries are present in India. An Indian textile industry contributes 80% of the country's total textiles and operates largely in clusters mainly intensified in states of Tamil Nadu (Tirupur and Karur), Punjab and Gujarat. With its great demand for water (80–100 m^3/ton of finished textile), safe disposal of the wastewater (115–175 kg of COD/ton of finished textile, a large range of organic chemicals, low biodegradability, colour, salinity) is yet another challenging issue that has to be unravelled due to its complex nature. Main pollution in textile wastewater is from dyeing and finishing processes. With high concentrations of salt these effluents accumulate in various trophic levels of

ecosystem resulting in a chaos for the agricultural land and water bodies (Bharti and Chauhan, 2013). Totally 6800 ha of agricultural land is affected in Tirupur district due to dye and textile effluents. Around 8.09 million ha is affected with menace of salinity in different climatic regions (Singh *et al.*, 2013). Textile industry wastewater is characterized by high value of BOD, COD, pH and colour. The pH range from 5.5 to 10.5 and the EC is 3.5 to 9.1 dSm^{-1}. The value of TDS ranges from 1500 to 12000 ppm, the TSS and chloride may go up to 8000 and 6000 ppm. A wide variation of 400 to 7900 ppm was observed in the sodium concentration of the effluent (Hussein, 2013; Rajeswari *et al.*, 2013; Eswaramoorthi *et al.*, 2008).

Salt accumulation in soils poses problems in two ways: the soil becomes less permeable, and the salt damages or kills the plants. Highly saline soils have high EC, ESP, SAR and are generally poor in availability of macronutrients, micronutrients (Pessarakli and Szabolcs, 1999), organic matter (Qadir *et al.*, 1997), mineralization rates and enzyme activities (McClung and Frankenberger, 1985). Despite which, there are soils with indigenous salt content that includes the clayey soils. The salt content of experimental region usually ranges from 4 to 10.2 dS m^{-1}. Increased salinity limits microbial growth and activity by causing osmotic stress, dehydration and lysis of cells (Wichern *et al.*, 2006). Wong *et al.*, (2008) also observed an increase in metabolic quotient (respiration per unit biomass) with increasing salinity and sodicity, indicating a more stressed microbial community. Intensified salinity also poses direct effect on plants such as a reduction in the osmotic potential of the soil solution that reduces plant available water, a deterioration in the physical structure of the soil such that water permeability and soil aeration are diminished and increase in the concentration of certain ions that have an inhibitory effect on plant metabolism (Grattan and Grieve, 1999).

Amelioration of saline soil involves physical technique (water leaching, deep ploughing, subsoiling, sanding, profile inversion), chemical technique (gypsum, calcium chloride, limestone, sulphuric acid, sulphur, iron sulphate), electro-reclamation (treatment with electric current) and the biological methods including phytoremediation using living or dead organic matter and using microorganisms (Feizi *et al.*, 2010). However, selection and adoption of these technologies depends on soil type, depth of soil to be ameliorated, water available for leaching, quality and depth of groundwater, desired rate of replacement of excessive exchangeable Na^{+}, occurrence of gypsum in soil, availability and cost of amendments, topographic features of the land, nature of the crops to be grown or the land use during and after amelioration, climatic conditions and time available for amelioration.

Salt stress upsets plant–microbe interactions, constituting a critical ecological factor that helps sustain and enhance plant growth in degraded ecosystems. To adapt to saline stressed environments, microorganisms have developed various biochemical strategies over time to maintain structural and functional stability of the cells. As a result, many bacteria are able to synthesize secondary metabolites, such as extracellular enzymes and bioactive compounds. Now there is increasing evidence that the use of beneficial microbes in agricultural production systems can enhance plant resistance to adverse environmental stresses drought, salts, nutrient deficiency and heavy metal contamination. Under adverse environmental stresses, it requires suitable biotechnology to improve not only crop productivity but also soil health through interactions of plant roots and soil

microorganisms. Development of such a stress tolerant microbial strain associated with roots of agronomic crops can lead to improved fertility of affected soils. *Sesuvium portulacastrum* is a pioneer plant species used for sand dune fixation, desalination and phytoremediation along coastal regions. The plant tolerates abiotic constraints such as salinity and drought. It is used as vegetables, fodder for domestic animals and as an ornamental plant. It grows at severe salinity of 1000mM NaCl (Lokhande *et al.,* 2013).

Plant growth promoting rhizobacteria (PGPR) – induced plants stress tolerance is considered to be an economic approach to alleviate the salt stress (Barassi *et al.,* 2006). Dodd and Alfocea (2012) reported that isolated PGPR from saline soils improve the plant growth at high salt and it can tolerate wide range of salt stress and enable plants to withstand salinity by hydraulic conductance, osmotic accumulation, sequestering toxic Na^+ ion maintaining the higher osmotic conductance and photosynthetic activities. The bacteria obtained from saline environment include *Flavobacterium, Azospirillum, Alcaligenes, Acinetobacterium, Pseudomonas, Sporosarcina, Planococcus* (Ventosa *et al.,* 1983) *Bacillus* (Upadhyay *et al.,* 2009) *Thalassobacillus, Halomonas, Brevibacterium, Oceanobacillus, Terribacillus, Enterobacter, Halobacillus, Staphylococcus and Virgibacillus.* Hence, this aims to assess the potential of *Sesuvium portulacastrum* and their interactions with rhizosphere in remediating salt affected soils (Fig. 1).

Materials and Methods

Isolation, screening and phylogenetic characterization of bacteria associated with rhizosphere of *Sesuvium portulacastrum*

Rhizosphere soil of *Sesuvium portulacastrum* brought from Pitchavaram village in Chidambaram district of Tamil Nadu was used for isolating salt tolerant bacterial strains using R2A agar medium with 6% sodium chloride (NaCl). These strains were further screened for salt tolerance and growth in R2A broth amended with various concentrations of NaCl (0.5%, 1%, 2% and 3%). The growth was measured at 600nm at 72 h.

The potential four salt tolerant bacteria were further selected for phylogenetic identification. Axenically maintained culture was used for DNA isolation. Colonies are picked up with a sterilized toothpick, and suspended in 0.5 ml of sterilized saline in a 1.5 ml centrifuge tube. Centrifuged at 10,000 rpm for 10 min. After removal of supernatant, the pellet is suspended in 0.5 ml of InstaGene Matrix (Bio-Rad, USA). Incubated 56a for 30 min and then heated 100^0c for 10 min. After heating, supernatant can be used for PCR.

For PCR amplification, 1 µl of template DNA was added to the 20 µl of PCR reaction solution. The primers 518F/800R was used for amplification, 35 amplification cycles were performed using the following programme 94^0c for 45 sec, 55^0c for 60 sec, and 72^0c for 60 sec. The PCR products were purified to remove the unincorporated PCR primers and dNTPs using Montage PCR Clean up kit (Millipore). The purified PCR products of approximately 1,400 bp were sequenced by using the primers (785F 5' GGA TTA GAT ACC CTG GTA 3' and 907R 5' CCG TCA ATT CCT TTR AGT TT 3'). Sequencing were performed by using Big Dye terminator cycle sequencing kit (Applied BioSystems, USA) and the sequenced products were resolved on an Applied Biosystems model 3730XL automated DNA sequencing system (Applied BioSystems, USA).

The culture sequences obtained were subjected to BLAST analysis, the phylogenetically similar type strains sequence

and other phylogenetic related sequence were selected from the GenBank and they were subjected to multiple sequence alignment and then then align sequences were trimmed to similar length in nucleotides and were subjected to phylogenetic tree (neighbour joining) analysis using MEGA 6. In the tree the numbers at the nodes indicate the levels of the bootstrap support [high bootstrap values (close to 100%) meaning uniform support] based on a neighbour-joining analysis of 1,000 re-sampled data sets. The bootstrap values below 50% were not indicated. Bar 0.005 substitutions per site.

Assay for plant growth promoting abilities

Indole 3-acetic acid production

Indole 3 – acetic acid (IAA) production was analysed calorimetrically (Gordon, 1951) and quantified by growing the bacterium for 7 days in LB- broth supplemented with 100mg/L tryptophan as precursor of IAA. For estimation of IAA in the presence of salt, LB – tryptophan was supplemented with different concentrations of NaCl. The grown culture was centrifuged at 10.000rpm. Supernatant was acidified (up to pH 2.8) with hydrochloric acid and extracted twice with equal volume of ethyl acetate (Tien *et al.,* 1979). The extracts were further air dried and analysed using high – performance liquid chromatography at a flow rate of 0.5ml/min on C – 18 column.

Siderophore production

The CAS solution was prepared by dissolving 60.5g of chrome azurol sulphate (CAS) in 50ml distilled water, and to this, 0.27g of FeCl3 was added and stirred well. To this, 364.6µl of concentration HCl was added and mixed well. It was slowly added to CATB solution (2.9g CATB in 40ml distilled water) while stirring, resulting in a dark blue solution (100ml total) and autoclaved at 121°C for 15 minutes. The basal media was prepared by adding 4g of succinic acid, 3g of K_2HPO_4 and 0.2g of ammonium sulphate. To this 50ml of CAS solution was added along the walls of the flask with constant stirring and the pH was adjusted to 7.0. The volume was then made upto 1L and agar was added and autoclaved. After autoclave, it was cooled and poured in sterile petriplates, each plate receiving approximately 25ml of blue agar.

After 24 hours (to check any contamination), all the isolates were spot inoculated on these plates and incubated at optimum growth temperature for 3 – 4 days. The isolates producing orange colour in the form of halo zone around the colonies were considered as siderophore producers.

Phosphate solubilisation

The quantitative estimation of solubilized P by bacterial isolates was done by the vanadomolybdophosphoric yellow colour method (Subba Rao, 1988) in NBRIP (National Botanical Research Institute's Phosphate growth medium) broth (Nautiyal 1999; Mehta and Nautiyal, 2001) containing 1000 µg/ml tri-calcium phosphate (TCP).

Pot culture experiment

Pot culture experiment was conducted to assess the role of bioamendments and bioinoculants in enhancing salt removal capacity of *Sesuvium* with the following combinations *viz.,* Soil + *Sesuvium portulacastrum,* Soil + *Sesuvium portulacastrum* + Vermicompost ($5tha^{-1}$), Soil+ *Sesuvium portulacastrum* + Vermicompost ($5tha^{-1}$) + ST-PGPR. The soil collected from Andipalayam village was used in the pot culture experiment. Observation on salt uptake was analysed in soil and plant samples at 0, 30 and 60 days after planting in pot culture study

Field study

The field experiment to assess the potential of *Sesuvium portulacastrum* on salt removal was established at Andipalayam Village of Tirupur District. *Sesuvium portulacastrum* was planted in the field size of 10ftx10ft. The microbial inoculants was mixed with vermicompost and applied to the field and control without inoculum was maintained to compare the salt removal efficiency. The soil physico-chemical characteristics and plant biometric characteristics were monitored at 0, 30 and 60 days after planting.

Results and Discussion

Salt tolerant bacteria from rhizosphere soil of *Sesuvium portulacastrum* taken from Pitchavaram were isolated and twenty five strains were selected based on the distinct morphological characters on R2A agar medium (3% NaCl) plates. Colonies were selected based on colour, shape, size and abundance. These strains were further screened for salt tolerance and growth in R2A broth amended with various concentrations of NaCl (0.5%, 1%, 2% and 3%). Among these strains, 10 isolates failed to grow during sub culturing and the remaining 15 were screened for further salt tolerance test. The growth was measured at 600nm at 72 h. Among the 15 strains only 4 strains (OPS2, OPS4, APS1 and APS3) shown the highest salt tolerance potential (Table 1). The existence of the isolated and screened bacterial strains (2 from Orathupalayam soil and 2 from Andipalayam soil) at high salt concentration was tested by further sub culturing in the R2A medium amended with NaCl and maintained for further characterization and also to study the plant growth promotion activities. The bacterial strain OPS2 has shown the highest removal of salt from the medium followed by OPS4, APS1 and APS3. The highest removal was observed in the 0.5% concentration. As the concentration increase the removal shown to be less (Table 2).

The morphological and biochemical test of the 4 isolated strains (OPS2, OPS4, APS1 and APS3) is tabulated in Table 3. On the basis of nucleotide sequences of the 16S rDNA fragments the selected strains were identified as *Paenibacillus alvei* (OPS4), *Bacillus aryabhattai* (APS1), *Bacillus vietnamiensis* (APS3) and *Bacillus megaterium* (OPS2). Both *Paenibacillus* and *Bacillus* have been reported to provide tolerance to host plants under different abiotic stress environments (Grover *et al.,* 2011). Upadhyay *et al.,* (2011) isolated *Paenibacillus* from Wheat (*T. aestivum*) which imparted some degree of tolerance towards salinity stress. *Bacillus* is one of the most dominant bacteria obtained from saline environment (Upadhyay *et al.,* 2009 and *Rodriguez-Valera*, 1988). Several halophilic *Bacillus* species have been isolated from soil samples and it exhibited halophilic properties. Furthermore, Siddikee *et al.,* (2010) reported that *Bacillus aryabhattai* is able to ameliorate salt stress of (150mM) in canola plants thereby producing more than 40 per cent increase in root length and dry weight compared to the control.

Moreover, the results of the plant growth promoting potential of the strains indicated that OPS2 (*Bacillus megaterium*) showed a positive result for all the three tests (IAA production, Siderophore production and Phosphate solubilisation) whereas the other three strains produced IAA only (Table 4). IAA produced by a halo tolerant bacterium will modulate the plant stress level through promoting root growth by stimulating plant cell elongation or cell division (Patten and Glick, 2002, Siddikee *et al.,* 2010). Production of siderophores, an elicitor of induced systemic resistance, is one of the direct stimulation on plant growth and

development by providing iron that has been sequestered by bacterial siderophores. Solubilisation of phosphorus in rhizosphere increases the nutrient availability to the host plant (Rashid *et al.,* 2004). These rhizobacteria are critical for the transfer of P from poorly available forms and are important for maintaining P in readily available pools.

Field study

The field experiment to assess the potential of *Sesuvium portulacastrum* on salt removal was established at Andipalayam Village of Tirupur District. *Sesuvium portulacastrum* was planted in the field size of 10 x10ft.

The microbial inoculants was mixed with vermicompost and applied to the field and control without inoculum was maintained to compare the salt removal efficiency. The soil physico-chemical characteristics and plant biometric characteristics were monitored at 0, 30 and 60 days after planting.

Significant growth of plants has been achieved over the experimental period due to the inoculation of microbial consortia. The plants in field study showed higher root (41.4 cm) and shoot length (42 cm) after 60 days of planting than the pot cultured plants whose root and shoot length are 37.8 cm and 29.7 cm respectively (Table 5).

Irrespective of pot and field study the highest biomass content of *Sesuvium* plant was recorded in the microbial cultures inoculated experiment compared to control at all days of growth.

The maximum biomass of 262 and 475 g $pot^{-1}$was recorded in the pot experiment and in the field study respectively at 60 DAP (Table 6). From the table 7, it is evident that the EC and sodium content of the soil tends to decrease over the experimental period. An higher proportional reduction has been attained in field study than in pot culture study, wherein an initial EC of 13.5 dSm^{-1} has reduced to 5.5 (30 DAP) and 3.2 (60 DAP) while the initial concentration of Sodium (3500 mg kg^{-1}) has been reduced to 1750 mg kg^{-1} (30 DAP) and 700 mg kg^{-1} (60 DAP).

Assessing the role of bioamendments and bioinoculants in enhancing salt removal capacity of *Sesuvium*- Pot culture experiment

Pot culture experiment was conducted to assess the role of bioamendments and bioinoculants in enhancing salt removal capacity of *Sesuvium* with the following combinations *viz.,* Soil + *Sesuvium portulacastrum,* Soil + *Sesuvium portulacastrum* + Vermicompost ($5tha^{-1}$), Soil+ *Sesuvium portulacastrum* + Vermicompost ($5tha^{-1}$) + ST-PGPR.

The soil collected from Andipalayam village was used in the pot culture experiment. Observation on salt uptake was analysed in soil and plant samples at 0, 30 and 60 days after planting in pot culture study.

The initial EC of the Andipalayam village soil ranges between 10.1 to 10.5 dS m^{-1}. Among the treatments, the maximum EC reduction was observed in the treatment Soil+ *Sesuvium portulacastrum* applied with Vermicompost ($5tha^{-1}$) and Salt tolerant growth promoting rhizobacteria (ST-PGPR) recorded the EC of 3.8 dS m^{-1} (30 DAP) and 2.8 dS m^{-1} (60DAP).

The initial sodium content Andipalayam village soil ranges from 2980 to 3200 mg kg^{-1} is reduced to 1420 $mgkg^{-1}$ (30 DAP) and 610 $mgkg^{-1}$ (60DAP) in the treatment Soil+ *Sesuvium portulacastrum* applied with Vermicompost ($5tha^{-1}$) and ST-PGPR (Table 8).

Table.1 Bacterial population assessed at 72 h as CFU ml^{-1}

Strains used	NaCl concentration				
	Control	0.5%	1%	2%	3%
OPS2	2.0x 10-3	6 x 10-7	4 x 10-7	7 x 10-6	8 x 10-6
OPS4	3.0x10-3	2 x 10-6	2 x 10-6	5 x 10-4	4 x 10-4
APS1	1.0x10-2	7 x 10-7	8 x 10-7	5 x 10-6	3 x 10-6
APS3	2.0x10-3	7 x 10-6	8 x 10-6	5 x 10-5	3 x 10-5

Table.2 Sodium content in the filtrate in mg L^{-1}

Strains used	NaCl concentration			
	0.5%	1%	2%	3%
OPS2	3570	5290	10100	15300
OPS4	2980	5090	9770	13700
APS1	2530	5350	9660	14400
APS3	3070	5380	7220	10800

Table.3 Morphological, biochemical identification and phylogeny results of the isolated bacterial strains

Sl. No.	Morphology and biochemical test	OPS2	OPS4	APS1	APS3
1.	Morphology	Small oval colonies	Round shiny colonies	Oval rough colonies	Small oval colonies with serrated margins
2.	Gram staining	Gram positive	Gram negative	Gram positive	Gram positive
3.	Spore shape	-	Oval and swollen sporangium	Ellipsoidal central	-
4.	Colony colour	Dull white to creamy	Yellow	Yellow	Red to pink coloured
5.	Catalase	+	+	+	+
6.	Oxidase	+	+	+	-
7.	Hydrolysis of casein	+	+	+	+
8.	Hydrolysis of esculin	+	+	-	-
9.	Hydrolysis of Gelatin	+	+	+	-
10.	Nitrate reduction	-	-	-	-
11.	Growth at 3 % NaCl	+	+	+	+
12.	Acid production from glucose	+	+	+	+
13.	Phylogenetic identification	Bacillus megaterium	Paenibacillus alvei	Bacillus aryabhattai	Bacillus vietnamiensis

Table.4 Plant growth promoting traits of salt tolerant bacterial strains used in this study

Sl. No.	Isolate number and location	IAA production	Siderophore production	Phosphate solubilization
1.	OPS2(Bacillus megaterium)	+	+	+
2.	OPS4(Paenibacillus alvei)	+	-	-
3.	APS1(Bacillus aryabhattai)	+	-	-
4.	APS3(Bacillusvietnamiensis)	+	-	-

Table.5 Growth parameters of *Sesuvium portulcastrum* inoculated with bacterial consortia

Days	Pot culture		Field study		Pot culture		Field study	
	Control	Inoculated	Control	Inoculated	Control	Inoculated	Control	Inoculated
	Shoot length (cm)		Shoot length (cm)		Root length (cm)		Root length (cm)	
0	13.25	13.26	12.1	13.08	16.6	17.3	15.9	14.4
30	22.78	20.3	24.1	30.6	34.6	27.3	23.4	32.7
60	32.93	29.7	38.4	42.0	30.4	37.8	30.8	41.4
Mean	23	21	25	29	27	27	23	30

Table.6 Biomass content (g pot^{-1}) of the *Sesuvium portulcastrum* inoculated with bacterial consortia

Days	Pot culture		Field study	
	Control	Inoculated	Control	Inoculated
30	56	172	90	255
60	173	262	270	475
Mean	115	217	180	365

Table.7 EC and Sodium content of the *Sesuvium portulcastrum* cultivated soil inoculated with bacterial consortia

Days	Pot culture		Field		Pot culture		Field	
	Control	Inoculated	Control	Inoculated	Control	Inoculated	Control	Inoculated
	EC (dSm^{-1})				Na ($mgkg^{-1}$)			
0	13.5	13.5	13.5	13.5	3500	3500	3500	3500
30	6.5	7.5	8.5	5.5	1950	1900	1850	1750
60	4.0	4.5	5.5	3.2	980	850	870	700
Mean	8	9	9	7	2143	2083	2073	1983

Table.8 EC and Sodium content of the *Sesuvium portulcastrum* cultivated soil

Treatments	EC (dSm^{-1})			Na($mgkg^{-1}$)		
	Initial	30 DAP	60 DAP	Initial	30 DAP	60 DAP
Soil + Sesuvium portulacastrum	10.3	6.2	4.5	3200	1800	880
Soil + Sesuvium portulacastrum + Vermicompost (5tha-1)	10.5	4.8	3.1	2980	1580	730
Soil+ Sesuvium portulacastrum + Vermicompost (5tha-1) + ST-PGPR	10.1	3.8	2.8	3095	1420	610

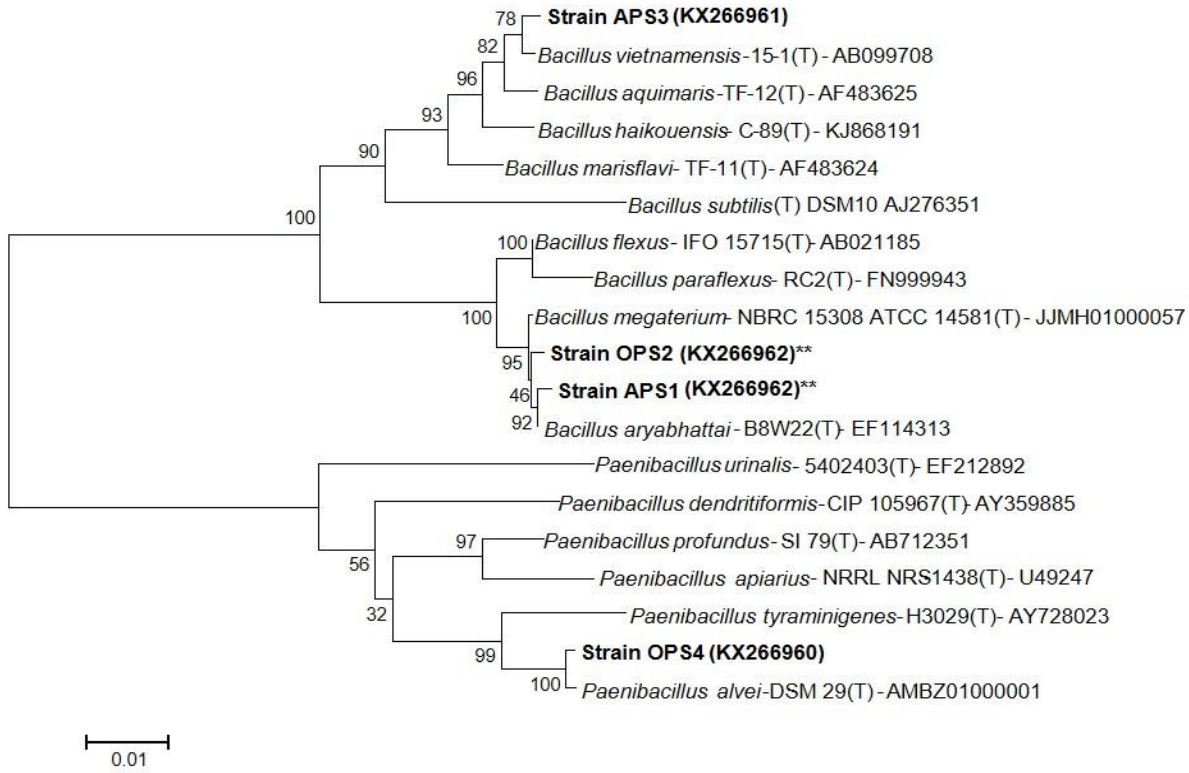
Table.9 Growth and biomass content of *Sesuvium portulcastrum*

Treatments	Root length (cm)			Shoot length (cm)			Biomass (g pot-1)		
	Initial	30 DAP	60 DAP	Initial	30 DAP	60 DAP	Initial	30 DAP	60 DAP
Soil + Sesuvium portulacastrum	-	12.3	18.5	5.8	16.2	21.5	25	163	175
Soil + Sesuvium portulacastrum + Vermicompost (5tha-1)	-	18.7	23.6	5.3	21.7	28.2	30	280	430
Soil+ Sesuvium portulacastrum + Vermicompost (5tha-1) + ST-PGPR	-	20.2	25.7	6.1	20.4	30.5	26	320	570

Table.10 EC and Sodium content of the *Sesuvium portulcastrum*

Treatments	EC (dSm-1)			Na(%)		
	Initial	30 DAP	60 DAP	Initial	30 DAP	60 DAP
Soil + Sesuvium portulacastrum	8.5	11.2	14.7	3.5	3.9	4.1
Soil + Sesuvium portulacastrum + Vermicompost (5tha-1)	8.7	10.3	11.9	3.1	3.4	3.7
Soil+ Sesuvium portulacastrum + Vermicompost (5tha-1) + ST-PGPR	8.4	9.7	10.6	3.0	3.2	3.5

Fig.1 Phylogenetic position of the isolates recovered from rhizosphere soil of *Sesuvium portulacastrum.* The phylogeny analysis was carried out using Mega 4.0, the distance was calculated using Kimura two parameter model and clustering with using Neighbour joining algorithm. Bootstrap values were determined based on 1000 replications. Bar indicates 0.01 substitutions per site



**Belongs to a taxonomic group (*Bacillus megaterium* group) includes species/subspecies that are not distinguishable by16SrRNA sequence

The maximum root length (25.7 cm), shoot length (30.5 cm) and biomass (570 g pot^{-1}) was observed in the treatment Soil+*Sesuvium portulacastrum* applied with Vermicompost (5tha^{-1}) and ST-PGPR (Table 9).

The initial sodium content *Sesuvium portulcastrum* is from 3.0 to 3.5%. The lowest sodium content of 3.5% was observed in the plant in the treatment Soil+ *Sesuvium portulacastrum* applied with Vermicompost (5tha^{-1}) and ST-PGPR at 60DAP.The highest sodium content was observed in the plant in the treatment Soil+ *Sesuvium portulacastrum*was4.1% at60DAP (Table 10).

The results in the pot culture experiment were mirrored on the field study also, as the plants able to withstand the salt stress and adopt to the soil conditions. It was already documented that is can able to withstand a salt spray and grows in the coastlines in the tropical and sub-tropical shore line (Lokhande *et al.*, 2012). Generally, under salt stress condition, in *S. portulacastrum* decrease in the root and shoots leading to lesser biomass accumulation. However, in the present study due to the presence of the plant growth promoting *Bacillus* sp. the roots, shoot and biomass level were increased in the inoculated plants. Similar effects on plant

growth promoting bacteria were documented earlier by halotolerant bacteria like *Brevibacterium, Planococcus, Zhihengliuella, Halomonas, Exiguobacterium, Oceanimonas, Corynebacterium, Arthrobacter,* and *Micrococcus* (Siddikee *et al.,* 2010). Thus the present also emphasise the need for a phytoremediating agent (*S. portulacastrum*) and bioinoculants to augment the salt stress under salt contaminated soils.

In conclusion, the experimental results revealed that these bacterial strains having salt tolerant potential and also PGPR activities so this can be effectively utilized as a bioinoculant for better crop growth in the salt affected soils.

References

Barassi, C., Ayrault G., Creus C., Sueldo, R and Sobrero M. (2006). Seed inoculation with *Azospirillum* mitigates NaCl effects on lettuce. Scientia Horticulturae, 109: 8–14.

Bharti, P.K., and Chauhan, A., (2013). Soil quality and contamination, Discovery Publishing House, New Delhi, India p. 186

Dodd, I.C., and Perez-Alfocea, F., (2012). Microbial amelioration of crop salinity stress. J. Exp. Bot. 63: 3415–3428

Eswaramoorthi, S., Dhanapal K., and Chauhan, D. (2008). Advanced in textile wastewater treatment: The case for UV-ozonation and membrane bioreactor for common effluent treatment plants in Tirupur, Tamil Nadu, India. Coimbatore, India: Environment with People's Involvement & Co-ordination in India.

Feizi, M., Hajabbasi, M.A. and Mostafazadeh, F.B. (2010). Saline irrigation water management strategies for better yield of safflower (*Carthamus tinctorius* L.) in an arid region. Aust. J. Crop Sci., 4: 408- 414.

Grattan, S.R., Grieve C.M. (1999). Mineral nutrient acquisition and response by plants grown in saline environments. In: Pessarakli M. (Ed.): Handbook of Plant and Crop Stress. Marcel Dekker, New York: 203–229.

Grover, M., Ali, Sk. Z., Sandhya, V., Rasul, A., Venkateswarlu, B. (2011).Role of microorganisms in adaptation of agriculture crops to abiotic stresses. World J. Microbiol. Biotechnol., 27: 1231-1240.

Gordon, S.A., and Weber R.P. (1951).Colorimetric estimation of indole-acetic acid. Plant Physiol., 26: 192-195.

Hussein, F.H., (2013). Chemical properties of treated textile dyeing wastewater. Asian J Chem 25:9393–9400.

Lokhande, V.H., Gor B.K., Desai N.S., Nikam T.D. and Suprasanna P. (2013). *Sesuvium portulacastrum*, a plant for drought, salt stress, sand fixation, food and phytoremediation. A review Agron. Sustain. Dev 33: 329-348.

Manzoor, Q., Rizwan Q., and Nawal A., (1997). Nutrient availability in a calcareous saline-sodic soil during vegetative bioremediation. Arid Land Res. Manag. 11: 343-352.

Mcclung, G., and Frankenberger, W., (1985). Soil nitrogen transformations as affected by salinity. Soil Science. 139: 405-411.

Mehta, S., and Nautiyal, C., (2001). An efficient method for qualitative screening of phosphate-solubilizing bacteria. Curr Microbiol 43: 51-56.

Nautiyal, C.S., (1999). An efficient microbiological growth medium for screening phosphate solubilizing microorganisms. FEMS Microbiol Lett. 170:265-70.

Patten, C.L., and Glick, B.R. (2002). Role of *Pseudomonas putida* indoleacetic acid

in development of the host plant root system. Appl. Envtl. Microbiol., 68: 3795-801.

Pessarakli, M., and Szabolcs I., (1999). Soil salinity and sodicity as particular plant/crop stress factor. In M. Pessarakli (ed.), Handbook of Plant and Crop Stress. Marcel Dekker Press Inc. New York, pp. 1-16.

Rajeswari, K., Subashkumar R., and Vijayaraman K. (2013). Physicochemical parameters of effluents collected from Tirupur textile dyeing and CETP and analysis of heterotropic bacterial population. J Microbiol Biotechnol Res 3:37-41.

Rashid, M., Khalil, S., Ayub, N., Alam, S and Latif, F. (2004). Organic acids production and phosphate solubilization by phosphate solubilizing microorganisms (PSM) under *in vitro* conditions. Pak. J. Biol. Sci., 7, 187-196.

Rodriguez-Valera, F., (1988). Characteristics and microbial ecology of hypersaline environments, pp. 71-84. In: Halophilic bacteria. Vol 1 (F. Rodriguez-Valera, (Ed.). Boca RatonJFL CRC Press Inc.

Siddikee, M., Puneet, C., Rangasamy, A., Gwang-Hyun H and Sa. T.M. (2010). Isolation, characterization, and use for plant growth promotion under salt stress, of ACC deaminase-producing halotolerant bacteria derived from coastal soil. J. Microbiol Biotechnol. 20: 1577-1584.

Singh, Y.P., Dubey U.C, Singh S and Dubey S.K. (2013). Interventions of sodic soil reclamation technologies and constraints in their adoption. Indian Res. J. Ext. Edu. 13:36-40.

Subbarao, N.S., (1988). Phosphate solubilizing microorganism. In: Biofertilizer in Agriculture and forestry. Regional Biofert. Dev. Centre, Hissar, India, 133-142.

Tien, T., Gaskin, M., and Hubbel, D. .(1979). Plant growth substances produced by *Azospirillum brasilense* and their effect on the growth of pearl millet (*Pennisetum americanum* L.). Appl Environ. Microbiol. 37: 1016–1024.

Upadhyay, S.K., Singh D.P and Saikia, R. (2009). Genetic diversity of plant growth promoting rhizobacteria isolated from rhizospheric soil of Wheat under saline condition. Curr. Microbiol., 59:489-496.

Upadhyay, S.K., Singh, J.S., Saxena, A.K and Singh D.P. (2011).Impact of PGPR inoculation on growth and antioxidant status of wheat under saline conditions. Plant Biol., 14: 605-611.

Ventosa, A., Ramos-Cormenzana, A and Kocur, M. (1983). Moderately halophilic Gram-positive cocci from hypersaline environments. Sys. Appl. Microbiol. 4: 564-570.

Wichern J., Wichern F., and Joergensen R.G., (2006). Impact of salinity on soil microbial communities and the decomposition of maize in acidic soils. Geoderma 137: 100-108.

How to cite this article:

Kalaiselvi, P., R. Jayashree and Poornima, R. 2019. Plant Growth Promoting *Bacillus* spp. and *Paenibacillus alvei* on the Growth of *Sesuvium portulacastrum* for Phytoremediation of Salt Affected Soils. *Int.J.Curr.Microbiol.App.Sci.* 8(04): 2847-2858.
doi: https://doi.org/10.20546/ijcmas.2019.804.332