

Abstract

Environmental factors like “salinity” and “biotic” stresses are alarming causes affecting the plant growth and crop production. Bacterial strains like *Pseudomonas spp.* show PGPR activity as well as biocontrol effect in presence of many pathogens and fungi. Also, PGPR traits have been reported in *Enterobacter spp.* and *Rhizobium spp.*, respectively. However, individual organisms cannot effectively ameliorate the stress due their metabolic restrictions. Therefore, a consortium of PGPR tends to be more effective, which is also subject to the compatibility within the consortium. All these factors lay an important role stress alleviation, which is the basis for this study. Hence, it is of crucial importance to understand the microbial interaction within the consortia under stress, so as to establish a consortium which can lead to alleviation of multiple stress factors in plants.

Herein, the PGPR traits have been studied for various PGPR strains belonging to multiple genera under salt stress individually as compared to in consortia of different combinations. Using the abundance of bacterial strains within the consortia, it was attempted to correlate their contribution in PGPR behaviour and survival under salt stress. This also helps us demonstrate their inter-dependence and synergy within the consortia. Plant inoculation studies were performed to assess the behaviour of the strains as assessed under laboratory conditions under pot trials using pigeon pea plants

Effect of salt stress on PGPR traits of selected strains individually and in consortium: Ten PGPR strains belonging to *Pseudomonas spp.*, *Enterobacter spp.* and rhizobial strains were subjected to growth in salt stressed conditions in media amended with 1 – 5% NaCl (w/v) and their capability for exhibition of various PGPR traits, including phosphate solubilization, indole acetic acid (IAA), siderophore, ammonia, biofilm and HCN production, individually as well as in consortium, under these saline conditions was assessed. The antagonistic activity via cross inoculation & fungal inhibition as well as the synergistic interaction via siderophore cross-utilization assays, among the PGPR consortia, was also established. It was found that several consortia demonstrated better stress tolerance and responded with increased PGPR effect suggesting synergy between these multispecies combinations. Among the 10 bacterial strains under study, varying levels of salt tolerance was observed depending upon the PGPR group to which the strain belonged. The PGPR traits in EC1D were least affected by salt stress. The rhizobial strains were good IAA producers while pseudomonads showed the increased biofilm and antifungal activity under salt stress. The PGPR traits showed different effect under stress, IAA and siderophore being negatively affected and biofilm positively affected by salt stress. The PGPR trait activity was enhanced substantially in consortium

especially for the biofilm, siderophore and phosphate solubilization which was dominated by the *Pseudomonas* strain.

Functioning and compatibility of the PGPR consortia in presence of different abiotic stresses using PCR based approach: Genus specific primers were designed based on the 16S partial gene sequences which were able to specifically amplify the individual members of mixed community. The bacteria were subjected to phosphate deficiency under saline condition to establish the interaction among the bacterial strains in a consortium by growth assays and to study the effect of consortium development on the habitat stress for sustenance under these harsh conditions. The bacterial and consortial samples were subjected to amplification using the genus specific primers and analysed by semi-quantitative PCR method and q-PCR methods for relative growth assessment in comparison to individual strains grown under the same condition. Results showed patterns of consortia member populations under the different conditions indicating which members were most abundant under different stress conditions. Most of the consortia strains demonstrated 2-fold increase in P-solubilization. The relative bacterial growth assessment via PCR studies revealed that the high optical density in growth assays did not correlate to the bacterial density. Evaluation of bacteria in consortium when subjected to salt stress revealed synergistic effect on bacterial growth under salt stress, especially *Rhizobium* spp. which were present at high salt concentrations where the growth in individual experiments was inhibited.

Plant inoculation studies with selected PGPR consortia for biotic and salinity stress with *Cajanus cajan* Seeds were subjected to various salt concentrations for *in vitro* germination via plate assay and hydroponic technique. Pot studies were carried out using the PGPR strains and the abiotic stress alleviation was assessed via plant growth parameters. In another set of experiments the plants were subjected to biotic stress using *Fusarium* infested soil. The seedlings treated with the PGPR consortia were assessed for the biotic stress alleviation via disease incidence and the infection symptoms observed on plants. Pigeon pea plant was found to be highly sensitive towards salt stress. PGPR treatment showed several folds enhancement in the biomass accumulation, protein, sugars, proline, carotenoid and even chlorophyll content. The consortia treatment was found to be more effective in stress alleviation as compared to individual strains. In biotic stress condition, the plants treated with PG22 and most of the consortia showed more than 50% decrease in the disease incidence.

In conclusion, the PGPR consortia with the ability to demonstrate salt stress as well as biotic stress alleviation in the plants has been established. This work can be applied in field studies for stress alleviation in pigeon pea and can be extended towards other leguminous and non-legume crops for the development of sustainable agricultural practices.