ABSTRACT

Utilization of plant growth promoting (PGP) endophytic bacteria could be an effective strategy in the implementation of sustainable agriculture to enhance crop productivity. The present study firstly explores the community structures, habitations, and potential PGP attributes of endophytic bacteria from the rice plant through next generation sequencing (NGS), which is a prerequisite to isolation, characterization and developing formulation of selected endophytes for testing PGP traits in rice crop. The taxonomic abundance and diversity of seed-transmitted endophytic bacteria inhabiting in vitro grown root, shoot and callus tissues of two aromatic rice cultivars were unraveled through 16S rRNA gene-based Illumina NGS. Wide variability in the number of bacterial operational taxonomic units and genera was observed between the two root and shoot samples, but not between the two callus samples, indicating tissue-specific and genotype-dependent bacterial community distributions in rice plant, even under similar gnotobiotic growth conditions. Once new insights on utilizing callus cultures for isolation of seed-transmitted endophytic bacteria were obtained; subsequently nine bacterial endophytes, designated as PB001-PB009, were isolated from *in vitro* grown calli of the two rice cultivars. Biochemical investigations validated in silico functional annotation that they possess several PGP traits, such as phosphate solubilization, indole acetic acid biosynthesis, nitrogen fixation, and production of ammonia, amylase, siderophore and 1-aminocyclopropane-1-carboxylic acid deaminase. Findings in gnotobiotic conditions revealed an increase in fresh and dry weights, and root and shoot lengths of seedlings germinated from endophyte-primed seeds than the control (uninoculated) set in a non-host and two host rice cultivars. In net house experiments, plants germinated from Micrococcus sp. PB001, Pseudomonas sp. PB002, Methylobacterium sp. PB005 and Methylorubrum sp. PB009-primed seeds of the non-host rice cultivar showed an increase in several vegetative and reproductive parameters than control plant sets, further validating intercultivar PGP abilities of these endophytes by modulating the abundance of certain sets of plant metabolites. Based on the growth compatibility between the bacterial isolates, four single-strain and two multi-strain talc-based formulations were developed, all of which exhibited a shelf-life of upto 90 days at room temperature. Preliminary field trials established that the single-strain bioinoculant formulation of Micrococcus sp. PB001 demonstrated the best intra-cultivar and inter-cultivar PGP traits, leading to improved rice crop productivity; thereby serving as a promising solution to practise eco-sustainable agriculture.

Keywords:

Bioinoculant formulation; Community structure; Endophytic bacteria; *In vitro* grown rice tissues; Intra- and inter-cultivar; Next generation sequencing (NGS); Gnotobiotic conditions; Plant growth promotion (PGP); Rice productivity.