Microbiome-based rhizosphere engineering for mitigation of salinity stress in *Vigna radiata*

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Abstract

The indiscriminate use of chemical fertilizers and pesticides in agriculture, and rising global temperatures, lead to salt accumulation in the soil, thereby negatively impacting plant physiology and crop yield worldwide. This has significantly affected the global economies, as agriculture is essential for ensuring food security and reducing poverty in many parts of the world. To address this grave issue, researchers have turned to eco-friendly approaches of utilizing beneficial microbes residing in the rhizosphere.

In present study, an indirect plant-assisted microbiome-selection strategy was adopted over several passaging cycles to acclimatize the indigenous microbiome such that it benefitted the growth of *Vigna radiata* plants under salt-stress conditions. The plant fitness was assessed over successive cycles in terms of plant growth and physiological parameters. Results showed an increased tolerance of plants to salinity stress, as evidenced by enhancement in plant growth attributes and decreased stress marker levels/indicators such as proline, malondialdehyde, and membrane stability index. Amplicon sequencing, employing the 16S rRNA and ITS as genetic markers, revealed significant changes in the microbial community inhabiting the rhizosphere of *Vigna radiata* under salinity stress across the passages.

Once the salt stress-adapted rhizospheric microbiome was obtained, the next objective was to understand the mechanism of action in salinity stress mitigation in *Vigna radiata*. Bacterial strains were isolated from stress-acclimatized microbiome using group-specific media to generate a culture bank. As determined by split plate assay, specific bacterial isolates were found to produce volatiles, possibly contributing to salt stress mitigation in *Arabidopsis*. The best-performing plant growth-promoting bacterial strains were selected to analyse their volatilomes by gas chromatography–mass spectrometry (GC-MS).

To overcome the technical limitations associated with the storage of such an acclimatized microbiome, synthetic microbial communities (SynComs) were designed from the culture bank generated from the acclimatized microbiome. SynComs were generated to perform *at par* with the stress-acclimatized microbiome, while mimicking its functionality in mitigating salinity stress in plants grown in arable land. The salinity stress mitigation potential of these SynComs was assessed first under control, and subsequently in field conditions.

The efficacy of the multi-passaging technique in rhizosphere engineering and the utilization of SynComs has been demonstrated through an extensive investigation conducted in laboratory and field settings. The study presents opportunities for implementing such a unique approach of combining top-down and bottom-up strategies with other crops and under diverse stress regimes. This approach promises an essential and favourable step toward ushering sustainable agricultural practices worldwide.