

Rhizosphere hybridization: Sustaining citrus production via microbiome changes

Rhizosphere security has become a pre-requisite to sustain the production by exploiting the microbiome profile of acid lime, and fortifying with microbes via rhizosphere hybridization is one effective way of deciphering the interaction between plants and microbes. The lost photosynthates from plants through root exudates creates a novel environment to harbour a diverse group of microorganisms, as a result, microbes-mediated rhizosphere exerted a diversified role in physiological plant growth and development. These microbes can be altered through various mean such as microbial fortification, soil amendments through various organic sources, rhizosphere engineering, inoculation of genetically engineered microbes to achieve a biased but desirable rhizosphere for growth and development of citrus. The biased but desirable changes in plant holobiont will reflect in changes in soil enzymes and plant available nutrients vis-a-vis crop phenology and its horticultural performance in the field. The process of rhizosphere hybridization is purely dependant on the selection of tree species with unique rhizosphere biomass, its activation of growth promoting rhizobacteria and its method of application, which all together makes a unique engineered rhizosphere. However, future studies must be directed towards upscaling the concept to a real field application, with more emphasis on soil-plant health safeguard as continuum.

FRUIT crops present an altogether different rhizosphere properties compared to any other field crops by the virtue of rootstock-scion combination, long juvenile phase, perennial nature of framework, 270-300 days of growing cycle etc., in addition to great variation in nature and composition of rhizodeposition triggering development of specific rhizosphere microbial communities, eventually quite different from rhizosphere properties of other crops. Around 40% of plant photosynthates has been observed to be lost by root systems, which resulted a higher nutrient status of rhizosphere as compared to bulk soil. Because of this, rhizosphere harbours a greater diversity of microbiome and its activity in rhizosphere soil than non-rhizosphere soil. As a result, microbes-mediated rhizosphere exerted a diversified role in physiological plant growth and development. Soil health with respect to physical and chemical properties is dependent on its biological activity, as microorganism plays a crucial role in active soil functions. These diversified microorganisms in soils are involved in maintenance of soil health and quality. Soil microbial community structure solely depend on soil type and plant type, which continue their diversified role in growth of plant in a complex manner. The microbiome diversity of specific rhizosphere depends on the genotypic characteristics of a plant such as species, cultivar, age of plant and root architecture. The microbial

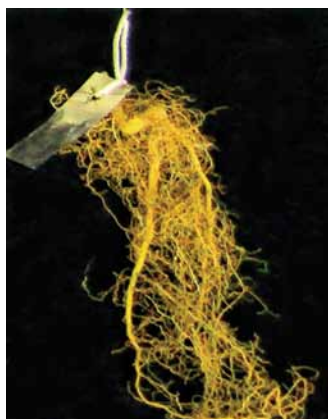
communities in alliance with acid lime either above or below ground part of the plant are known to constitute two tier structural niches for plant microbiome, which support genetically diverge microbial communities viz., bacteria, fungi, actinomycetes linked either with different compartment of plant microbiome such as rhizosphere, phyllosphere or endosphere. In terms of microbial diversity of microbiome, rhizosphere and endosphere microbiome is reported to have higher diversity than phyllosphere and epiphytic microbiome.

Rhizosphere properties of citrus

Soil microorganisms inhabiting citrus rhizosphere play a crucial role in improving soil ecology by changing the bio-physico-chemical properties of soil and transform the nutrient in readily available form for the growth and development of plant. In response to the biological indicator for soil fertility, soil microorganisms are the main factor to be considered. Previous report on citrus rhizosphere enlists a lot of fungi including *Absidia corymbifera*, *Aspergillus fumigatus*, *Emericella nidulans*, *Penicillium diversum*, *Paecilomyces variotii*, *Rhizomucor pusillus*, *Talaromyces thermophilus* and *Thermomyces lanuginosus*. In addition, some Arbuscular Mycorrhizal Fungi (AMF), including *Glomus*, *Acaulospora*, *Entrophospora*, *Gigaspora* and *Scutellospora* species were frequent in occurrence in



Control



Hybridized rhizosphere soil
of banyan, bamboo and
neem

Root architecture affected by rhizosphere hybridization

citrus orchards. The dynamic environment created by the soil microbes in plant rhizosphere help in growth and development through decomposition and nutrient cycling. Hence, the diversity in microbial population solely depends on root density and ability of the plant to absorb nutrients from the soil. Hence, microbial environment of plant rhizosphere is expected to differ according to plant species grown on the same rhizosphere soil.

Sacred trees with unique rhizosphere properties

The tree species of banyan (*Ficus benghalensis* L.), bamboo (*Dendrocalamus strictus* (Roxb.) Nees), neem (*Azadirachta indica* A. Juss) possessing inherent ability of high biomass production, higher microbial activity and higher geographic adaptability should be considered to be used as bio-inoculant. In the traditional agriculture, the virgin rhizosphere soil of these plants were considered to be sacred and used in festivals, wedding and agriculture to make nutrient rich product like *Jeevamrit* and *Vijamrit*. However, their use as nutrient source was confirmed by the presence of microbes in their respective rhizosphere in various literature.

Rhizosphere hybridization: An upshot

Inoculation of rhizosphere soil with rhizospheric or endophytic microbes, is likely to improve the functionality of microbiome for improved nutrients supply to various plant tissues including roots, leaves, and fruits. Artificially,

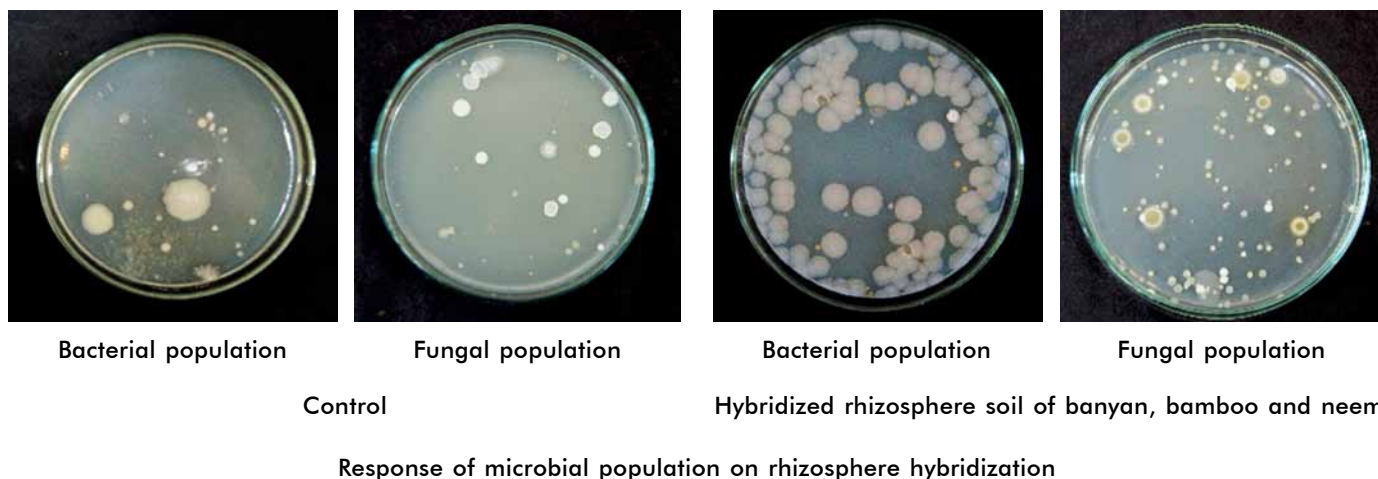
the rhizosphere is often modified or reconstructed matching to plant metabolism for reinforcing the physiological efficiency of plants by the process of rhizosphere engineering, rhizosphere hybridization, creating a better simulated environment for microorganisms to create a wide network of biological niche helping the growing acid lime plants through an elevated safeguard system against pathogens and supplying staggered concentration of nutrients through nutrient recycling. Such attempts of rhizosphere hybridization exploiting both functional and structural diversity of microbiome are almost non-existent. To develop a better rhizosphere environment for acid lime, microbes from wild species such as banyan, bamboo and neem can be introduced.

Methodology

To carry forward the rhizosphere hybridization, collection of unique rhizosphere soil from wild and sacred species is utmost important. These trees should be geographically away from the locality, unhampered by human interference, completed their juvenile period, producing high biomass in terms of luxuriant vegetation, fruit yield, tertiary root density and most importantly free from any kind of soil borne disease and pest. After the identification of tree species, it is very much important to activate the rhizosphere microbes. During the rainy season, the microbial activity is always at the peak. However, during winter and summer, the rhizosphere zone should be watered thrice in subsequent day to activate the rhizosphere microbes. The soils adhering to the tertiary root zone are considered to be the best for rhizosphere hybridization as these zone collect ample amount of plant growth promoting rhizobacteria. These rhizosphere soils can be used directly as a bio-



Total biomass comparison between hybridized rhizosphere and untreated control



inoculants or indirectly by rhizosphere water extract to carry forward the hybridization. Similarly, on recipient plant, rhizosphere zone should be activated prior to hybridization. To achieve the best result, the amount of rhizosphere soil taken from the donor parent must be incorporated into the rhizosphere of recipient parent in the same day of collection during early morning hour. To make rhizosphere water extract, rhizosphere soil and distilled water should be mixed in a ratio of 1:1 and kept untouched for 24 hours to release the microbes from soil to water. The supernatant liquid of this extract can be used as source of biofertilizers as rhizosphere water extract.

Seeing is believing

An experiment was set up at ICAR-Central Citrus Research Institute, Nagpur, Maharashtra, to observe the subsequent result of rhizosphere hybridization in acid lime. Earlier, we observed the value added response of rhizosphere hybridization carrying rhizospheres of banyan, bamboo and neem with acid lime rhizosphere either alone or in combination. The effect of the combined rhizosphere of banyan (*Ficus benghalensis*), neem (*Azadirachta indica*) and bamboo (*Bambusa vulgaris*) was significantly superior to the rest of the treatments in increasing the biometric (seedling height, seedling diameter and number of branches per seedling), number of leaves per seedling, root architecture (tap root length, tap root diameter and number of secondary roots per seedling) and microbial biomass (fungal count and bacterial count) response of acid lime seedling compared to control treatment. Rhizosphere soil treatment was found to be superior over rhizosphere water extract treatment. Among various rhizosphere water extract treatments, acid lime rhizosphere water extract performed better in biometric response of acid lime seedling.

While practicing such concept in field, we have to be absolutely sure that rhizosphere soil does not carry any

pathogenic microbe to infect host plant, rhizosphere has excellent microbial diversity evident from plant stand and rhizosphere soil is inoculated into the rhizosphere of host plant at a time when rhizosphere has sufficient moisture.

Future prospects

Microbial communities of any rhizosphere solely depends on genotype of the crop, its age, its developmental period, edaphological condition, climatic periodicity. However, these microbial communities can be altered by use of innovative techniques like rhizosphere hybridization, where a complete representative of plant holobiont is crossed or inoculated to an entirely new rhizosphere. The homologous microbiome has been proved to impart better result over non-homologous microbiome. On the contrary, forest tree species possessing biotic and abiotic stress tolerance may be considered as an alternative. Introduction of these new technologies may sustain the crop productivity in long term as the world is shifting from inorganic to organic fruit production. Instead of using large scale production of biofertilizers, these indigenous methods may change the scenario of biofertilizers use in fruit crop production. Such novel concept needs to be domesticated for farmers in not only other horticultural crops, but field crops as well. The concept has already been recognised by ICAR as a part of international training course (Dr A.K. Srivastava as Course Director), and can be referred through www.icar.org.pp.57 for more details.

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**Flowers always make people better, happier, and more helpful;
they are sunshine, food and medicine for the soul.**

– Luther Burbank