Original Article

Studies on Rhizosphere Hybridization and Nutrients Dynamics in Sweet Orange Growing Soil

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Abstract

The present investigation entitled was carried out to "Studies on Rhizosphere hybridization and nutrient dynamics in sweet orange growing soils was carried out during 2013-14 at department of Soil Science and Agricultural Chemistry,VNMKV, Parbhani. The Rhizosphere soil samples were collected from three religious trees such as *Ficusreligiosa* (Pimpal), *Ficusbenghalensis* (Banyan tree / wad) and *Ficus racemosa* (Umber tree). The soil samples were collected from 15-20 cm depth of root zone and used for pot culture experiment. The seedlings of sweet orange were planted in pots filled with rhizosphere soils collected from 3 *Ficus* species. Pot culture experiment was carried out with 9 treatments and 3 replications. The treatments were T_1 (Control Vertisol soil), T_2 (Inceptisol soil), T_3 (Soil under *Ficus religiosa* tree), T_4 (Soil under *Ficus benghalensis* tree), T_5 (Soil under *Ficus racemosa* tree), T_6 (*Ficus religiosa* + Inceptisol), T_7 (*Ficus benghalensis* + Inceptisol), T_8 (*Ficus racemosa* + Inceptisol), T_9 (Soil from sweet orange orchard). Results indicated the positive effect of rhizosphere hybridization on sweet orange seedlings growth parameters viz., number of leaves, height of seedling and stem diameter. It was noticed that all growth parameters were improved significantly in rhizosphere soils of *Ficus religiosa*, *Ficus benghalensis* and *Ficus racemosa* tree species and its hybridized soils over control, normal soil and soil from sweet orange orchard. The improvement in growth attributed to the improvement in physical properties, favorable pH, macro and micro-nutrient availability due to rhizosphere soil and its mixing with normal sweet orange garden soil.

Keywords

Rhizosphere; Hybridization; Ficus religiosa; Ficus; benghalensis; Ficusracemosa; Sweet orange growth and Development.

Introduction

The rhizosphere refers to the soil region nearest to the plant root system and is characterized by a high microbial activity. Roots release large quantity of metabolites from living root hairs or fibrous root systems into rhizosphere. The rhizosphere inhabits lots of soil micro-organisms, such as bacteria, fungi and actinomycetes which compete for water, nutrients, space and sometimes improve their competitiveness by developing an intimate association with plant generally, about 3,000 to 5,000 kg of soil living organisms occurs in one hectare of soil. The metabolites act as chemical signals for bacteria to move to the root surface, also represent the main nutrient sources available to support growth and persistence in the rhizosphere. Some of the microbes that inhabit this area are bacteria that are able to colonize very efficiently the roots or the rhizosphere soil of crop plants. These bacteria are referred to as plant growth promoting rhizobacteria (PGPR). Direct plant growth promotion may result either from improved nutrient acquisition and/or from hormonal stimulation. Diverse mechanisms are involved in the suppression of plant pathogens, which is often indirectly connected with plant growth.

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In Hindu literature *Ficusreligiosa* (Pimpal), *Ficusbeng halensis* (Banyan tree) and *Ficus racemosa* (Umber tree) tree species are worshiped. It was observed from review of literature that the soil underneath the tree is full of beneficial organisms due to congenial environment for growth and development of the microorganisms. Hence the present investigation was carried out to find out the effect of rhizosphere soil from these trees and its influence on sweet orange.

Materials and Methods

Under the umbrella of project "studies on rhizosphere hybridization and nutrient dynamics in sweet orange tree" subprojects were carried out to achieve the objectives outlines for the study. In the fine phase soil samples were collected underneath the *Ficus* species trees to develop the microbial consortium for growth enhancement of sweet orange. This microbial consortium was then tested by conducted the pot culture experiment on sweet orange seedling in second phase and third phase, effect of this developed value added consortium on studies on growth ,yield and quality of sweet orange in farmer field. The pot culture experiment was carried out department of soil science and Agricultural Chemistry, Vasaantarao Naik Marathwada Krishi Vidyapeeth, Parbhani. during 2013-2014.

Collection of Soil Sample

The rhizosphere soil samples were collected underneath the three different trees viz. *Ficusreligiosa* (*Pimpal*),*Ficusbenghalensis* (*Banyan tree*)and *Ficus racemosa* (*Umber tree*). The soil samples were collected from 0 to 20 cm. depth below the tree rhizosphere. About 150 to 200 kg rhizosphere soil was collected from each tree for pot culture experiment. The soil samples were made free of pebbles and twigs and dried in shade and used for physical, chemical and biological properties.

Pot Culture Experiment

A pot culture experiment was conducted during September 2013 to October 2014 for 13 months on sweet orange seedlings. There were nine treatments viz. T_1 Normal, T_2 Inceptisol (Normal medium black cotton soil), T_3 Soil under *Ficus religiosa* tree T_4 Soil under *Ficus benghalensis* tree, T_5 Soil under *Ficus racemosa* tree, T_6 *Ficus religiosa* + Inceptisol, T_7 *Ficus benghalensis* + Inceptisol, T_8 *Ficus racemosa* + Inceptisol and T_9 Sweet orange orchard soil, replicated in to three replications. Each pot was filled with 20 kg soil of respective treatments except treatment T_6, T_7 and T_8

wherein 10 kg rhizosphere soil of respective tree and 10 kg normal soil was hybridized and used for pot culture experiment. All the pots were lined by plastic to avoid the nutrient loss. The initial microbial population, enzyme activity, macro and micronutrient status of each treatment was determined to know the supplying capacity of substrate and the relevant data are presented in Table1 and Table 2. The microbial population from these soil samples was evaluated. The soil samples showing maximum population of beneficial organisms i.e. *Rhizobium, Azotobacter, Azospirillum, PSB, Pseudomonas and Trichoderma* and hence these soils were hybridized with normal soil to find out the influence of hybridized soil on sweet orange seedlings.

Results and Discussion

Physico-Chemical Properties of Rhizosphere Soil

The data on physico-chemical properties are presented in Table 2. It is observed that rhizosphere soil *Ficus racemosa* recorded minimum bulk density (1.13 Mgm-1) followed by *Ficus benghalensis* rhizosphere soil (1.17 Mgm-1) and *Ficus religiosa* rhizosphere soil (1.21 Mgm-1). Further hybridization of these three rhizosphere soil with normal soil (Treatment T_6, T_7 and T_8) showed reduction in bulk density and increase in porosity to a greater extend while rest of the soils showed higher bulk density and reduced porosity. The reduction in bulk density and increase in porosity are the favorable soil property changes occurring due to hybridization. The improved porosity allows balanced water and air balance in soil that will influence the nature of root effluxes and may influence the colonization and activity of microorganism in the soil. Organic carbon content was very high in all rhizosphere soils of *Ficus* species as compared to normal soil, Inceptisol soil and soil collected from sweet orange orchards. Further there was increase in organic carbon to a greater extend because of hybridization. This increased organic carbon will act as food and energy source for rhizosphere flora and Fauna. All the soil pH in the rhizosphere of all tree species was tended towards alkalinity. However the soil under *Ficus* species trees more close to soil pH 7. The soil pH between 6.5 to 7.5 found to provide favorable condition for growth and development of microorganism.

These changes in favor of the rhizosphere flora were correlated with soil pH. The general trend of soil acidification by trees has been observed under a variety of circumstances (Augusto *et al.*, 2002; Jobbagy and Jackson, 2003; Sinha *et al.*, 2009) and has been

attributed to release of H+ ions from the respiration of plant roots/soil microorganisms (Hinsinger, *et al.*, 2006) or release of acidic or alkaline exudates in the rhizosphere (Hagen-Thorn *et al.*, 2004). The rhizosphere environment generally has a lower pH, lower oxygen and higher carbon dioxide concentrations. However, exudates can make the soil in the rhizosphare more acid as well as neutral and alkaline. The electrical conductivity is highest in control treatment is (0.51 dSm^{-1}) is followed by T_5 (0.23dSm^{-1}) T_4, T_3, T_8 T_7, T_6, and T_9 also safe in all the rhizosphere soil and its hybridized soils. The various in the EC are probably due to variation in roots exudates of these tree species because roots exudates increase salinity of the rhizosphere soil. Shen *et al* (2001). The organic carbon content was highest in treatment no T_5 (38.80 g /kg^{-1}) is followed by T_4, T_3, T_8 T_7, T_6, and T_9 and control treatment. All rhizosphereic plant species except control recorded a considerable amount of organic carbon in rhizosphere soil as compared with the critical values suggested by Masto, *et al* (2007). A tree rhizosphere is likely developing a microenvironment continuously under the effect of the root exudates, soil characteristics and climatic factors, giving an opportunity for development of a specialized rhizoflora.

Soils of all treatments did not show any specific trend in calcium carbonate content. This might be because the content of calcium carbonate is governed by soil genesis, as it being inherited from the parent material which takes more time to alter.

Macro and Micro Nutrient Content in Rhizosphere Soil

Macronutrients

The available N, P, K is highest in the rhizosphere soil under *Ficus* species (Treatment T_3, T_4 and T_5) followed by hybridized soils (T_6, T_7 and T_8). When a plant takes up nitrogen as ammonium it releases hydrogen ions which will make the rhizosphere more acid. When a plant takes up nitrogen as nitrate, it releases hydroxyl ions which make the rhizosphere more alkaline. This action doesn't usually affect the bulk pH of the soil but is important for the small organisms that live in the rhizosphere because many soil organisms do not move far in the soil. The availability of nitrogen in most soils is low because of the leaching losses of soluble nitrate (NO_3^-) with infiltrating rainwater, fixation of ammonium (NH_4^+) in clays and soil organic matter and bacterial denitrification. Plants respond differently depending on the form of nitrogen in the soil. Ammonium has a positive charge, and thus the plant expels one proton (H^+) for every NH_4^+ taken up

resulting in a reduction in rhizosphere pH. When supplied with NO_3^-, the opposite can occur where the plant releases bicarbonate (HCO_3^-) which increases rhizosphere pH. These changes in pH can influence the availability of other plant essential micronutrients (e.g., Zn, Ca, Mg). Phosphate (PO_4^{3-}), the form of P used by plants, is highly insoluble in soils, binding strongly to Ca, Al and Fe oxide, and soil organic matter rendering much of the P unavailable to plants. Under P deficiency, plants have evolved special mechanisms to obtain PO_4^{3-} which depend on plant type (dicot vs.monocot), species and genotype. Plant roots can exude organic acids such as malic and citric acids into the rhizosphere which effectively reduced rhizosphere pH and solubilize P bound in soil minerals. Plants also liberate PO_4^{3-} from organic sources by releasing enzymes such as acid phosphates. High amount of available potassium in rhizosphere soil is probably because of higher potassium bearing minerals like feldspar and mica in the parent material. High organic matter is also presence in rhizosphere soil which is increase the level of potassium in soil.

Micronutrients

The DTPA- Fe, Mn, Cu, and Zn is highest in treatment T_5 followed by T_4, T_3, and T_8,T_7, T_6 and T_9as compare to control treatment. Plant growth promotion can be shown to work directly on the plant in the absence of root pathogens by the release of plant growth stimulating compounds (e.g. phytohormones such as auxins or cytokinins) and improvement in mineral uptake (e.g. siderophore release increasing Fe availability). Plant growth promotion can also occur indirectly by control of pathogens (biocontrol) via synthesis of antibiotics or secondary metabolite-mediated induced systemic resistance (ISR) (van Loon, *et al.*, 1998, 2007).

Biological Properties of Rhizosphere Soil

The data presented in Table 3 indicated that microbial population such as bacteria, fungi and actinomycetes is very high in all treatment except control treatment. The largest number of Bacteria, Fungi and Actinomycetes was isolated from the *Ficus sp* rhizosphere soils. It was shown that the activity and growth of most soil inhabiting bacteria and fungi are enhanced by plant root exudates. This difference between the two populations of microorganisms can be explained through the beneficial effect that the plant roots exercise the microorganism in the rhizosphere. Further underneath the tree soil temperature ranges between 20 to 30 degree centigrade which might have

Table 1: Physico chemical properties and nutrient content of rhizosphere and hybridized soils

Tret no	Treatments	Physical properties			Chemical properties				Macro Nutrients				Micro Nutrients			
		B.D (Mg m⁻³)	P.D (Mg m⁻³)	Porosity (%)	pH	EC (dSm⁻¹)	OC (g kg⁻¹)	CaCO₃ (%)	Avail. N (kg/ha)	Avail P (kgha⁻¹)	Avail. K (kg ha⁻¹)	DTPAFe (mgkg⁻¹)	DTPA-Mn (mg kg⁻¹)	DTPACu (mgkg⁻¹)	DTPAZn (mgkg⁻¹)
T1	Normal	1.83	2.66	31.38	8.1	0.51	3.75	9.19	222.65	10.39	369.58	2.74	7.74	1.57	0.21
T2	Soil Inceptisol	1.53	2.38	35.74	7.9	0.33	7.95	7.75	520.57	18.02	448.13	5.39	15.73	2.12	0.54
T3	Soil under Ficus religiosa tree	1.21	2.45	50.81	7.0	0.29	23.70	5.12	1083.64	32.12	3157.05	15.18	26.28	8.13	0.68
T4	Soil under Ficus benghalensis tree	1.17	2.42	51.65	7.2	0.27	28.65	3.50	1207.17	37.19	3350.04	16.26	24.24	12.44	4.66
T5	Soil under Ficus racemosatree	1.13	2.49	54.98	7.5	0.23	38.80	2.07	1226.17	39.92	3541.37	18.89	38.21	15.52	6.84
T6	Ficus religiosa + Inceptisol	1.33	2.41	44.81	7.1	0.26	14.25	6.25	774.59	25.44	2581.82	14.23	20.12	13.14	1.53
T7	Ficus benghalensis + Inceptisol Soil	1.31	2.45	46.53	7.3	0.28	13.35	6.50	865.53	28.29	2638.27	12.15	22.23	11.19	3.54
T8	Ficus racemosa + Inceptisol	1.28	2.52	49.20	7.3	0.25	22.35	3.19	1101.08	29.11	3181.24	17.19	29.18	9.25	3.98
T9	Soil from Sweet orange orchard	1.38	2.29	40.61	7.7	0.33	7.81	5.87	620.92	19.76	669.54	8.17	18.8	3.85	0.80

Table 2: Biological properties of rhizosphere soils of ficustree species and other treatments

Tr.no	Treatments	Soil Microbial Population			Enzyme activity		
		Bacteria (Cfu x 10⁻⁶)	Fungi (Cfu x 10⁻⁴)	Actinomycetes (Cfu x 10⁻⁵)	Acid Phosphatase (ug p-N g⁻¹ soil ha⁻¹)	Alkaline Phosphatase (ug p-N g⁻¹ soil ha⁻¹)	Dehydrogenase (ug p-N g⁻¹ soil ha⁻¹)
T₁	Normal	32	03	12	34.78	155.23	62.34
T₂	Soil Inceptisol (Medium black)	42	07	19	55.89	173.76	79.80
T₃	Soil under Ficus religiosa tree	133	15	41	155.10	273.23	201.76
T₄	Soil under Ficus benghalensis	156	18	52	155.34	282.99	279.90
T₅	Soil under Ficus racemosa tree	186	21	58	157.82	287.59	298.13
T₆	Ficus religiosa + Inceptisol	112	12	38	133.03	174.94	132.17
T₇	Ficus benghalensis + Inceptisol	119	14	42	143.94	181.86	120.53
T₈	Ficus racemosa + Inceptisol	127	17	49	151.46	185.34	147.14
T₉	Soil from sweet orange orchard	88	11	31	67.59	178.77	81.46

Table 3: Growth observation of sweet orange seedling Pot culture experiment

Tr. no	Treatments	Initial Stage			Final Stage		
		Height of seedling (cm)	No. of Leaves	Steam diameter (mm)	Height of seedling (cm)	No. of Leaves	Steam diameter (mm)
T1	Normal	29.30	15.33	7.69	63.56 (34.26)	56.89 (41.56)	13.89 (6.20)
T2	Soil Inceptisol (Medium black)	33.43	15.66	7.43	67.25 (33.82)	69.61 (53.95)	14.68 (7.25)
T3	Soil under Ficus racemosa tree	29.26	16.00	7.70	81.96 (52.70)	96.69 (80.69)	15.59 (7.89)
T4	Soil under Ficus benghalensis	30.26	18.33	7.33	83.52 (53.26)	98.59 (80.26)	17.12 (9.79)
T5	Soil under Ficus racemosa tree	25.40	14.33	6.54	110.22 (84.82)	128.26 (113.93)	18.98 (12.44)
T6	Ficus religiosa + Inceptisol	33.13	13.33	7.61	79.54 (46.41)	82.35 (69.02)	16.56 (8.95)
T7	Ficus benghalensis + Inceptisol	24.43	16.10	7.90	85.69 (61.26)	95.66 (79.56)	16.92 (9.02)
T8	Ficus racemosa + Inceptisol	23.56	15.00	7.57	99.65 (76.09)	108.14 (93.14)	17.86 (10.29)
T9	Soil from sweet orange orchard	24.30	14.33	7.27	64.25 (39.95)	85.26 (70.93)	15.51 (7.24)

helped to increase the population of microbes in addition to high organic carbon and optimum pH.

The enzyme activity is also very high in T_5 is fallowed T_4, T_3, T_8 T_7, T_6 and T_9 is compare with control treatment. Acidic phosphates enzyme activity is low as compare to the alkaline phosphates activity. Soil enzyme assayed in this study were chosen because they play central role in the C, N, and P cycling in soils and application of N or P would affect the release of N or P. Therefore the P-releasing enzyme acid phosphatase and C- releasing compound increasing alkaline phosphatese. Dehydrogenase activity also high in Treatment T_5 *Ficus racemosa* is followed by T_4 *Ficus benghalensis* and T_3 *Ficus religiosa.*

Growth Observation of Pot Culture Experiment

The data on sweet orange seedling growth collected from one year pot culture experiment are presented in Table 4. The initial and final height, number of leaves and stem diameter was recorded and given in Table. Similarly to find out the effect of rhizosphere soil and hybridized soil treatments over other treatments incremental height, number of leaves and stem diameter was calculated and presented in parenthesis. It was noticed that all growth parameters were improved significantly in rhizosphere soils of *Ficus religiosa, Ficus benghalensis* and *Ficus racemosa* tree species and its hybridized soils over control, normal soil and soil from sweet orange orchard. The improvement in growth attributes attributed to the improvement in physical properties, favorable pH, macro and micronutrient availability due to rhizosphere soil and its mixing with normal sweet orange garden soil.

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