

Fungal Diversity in the Rhizosphere of Tropical Homestead and Plantation Crops of Kerala

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Abstract

A study was conducted on the fungal diversity in the home gardens of three important districts of Kerala. The rhizosphere and rhizoplane samples were collected from different homestead crops representing diverse and distinct areas of coastal region (Ernakulam district), High range (Wayanad) and Dry region (Palakkad district) districts of Kerala. Fungal population was highest (82×10^3 cfu g⁻¹ of soil) in black pepper (Wayanad) followed by coconut (70×10^3 cfu g⁻¹ of soil) in Ernakulam and 22×10^3 cfu g⁻¹ of soil in banana (Palakkad). There was greater diversity of fungi in the homesteads of Ernakulam, Wayanad and Palakkad districts. In all the three districts surveyed, *Penicillium* sp. and *Aspergillus* sp. were predominant fungi in most of the crops and or locations, even though other fungi were also recorded. However, there functional diversity needs to be studied and harnessed in order to assess the beneficial effects and relative efficiency on the host plants.

Highlights

- The rhizosphere and rhizoplane samples were collected from different homestead crops representing diverse and distinct areas of coastal (Ernakulam district), High range (Wayanad district) and Dry (Palakkad district) regions of Kerala.
- Fungal population was highest (82×10^3 cfu g⁻¹ of soil) in black pepper (Wayanad) followed by coconut (70×10^3 cfu g⁻¹ of soil) in Ernakulam and 22×10^3 cfu g⁻¹ of soil in banana (Palakkad).
- In all the three regions, *Penicillium* sp. and *Aspergillus* sp. were predominant fungi in most of the crops and / or locations, even though other fungi were also recorded
- However, there functional diversity needs to be studied in detail and harnessed in order to assess the beneficial effects and relative efficiency on the host plants.

Keywords: fungal diversity, homesteads, rhizosphere, rhizoplane

Homestead farming is a unique system adopted by majority of the farmers in Kerala in which small sized landholdings predominate. Homegardens are traditional farming systems which may have evolved over time from the practices of hunters / gatherers and continued in the ancient civilization

upto modern times, therefore is one of the oldest agro-ecosystems that exist throughout the world (Somerwato, 1987; Somerwato and Conway, 1992). Home gardens are an integral part of the livelihood systems, and could contribute to the family food, income and conservation of



biodiversity.(Shrestha, *et al.*, 2004). A typical Kerala homegarden consists of a dwelling house with small garden in front and a variety of annual and perennial crops (predominantly horticultural crops) grown in mixture on a small piece of land. As the homesteads are mostly cultivated in organic way, the rhizosphere microflora will differ from the fertigated fields. Microbial community of the rhizosphere is not only influenced by factors such as root exudates, phenology, and nutrient uptake but also by the plant species. The rhizosphere-host interactions have implications on the nutrient cycling and on physical and chemical nature of soil. There have been many studies of the microbial community of the rhizosphere soil of both cultivated and wild plants, revealing how this community is influenced by the presence of plant roots (Brodie *et al.*, 2002; McCaig, *et al.*, 2001). The host plant not only influences its own rhizosphere microbial community, but also the microorganisms play important roles in the growth of plant host and its ecological fitness. The rhizosphere microbes affect the plant by altering the supply and availability of inorganic nutrients, nitrogen fixation, producing plant growth hormones (Nieto and Frankenberger, 1989) and repressing plant pathogens (Berg *et al.*, 2005).The rhizosphere is also considered being chemically and microbiologically distinct from bulk soil (Kowalchuk *et al.*, 2002; Marilley and Aragno, 1999; Marilley *et al.*,1998).

A total of 45 genera distributed in 85 species were reported in agricultural soils collected from Aurangabad district of Maharashtra (Nagmani *et al.*, 2005). *Aspergillus* was dominant in all the three types of soils followed by *Alternaria*, *Cladosporium*, *Trichoderma*, *Gliocladium* and *Gloeosporium*. It was also found that more number of genera and species of fungi exist in soil than in any other environment and contributes to the nutrient recycling and maintenance of ecosystem. Fungi play an important role in soil formation, soil fertility, soil structure and soil improvement (Hao-quin *et al.*, 2008).Fungi decompose organic matter from humus, release nutrients, assimilate soil carbon and fix organic nutrients. Although some genera of soil fungi are intensively studied, little is known about fungal community structure and community dynamics in agricultural soils (Hagn *et al.*,2003). It is generally agreed that only a small percentage of the estimated 1.5 million fungi worldwide (Hawksworth and Rossman, 1997) have been cultivated yet. However, studies of microbial communities especially fungal diversity associated with crops grown in homegardens are lacking in India and

particularly in Kerala. Hence, the present study was undertaken with the main objective of probing the fungal diversity and its predominance in three different ecological homesteads of Kerala. The study involved isolation, enumeration and identification of fungi in the rhizosphere of different crops grown in the homesteads.

Materials and methods

A total of 45 rhizosphere soil samples were collected from different crops grown in homesteads representing diverse and distinct areas of coastal region (Ernakulum district), high range (Wayanad) and dry region (Palakkad) districts of Kerala. The samples were collected from major crops viz., banana, coconut, jack, black pepper, cashew, nutmeg, mangosteen, arecanut, mango, colocasia, gummigutta, mahagony, sapotta, rambuttan, betel vine, coffee, ginger, rubber, coconut, cocoa and teak in Ernakulam district. Similarly, 16 rhizosphere soil samples were collected from coconut, pepper, coffee, banana, cardamom, arecanut, rubber, cocoa, clove, nutmeg and vanilla in Wayanad district and 5 rhizosphere soil samples were collected from coconut, banana and arecanut in Palakkad district.

The fungi were isolated by the serial dilution and plate count method (Johnson and Curl, 1972) from each of the host. For isolation of total fungi, 10^{-3} and 10^{-3} dilution were used with Martin's Rose Bengal agar media and Nutrient agar media. After incubation at $28^{\circ}\text{C} \pm 2$, the fungal population was counted after 5 to 7 days. The fungi were purified by single-spore method (Choi *et al.*, 1999) on the suitable media and maintained for further studies.

All the fungal isolates obtained from different homesteads in different districts of Kerala were identified. The fungi isolated from Ernakulam, Wayanad and Palakkad districts of Kerala were repeatedly purified. Pure cultures of fungus from Ernakulam (128 No.), Wayanad (62 No.) and Palakkad (12 No.) districts were sent for identification to the M/s. National Centre for Fungal Taxonomy, New Delhi, India.

Results and discussion

Enumeration of fungal population: A total of 199 fungal isolates (128 isolates from Ernakulam, 59 isolates from Wayanad and 12 isolates from Palakkad) were obtained from rhizosphere soils of major crops of home-gardens namely coconut, black pepper, coffee, banana, cardamom, arecanut, rubber, cocoa, clove, nutmeg and vanilla.



In Ernakulam district, the fungal population varied from 5×10^3 to 70×10^3 cfu g^{-1} (Table.1). The maximum fungal population was recorded in the rhizosphere of coconut (70×10^3 cfu g^{-1}) followed by colocasia (51×10^3 cfu g^{-1}). The minimum fungal population was recorded in the rhizosphere soil of mangosteen (5×10^3 cfu g^{-1}). The least fungal population was in the rhizosphere of mangosteen as the mangosteen lacks root hairs. It clearly shows that the host plant also has a major role in influencing the rhizospheric fungi. In Wayanad district, the fungal population varied from 0 to 82×10^3 cfu. g^{-1} . The maximum fungal population was recorded in the rhizosphere of black pepper (82×10^3 cfu g^{-1}) and in some samples of pepper rhizosphere, the fungal population was 0 and in one location it was 62×10^3 cfu g^{-1} . This was followed by Cardamom (53×10^3 cfu g^{-1}). The minimum fungal population observed were in the pepper (0×10^3 cfu g^{-1}) and coconut (1×10^3 cfu g^{-1}) respectively. There was wide variation in the rhizosphere fungal population in pepper grown in Wayanad district. In Palakkad district, the fungal population varied from 13×10^3 to 22×10^3 cfu g^{-1} . The maximum fungal population was recorded in the rhizosphere soil of banana (22×10^3 cfu g^{-1}) followed by coconut and arecanut (20×10^3 cfu g^{-1}). The minimum fungal population was recorded in the rhizosphere soil of coconut (13×10^3 cfu g^{-1} .)

In general, the highest populations of fungi were recorded in black pepper (Wayanad), followed by coconut (Ernakulam) and banana (Palakkad). The studies clearly indicated that fungi are better adapted to the crop which is widely cultivated in the particular region.

Identification of fungal isolates

Coastal region (Ernakulam district)

A total of 129 fungi were recorded in different hosts under homesteads of Ernakulam district (Table 2). The common fungi found in the rhizosphere soils of colocasia, nutmeg, coconut, mangosteen, cocoa, banana, black pepper, jack tree, rubber, mahogany, mango, ginger were different species of *Penicillium* and *Aspergillus*. However, in the rhizosphere of arecanut and coffee, the fungi found were *Trichoderma viride* and *Trichoderma harzianum* respectively. *Trichoderma viride* and *Trichoderma harzianum* are biocontrol agents which are widely used in agriculture for the control of soil-borne diseases. The presence of these fungi in the rhizosphere helps in controlling soil-borne pathogens. In the present studies, these fungi in rhizosphere of major crops of Kerala indicated

that they can be potential biocontrol agents for soil borne plant pathogens. However, other fungi in the rhizosphere were either plant pathogen like *Choenophora cucubitarium* or human pathogen such as *Acremonium restrictum*. Some of the beneficial fungi found in the rhizosphere were *Chaetomium indicum* in banana and *Cunninghamella echinulata* in nutmeg which are beta, 1,3, glucanase producer and bio-absorbant of heavy metal in water respectively.

The fungi present in the rhizosphere of homestead crops had less number of beneficial fungi indicating need for increasing its population in the rhizosphere. Fungi are fundamental for soil ecosystem functions (Warcup, 1951) especially in forest and agricultural soils where essential processes such as organic matter decomposition, elemental release by mineralization and protection against leaching by elemental storage in biomass (Christensen, 1989). Their mycelia contribute to soil aggregate stability, thereby avoiding erosion. They develop associations with more than 90% of all land plants (Rayner, 1992) enabling and facilitating the transport of nutrients and water from soil to plant roots.

High range region (Wayanad district)

A total of 64 isolates of fungi were found from different hosts under homesteads of Wayanad district (Table 3). The Wayanad district is well known for home stead farms. The fungi were found in the rhizosphere of only arecanut, cocoa, banana, coffee, pepper, cardamum, coconut and banana. The fungi found in the homesteads of Wayanad districts were *Taloromyces flavus*, *Choenophora cucubitarum*, *Trichoderma hamatum*, *Penicillium* sp. *Aspergillus* sp, *Byssoclamus niveus*, *Trichoderma viride* and *Acremonium restrictum*. In this district, a critical scrutiny of the results point to the fact that the beneficial fungi were more in the rhizosphere than pathogens. The Wayanad is a district where organic farming is widely practiced and the addition of organic matter might have increased the beneficial fungi. However, there is also a need to inoculate efficient fungi which are beneficial to the agricultural crops. There is evidence from both crops and temperate trees of ways in which plants fundamentally alter the chemistry of the soil around their fine roots through exudates (Acosta-Martinez, 2008; Berg and Smalla, 2005) and through the activities of mutualistic fungi that inhabit the roots (Streitwolf and Sanders, 1997). The effects of root exudates and nutrient uptake are most intense on the rhizoplane (Wieland *et al.*, 2001). Therefore, the studies have clearly revealed the



effect of host plant on the fungal microflora.

Dry region (Palakkad district)

A total of 12 isolates of fungi were found from different hosts under homesteads of Palakkad district (Table 4). The fungi were found in the rhizosphere of coconut, banana and arecanut. The fungi found in the homesteads of Palakkad districts were *Penicillium* sp. and *Aspergillus niger* which are phosphate solubilizing microorganisms. Hence, there is need to inoculate the other beneficial fungi such as *Trichoderma* sp. which will help in the plant growth indirectly by suppressing soil-borne diseases.

Among the three districts with distinct ecological conditions, the black pepper had highest population of fungi (82×10^3 cfu g⁻¹) in Wyanad district followed by coconut (70×10^3 cfu g⁻¹) in Ernakulam district. Banana in Palakkad district had the least fungal population (22×10^3 cfu g⁻¹). The studies clearly indicated variation in the fungal population depending on the soil nutrient status. With respect to the diversity of fungal species, *Penicillium* sp., *Aspergillus* sp., *Trichoderma viride* and *Trichoderma harzianum* were observed in Ernakulam district where as in Wyanad district, other fungi like *Talaromyces flavus*, *Choenophora cucurbitarum*, *Byssoclamus nivens* and *Acremonium restrictum* were recorded in addition to the fungi recorded in Ernakulam district. The rhizosphere soils of different hosts collected from Palakkad district showed the presence of phosphate solubilizing fungi *Penicillium* sp. and *Aspergillus* sp. The studies confirmed the urgent need to enrich the ecosystem by inoculating beneficial fungi in all the three districts as they are important in nutrient cycling through organic matter decomposition.

Conclusion

The state of Kerala is one of the “hot spots” of microbial diversity and also the land of homestead farming. The different crops grown in the home stead are unique due its cultivation nature which is largely under organic cultivation. The soils of home stead are also unique due to the large application of organic fertilizers. In the present studies, the highest populations of fungi were recorded in black pepper (Wayanad), coconut (Ernakulam), and banana (Palakkad) among the samples collected and it was also found that there is greater diversity of fungi in the homesteads of the three districts. In all the three districts surveyed, *Penicillium* sp. and *Aspergillus* sp. were predominant compared to the other fungi. However, the functional diversity of different fungi present in the

homesteads needs to be studied in detail so that the beneficial effects of these rhizosphere microflora are fully exploited and utilized.

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References

- Acosta-Martinez, V., Dowd, S., Sun, Y. and Allen, V. 2008. Tag-encoded pyrosequencing analysis of bacterial diversity in a single soil type as affected by management and land use. *Soil Biology Biochemistry* **40**: 2762–2770.
- Berg, G. and Smalla, K. 2005. Impact of plant species and site on rhizosphere-associated fungi antagonistic to *Verticillium dahliae* Kleb. *Applied Environment Microbiology* **71**:4203–4213.
- Brodie, E., Edwards, S. and Clipson, N. 2002. Bacterial community dynamics across a floristic gradient in a temperate upland grassland ecosystem. *Microbial Ecology* **44**:260–270.
- Brundrett, M. 1991. Mycorrhizas in natural ecosystems. *Advances Ecology Research* **21**:171–313.
- Choi, Y.W., Hyde, K.D. and Ho, W.H. 1999. Single spore isolation of fungi. *Fungal diversity* 29-38
- Christensen, M. 1989. A view of fungal ecology. *Mycologia.*, **81**:1-19.
- Hagn, A., Karin, P., Michael S. and Jean, C. M. 2003. Fungal diversity in agricultural soil under different farming management systems, with special reference to biocontrol strains of *Trichoderma* spp. *Biology Fertility Soils* **38**: 236–244
- Hawksworth, D.L. and Rossmann, A.Y. 1997. Where are all the undescribed fungi. *Phytopathology* **87**:888–891.
- Johnson, L.F. and Curl, E.A. 1972. Methods for research on the ecology of soil borne plant pathogens. Burgess Publishing Co. Minneapolis, MN USA. 178.p
- Kowalchuk, G.A., Buma, D.S., de Boer W., Klinkhamer, P.G. and Van Veen, J.A. 2002. Effects of above ground plant species composition and diversity on the diversity of soil-borne microorganisms. *Antonie van Leeuwenhoek*. **81**:509–520.
- Marilley, L. and Aragno, M. 1999. Phylogenetic diversity of bacterial communities differing in degree of proximity of *Lolium perenne* and *Trifolium repens* roots. *Applied Soil Ecology* **13**:127–136
- Marilley, L., Vogt, G., Blanc, M. and Aragno, M. 1998. Bacterial diversity in the bulk soil and rhizosphere fractions of *Lolium perenne* and *Trifolium repens* as revealed by PCR restriction analysis of 16S rDNA. *Plant Soil* **198**:219–224.
- McCaig, A.E., Glover, L.A., Prosser, J.I. 2001. Numerical analysis of grassland bacterial community structure under different land management regimens by using 16S ribosomal DNA sequence data and denaturing gradient gel electrophoresis banding patterns. *Applied Environment Microbiology* **67**:4554–4559.



- Nagmani, A., I.K., Kunwar, Manoharachary C .2005. Handbook of soil fungi. Published by I. K. International Pvt, Ltd. New Delhi.
- Nieto, K.F. and Frankenberger, W.T. 1989. Biosynthesis of cytokinins in soil. *Soil Science Society American. Journal* **53**:735–740.
- Rayner, A.D.M.1992. Introduction. In: Carroll, G.C. Wicklow, D.T.(eds).The fungal community. Dekker, New York, pp 17-24.
- Soemerwato, O.1997. Homegardens: a traditional agroforestry system with a promising future. In: Stepple, H.A. and Nair, P.K.R. (Eds) .Agroforestry: A head of Development. ICRAF, Nairobi, Kenya. pp 157-170.
- Soemarwato A. and Conway, G.R.1992. The Javanese home garden. *Journal of Farming Sytem Research Extension* **3**: 95-118.
- Sreshtha, P.K., Gautham, R., Rana, R.B. and Sthapit, B.R.2004. Homegardens and Agrobiodiversity. In: Eyzaguirre, P.B. and Linarres, O.F. (Eds.).Managing diversity in various ecosystems: Home gardens of Nepal. Smithsonian Book, Washington. 135 p.
- Streitwolf, E. R., Boller, T., Wiemken, A. and Sanders, I.R.1997. Clonal growth traits of two *Prunella* species are determined by co-occurring arbuscular mycorrhizal fungi from a calcareous grassland. *Journal of Ecology* **85**:181–191.
- Warcup, J.H. 1951. The ecology of soil fungi. *Transactions of the British Mycological Society* **34**:376–399.
- Wieland, G., Neumann, R. and Backhaus, H.2001. Variation of microbial communities in soil, rhizosphere, and rhizoplane in response to crop species, soil type, and crop development. *Applied Environmental Microbiology* **67**:5849–5854.