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DIVERSITY OF PHYLLOPLANE MYCOBIOTA OF SOME ROADSIDE AND GARDEN PLANTS OF KARACHI: ALPHA, BETA AND GAMMA DIVERSITY

Faisal Hussain[1], S. Shahid Shaukat[2], Zamarrud Fatima Tajuddin[3], Moazzam A. Khan[2] and Abid Raza[4]

[1]*Department of Agriculture and Agribusiness Management, University of Karachi, Karachi-75270*
[2]*Department of Environmental Studies, University of Karachi, Karachi-75270, Pakistan*
[3]*Department of Botany, Karakoram International University, Gilgit, Pakistan*
[4]*Department of Environmental Science, Federal Urdu University, Karachi, Pakistan*

ABSTRACT

This investigation determines the species composition and diversity of phylloplane mycobiota of twenty different plant species, mostly trees, growing on roadsides in Karachi city or gardens (particularly Karachi University nursery and various Departmental gardens on the campus). A total of twenty- two microfungal species and 12 genera were recorded. The highest number of microfungal species (7) and genera (5) were recorded from *Eucalyptus camaldulensis* and lowest number of species (2) and genera (2) were observed on the phylloplane of drumstick tree (*Moringa oleifera*). Species diversity and its components, i.e., species richness and equitability, for the fungal communities were ascertained. The diversity analysis disclosed that the microfungal general diversity (H) was high for neem or Indian Lilac (*Azadirachta indica*), curry leaf (*Murraya koenigii*) and Eucalyptus (*Eucalyptus camaldulensis*) while it was low for Indian rosewood (*Dalbergia sissoo*), golden-shower tree (*Cassia fistula*) and drumstick tree (*Moringa oleifera*). On the other hand, equitability (J) was higher for neem (*Azadirachta indica*), Buttonwood (*Conocarpus erectus*), oleander (*Nerium oleander*) and Ashok tree (*Polyalthia longifolia*) and low for Indian rosewood (*Dalbergia sissoo*), wild almond (*Terminalia catappa*) and Golden-shower tree (*Cassia fistula*). The microfungal assemblages were generally dominated by the genus *Aspergillus* (*A. niger*, *A. flavus*, *A. terreus*, *A. fumigatus*). Additionally, *Nigrospora sphaerica* was also abundant. High qualitative similarities of phylloplane microfungal assemblages were demonstrated between the 20 plant species. Alpha, beta and gamma diversity of the phylloplane mycobiota was determined. Alpha diversity was low ($\bar{\alpha}$ = 4.35) while average beta diversity was slightly higher than alpha diversity ($\bar{\beta}$ =6.26). Similarity of mycobiota associated with phylloplane of various plant species was examined and average similarity based on Sorensen's index was found to be 23.80 percent. The similarity was zero for 101 pairs of assemblages. The phylloplane fungal assemblage showed a great deal of correspondence with the airspora of the area.

Key-words: Biodiversity, phylloplane mycobiota, garden plants.

INTRODUCTION

The shoot surfaces of plants such as those of leaves and stem together known as phyllosphere or phylloplane, provide complex natural microhabitats that are characterized by the occurrence of heterogeneous populations of micro-organisms including mycelial fungi, bacteria, yeasts , actinimycetes and algae (Andrew and Harris, 2000; Lindow and Brandl, 2003; Levetin and Dorsey, 2006; Newton *et al*., 2010). Functionally, the phylloplane biota comprises of pathogens, saprobes and epiphytes (Newton *et al*., 2010). Phylloplane mycobiota has often been categorized into two main groups (Lee and Hyde, 2002) known as residents and casuals (Norse, 1972). Whereas resident fungi can reproduce and grow on surface of healthy leaves without exerting much effect on the host plant, the casuals land on the phylloplane but they do not grow because of unfavourable conditions and competition (Leben, 1965; Hudson, 1992). Phylloplane microhabitat is characterized as a harsh and unfriendly environment since a number of factors including unavailability of water; insolation, nutrient regime and exposure to pollutants alter periodically as well as seasonally (Breeze and Dix, 1981; Lindow and Leveau, 2002; Joshi, 2008). The phylloplane microorganisms may play a vital role with regard to the host plant by acting as pathogens, natural antagonists to several detrimental organisms or serving as a source of plant growth stimulators (Blakeman, 1991; Andrews, 1992; Braga *et al.*, 2009). Potentially, a great number of natural antagonists of some significant plant pathogens can colonize on leaf surfaces which would lead to considerable economic benefit in the biocontrol of plant pathogens (Dighton, 2003; Yadav *et al*., 2011; Shamsi *et al*., 2012). Numerous studies have been conducted on the phylloplane microbial assemblages of different plant species (Lindsey and Pugh, 1976; Breeze and Dix, 1981; Mishra and Dickinson, 1981; De Jager *et al*., 2001; Andrews *et al*., 2002; Bakker *et al*., 2002; Osono, 2002; Osono *et al*., 2004; Kishore *et al*., 2005; De Costa *et al*., 2006; Levetin and Dorsey, 2006). Nicholson (1972) noticed that the

microorganisms prevailing on the leaf surfaces also commonly occur either in the soil or air. Thus it was doubtful whether these microbiota are merely casual contaminants (constituted by random mingling of species) and do not constitute an organized community (*sensu* Sugihara, 1980; Anderson and Calmay, 2004; Ferreira and Petrere, 2008; Meyer and Leveau, 2012). With the help of suitable experiments, Nicholson (1972) demonstrated that the populations of microorganisms do interact, grow and multiply on the phylloplane and eventually give rise to organized communities. The interaction of microbial populations, in particular, plays an eminent role in determining the structure and composition of the phylloplane microbial community. The density and diversity of the microbial populations change with time depending on host plant species, its growth stage, growing season and changes in physico-chemical characteristics of the leaf surfaces (Hirano and Upper, 2000; Mercier and Lindow, 2000). The interaction of microbial populations, particularly competition, which is often strong, plays a prominent role in determining the structure and composition of the phylloplane microbial community. The competitive abilities of microbial populations comprising the communities on the leaf surfaces can be altered by various inhibitory agents such as heavy metal concentrations present in the leaves or as a result of chronic exposure to gases like SO_2 and O_3 (Smith, 1977; Fenn *et al*., 1989). The concept of diversity is of paramount importance with regard to the structure of communities (cf. Ricklefs and Schluter, 1994; Magurran, 2004). The diversity of phylloplane mycobiota or miroflora has been investigated less frequently (Thomas and Shattock, 1986; Joshi, 2008; Shaukat *et al.*, 2013, 2014). Taxonomic diversity of an area or landscape (ecosystem) can be divided into alpha, beta and gamma diversity (see Whittaker, 1972; Brown and Gibson, 1983; Jost, 2006). Alpha diversity is the species richness of a single locality or area within the total 'landscape' Beta diversity is the species turnover rate or change in diversity along gradients, alternatively it can be defined as taxonomic differentiation of biota between sites or assemblages. Gamma diversity is regarded as the total diversity of the 'landscape'; alpha and beta diversity are regarded as the components of gamma diversity (Veech and Crist, 2010) and these can be measured by several indices (Magurran and McGill, 2011). The literature on the diversity of phylloplane mycobiota where diversity is quantitatively determined is scarce (Thomas and Shattock, 1986; Stanwood, 2009; Shaukat *et al*., 2013, 2014) while no report exists where phylloplane diversity is dealt with in terms of alpha, beta and gamma diversity.

The study was undertaken with the following objectives: 1) to assess the abundance and composition of phylloplane fungi of twenty different plant species (see Materials and Methods), 2) to quantify the similarity of phylloplane microfungal assemblages of the selected species growing at roadsides in the city and gardens located at Karachi University campus, 3) to measure the species diversity and its components (species richness and equitability) for the fungal assemblages under investigation, 4) to partition the total species richness (i.e., microfungal species occurring in the pylloplane assemblages of all plants examined) i.e., gamma diversity into alpha and beta components.

MATERIALS AND METHODS

Sampling:

Sampling was performed during December 2014 to March 2015. Five different localities were deterministically chosen, Departmental gardens and University of Karachi Nursery (where a large number of trees are growing) were sampled at Karachi University campus. Well-developed departmental gardens were selected to collect the leaves of different species. Following 20 plant species were selected for the study including *Azadirachta indica* (Adr.) Juss., *Guaiacum officinale* Linn., *Murraya koenigii* (Linn.) Spreng., *Dalbergia sissoo* Roxb., *Eucalyptus camaldulensis* Dehnh., *Ficus glomerata* Roxb., *Ficus benghalensis* L., *Citrus limon* (Linn.) Burm. f., *Ficus religiosa* L., *Salvadora persica* Linn., *Nerium oleander* Linn., *Conocarpus erectus* L., *Polyalthia longifolia* (Sonnerat) Thwait., *Cassia fistula* Linn., *Mangifera indica* L., *Moringa oleifera* Lam., *Tamarindus indica* Linn., *Parkinsonia aculeata* L., *Terminalia catappa* L. and *Mimusops elengi* L.

Leaves were collected from 0.8.0 to 1.5 m above ground and in natural condition, only photosynthetically active (non-senescent) leaves were sampled. From each site 5 leaves of each plant species were collected from 3 randomly chosen plants. Any disturbance of the experimental leaves was avoided by cutting the petiole and adjacent branches, the collected leaves were immediately brought to laboratory in sterile polythene bags. The assay of mycobiota was carried out within 24h of sampling.

Fungal cultures and assessment of mycobiota:

For each leaf four 1 cm^2 areas were cut with a sterile stainless steel template with 1 cm^2 opening to ensure consistent leaf sample area and care was taken to avoid the central midrib of the leaf. The four leaf sections were rinsed together in 2 ml sterile distilled water by vortexing for 1 minute (Levetin and Dorsey, 2006). A 0.5 ml aliquot of the suspension was plated onto Czapex Dox Agar (CDA) medium, in 9 cm diameter sterile glass Petri plate,

supplemented with Penicillin and streptomycin sulphate. After incubation at 28° C, the plates were examined for number of fungal colonies, and then observed under a microscope. Most isolates were obtained after a few days of incubation (generally 5-6 days), but plates were checked over several weeks to allow isolation of slow growing fungi. Each colony was assumed to have originated from a unit propagule. Developing fungal colonies were sub-cultured into pure isolates and identified by their microscopic morphology and colony characteristics using standard mycological literature (Thom and Rapper, 1945; Booth, 1971; Domsch *et al*., 1980; Barnett and Hunter, 1998; Ellis and Ellis, 2009). Results were expressed as colony forming units (CFUs)/cm^2 of leaf area. Four replicates were kept for each of the plant species sampled. A single-factor analysis of variance (ANOVA) was performed for the abundant fungal species separately, This was followed by Fisher's least significant difference (LSD) test and Duncan's multiple range test (Zar, 2009). The program for analysis of variance (ANOVA) together with the post-hoc tests was developed by one of the author (S. Shahid Shaukat) in C++ and FORTRAN.

Measurement of diversity and similarity

Diversity indices

A host of diversity indices have been proposed to measure species diversity (Magurran, 2004). Indices of diversity provide a useful means for quantifying community diversity and have been instrumental in revealing the microorganism diversity and community structure such as that associated with the phylloplane (Thomas and Shattock, 1986; Natsch *et al*., 1997; Joshi, 2008; Shaukat *et al*., 2013, 1014). A wide variety of diversity indices have been employed to compare the phylloplane mycobiota inhabiting plant species collected from different sites. Various diversity measures estimate different aspect of community structure. The general species diversity of the fungal communities was measured by the popular Shannon–Wiener information theory function:

$$H = -\sum P_i \log P_i \qquad i=1\ldots. S$$

Where H is the general species diversity and P_i the proportion of total number of CFUs/cm^2 for fungal species belonging to the i*th* species and S equals the total number of species in the assemblage (Shannon and Weaver, 1963; Southwood and Henderson, 2000). The variance of general diversity Var (H) was calculated in accordance with Magurran (2004), as follows:

$$\text{Var}\,(H) = \sum P_i (\log P_i)^2 - (\sum P_i \log P_i)^2 / N + (S\text{-}1) / 2N^2 \quad i=1,\ldots,S$$

The general diversity incorporates two components of diversity: species richness, which expresses the number of species S as a function (ratio) of the total number of individuals N; and equitability that measures the evenness of allotment of individuals among the species (Magurran, 2004). The equitability component of diversity and its variance were measured in accordance with Pielou (1975):

$$J = H / H_{max} = H / \log S$$

The equitability index J is the ratio between observed H and maximal diversity $H_{max.}$: Variance of equitability was estimated as:

$$\text{Var}\,(J) =: (H) / (\log S)^2$$

Alpha, beta and gamma diversity:

Whittaker (1972) distinguished between alpha, beta and gamma diversity. Alpha diversity refers to local diversity. It is estimated either as species richness or using one of the common diversity indices such as Shannon, Simpson and McIntosh index (Shaukat *et al*., 1981). Beta diversity is the spatial differentiation or it may be defined as the species turn over rate among sites or along gradients in a given region (Legendre, 2014) A variety of measures have been proposed to estimate beta diversity (Koleff *et al*., 2003; Jost, 2006; Graham and Fine, 2008; Magurran and McGill, 2011). On the other hand, gamma diversity is the measure for regional diversity or total species diversity of a region, estimated by pooling the alpha diversity of all sites within a region. We used the simplest measure of species richness, i.e., the number of species (S) to ascertain the α (alpha) diversity. Beta diversity can be calculated directly or indirectly using various measures. Beta (β) diversity was calculated as follows:

$$\beta = (S_1 - C) + (S_2 - C)$$

Where S_1 equals the number of species recorded in the first community, S_2 equals the number of species recorded in the second community and C the number of species common to both communities. Beta diversity was computed between each of the fungal assemblages thereby obtaining a matrix of beta diversity.

Gamma diversity was set equal to total number of fungal species recorded in the overall phylloplane survey. Diversity partitioning can be achieved either additively or multiplicatively (Jost, 2006). We quantified α, β and γ-diversity at a small scale i.e., at the level of phylloplane of plants. However, these diversity concepts are equally applicable to small scale habitats (microhabitats).The programs DIVER and ABGDIV were developed for the computation of diversity by one of the author (S. Shahid Shaukat) in C++ and FORTRAN languages (cf. Shaukat and Siddiqui, 2005) and GWBASIC (Ahmed and Shaukat, 2012). These computer programs are available from one of us (S.S.S.) at a nominal cost.

Measurement of Similarity:

Similarity between fungal assemblages was computed qualitatively using Sorensen's similarity coefficient (Kenkel and Booth, 1992) as follows:

$$\mathbf{C_{jk} = [2a / (A + B)] X 100}$$

Where S_{jk} is the similarity between sites j and k , ' a' is the usual notation of the 2 X 2 contingency table indicating species common to assemblages j and k while A and B are the total number of fungal species in assemblages j and k. The program SIMIL for computation of similarity matrix using eleven different similarity indices was developed by S.S.S. in GWBASIC, FORTRAN and C++ (cf. Shaukat and Siddiqui, 2005; Ahmed and Shaukat, 2012).

RESULTS AND DISCUSSION

The composition of phylloplane fungal assemblages and the abundances in terms of average CFU/cm^2 for the twenty plant species are presented in Table 1. The number of fungal species varied with the assemblage. On an overall basis twenty- two microfungal species and 12 genera were recorded. The highest numbers of microfungal species (7) and genera (5) were recorded from *Eucalyptus camaldulensis* and lowest number of species (2) and genera (2) were observed on the phylloplane of *Moringa.*

Generally, the phylloplane mycobiota was predominated by the genus *Aspergillus.* Particularly, *A. flavus*, *A. fumigatus* and *A. niger* that were found to be the most abundant species. ANOVA for combined *Aspergillus* spp., showed significant difference in abundance (CFU's) with respect to plant species examined ($P<0.05$) The abundance of *Aspergillus flavus* also exhibited significant difference (using single factor ANOVA) with regard to phylloplane assemblages associated with different species ($P<0.05$). However, *A. niger* did not disclose significant difference with regard to plant species. Additionally, *Nigrospora sphaerica* also occurred on the phylloplane of 50 percent of the plant species tested. ANOVA for *N. spherica* resulted in a significant difference in abundance (CFU's) between species ($P<0.01$). The present study accords well with the findings of Mehdi and Saifullah (1992) who also reported high abundance of *Aspergillus* species (i.e., *A. niger* and *A. flavus*) on the phylloplane of grey mangrove *Avicennia marina* growing at Clifton and Korangi Creek in Karachi area. Naikwade *et al*. (2012) recorded 9 different species of *Aspergillus* from the phylloplane of a mangrove species *Ceriops tagal*; they also found *Alternaria alternata* on the phylloplane. Shaukat *et al*. (2013) working with the phylloplane mycobiota of *Avicennia marina* and *Rhizophora mucronata*, also observed dominance of *Aspergillus* species. El-Said (2001) recorded nine species of *Aspergillus* from the phylloplane of banana in Egypt. The phylloplane microfungal assemblages for the plants selected from Karachi University nursery showed relatively lower number of genera as well as species. Kuthubutheen (1981) in an extensive study involving 9 mangrove species, reported various fungal species included in the genera like *Aspergillus*, *Cladosporium*, *Curvularia*, *Fusarium*, *Penicillium* and *Trichoderma* which have been reported in the current study as well. The fungal assemblages developed in the cultures included many pioneer species that colonize the phylloplanes in early succession and subsequently their density increases substantially (Dix and Webster, 1995). The pioneer species tend to be fast growing, short-lived, and capable of rapid and widespread dispersal (Luczkovich and Knowles, 2000). Thus profusely sporulating fungi like *Aspergillus*, *Penicillium* and *Cladosporium* were predominant. *Fusarium* species found (*F. oxysporum*, *F. solani* and *F. moniliformis*) were presumably non-pathogenic and occurred simply as epiphytes as no visible pathogenecity symptoms were noticed (Luczkovich and Knowles, 2000). These results correspond well with those of earlier workers with regard to phylloplane mycobiota of some plant species (Luczkovich and Knowles, 2000; El-Said, 2001). The diversity analysis disclosed that the microfungal general diversity (H) was high for neem (*Azadirachta indica*), curry leaf (*Murraya koenigii*) and

Eucalyptus (*Eucalyptus camaldulensis*) while it was low for Indian rosewood *Dalbergia sissoo*, golden-shower tree *Cassia fistula* and drumstick tree (*Moringa oleifera* (Table 2).

Table 1. Fungal species density cfu/cm^2 of leaf surface are for twenty plant species samples in the survey.

Plants	*A. alternat*	*A. flavu*	*A. fumigate*	*A. niger*	*A. terreu*	*B. cinerari*	*C. cladosporioide*	*C. fusiform*	*C. herbaru*	*C. brachyspor*	*C. clavat*
1. Azadirachta indic	0	1	0	0.66	0	0	1	0	0	0	0
2. Guaiacum officin	0	0	0	3.66	1	0	0	1	0	0	0
3. Murraya koenigi	0	0.66	0	1	1.66	0	0	1	0	0	0
4. Dalbergia sissoo	0	1.66	0	9.33	2.66	0	0	0	0	0	0
5. Eucalyptus cama	1	1	0.66	1	0	0	0	0	1.33	0	0
6. Ficus glomerata	0	0	0	4.66	0	0	0	0	2.33	0	0
7. Ficus benghalen	0	0	1	3	0	0	2.33	0	0	0	0
8. Citrus limon	2.66	2.33	1	1	0	0	2.66	0	0	0	0
9. Ficus religiosa	0	0	0	0	0	0	0	4.33	0	1.33	0
10. Salvadora persi	0	1.33	0.66	0.66	0	0	0	0	2	0	0
11. Nerium oleande	0	0	0	0	0	0	1	0	0	0	1.33
12.Conocarpus erec	0	2.66	2	0	0	0	0	0	0	0	0
13. Polyalthia long	2	0	0	0	0	0	0	0	0	2.33	0
14. Cassia fistula	0	0	0	3.33	0	0	1.33	0.66	0	0	0
15. Mangifera indic	1.66	0	0	0	0	0	0	0	0	0	0
16. Moringa oleifer	0	0	2.66	0	0	0	0	0	0	0	0
17.Tamarindus indi	0	3.33	0	3.33	0	0	0	0	0	0	0
18.Parkinsonia acu	0	0	0	0	2	0	1.66	0	0	0	0
19. Terminalia cata	0	0	0	0	1.66	0	0	0	0	1.66	0
20. Mimusops eleng	2.33	0	0	0	0	2	0	0	0	0	0

Plants	*C. lunat*	*D. hawaiiensi*	*D. biseptat*	*F. moniliform*	*F. semitectu*	*N. sphaeric*	*P. oxalicu*	*P. commun*	*Rhizopus s*	*T. harzianu*	*Verticillium s*
1	0	2	0	1.33	0	1.33	0	0	0	0	0
2	0	1.33	0	1	0	0.66	0	0	0	0	0.66
3	0	0	0	2.66	0	1	0	0	0	0	0
4	0	0	0	0	0	0	0	0	0	0	1
5	0	1.33	0	0	0	4	0	0	0	0	0
6	0	1	1.33	0	0	0	0	0	0	0	0
7	1	0	0	0	0	2.66	0	0	0	0	0
8	0	0	0	0	0	0	0	0	0	0	0
9	0	0	0	0	0	4.33	0	0	0	0	0
10	0	0	0	0	0	4	0	0	0	0	0
11	0	1.66	0	0	0	0	0	0	0	1.66	0
12	0	0	0	0	0	0	2	0	0	1.66	0
13	0	0	0	0	2.33	0	0	0	0	0	0
14	0	0	0	0	0	0	0	0	0	0	0
15	0	0	0	0	0	0	1.33	0	1.33	0	0
16	0	0	0	0	0	0	0	1.66	0	0	0
17	0	0	0	0	2	0	0	0	0	0	0
18	0	0	0	0	0	4	0	0	0	2	0
19	0	0	0	0	0	4.66	0	0	0	0.33	0
20	0	0	0	0	0	3	0	0	0	0	0

Table 2. Diversity measure for fungi occurring on different garden and road side plants of Karachi. Species diversity=H, equitability= J, variance of H=Var(H), variance of J= Var(J), Species richness= d_1, dominance= D, # of species= α.

Plant Species	**H**	**Var (H)**	**J**	**Var (J)**	**d_1**	**D**	**α**
1. Neem	1.735	0.066	0.968	0.0208	2.267	0.05	6
2. Lignum	1.596	0.919	0.890	0.028	2.121	0.150	6
3. Curry leaf	1.679	0.083	0.937	0.026	2.267	0.029	6
4. Indian rosewood	0.835	0.041	0.760	0.034	0.832	0.482	3
5. Eucalyptus	1.799	0.073	0.899	0.019	2.213	0.131	7
6. Dumar	1.236	0.049	0.891	0.025	1.333	0.252	4
7. Banyan	1.513	0.042	0.940	0.016	1.666	0.150	5
8. Lemon	1.523	0.041	0.946	0.015	1.666	0.142	5
9. Pepal	0.993	0.030	0.904	0.025	1	0.325	3
10. Salvadora	1.375	0.084	0.854	0.032	1.767	0.211	5
11. Oleander	1.366	0.067	0.986	0.035	1.788	0.099	4
12. Buttonwood	1.371	0.022	0.989	0.014	1.414	0.156	4
13. Ashok	1.096	0.022	0.997	0.023	1.224	0.217	3
14. Golden shower	0.898	0.110	0.818	0.091	1.341	0.346	3
1**5.** Mango	1.092	0.065	0.994	0.054	1.5	0.137	3
16. Drumstick	0.666	0.04	0.960	0.092	1	0.384	2
17. Tamarind	1.07	0.021	0.977	0.017	1.060	0.264	3
18. Parkinsonia	1.319	0.034	0.952	0.017	1.333	0.204	4
19. Wild almond	1.095	0.077	0.790	0.040	1.414	0.313	4
20. Memosops	1.084	0.022	0.987	0.020	1.133	0.239	3

Table 3. Matrix of beta diversity between the phylloplane mycobiota assemblages of twenty plant species. Plant species 1-20 are the same as given in Table 1.

	1	**2**	**3**	**4**	**5**	**6**	**7**	**8**	**9**	**10**	**11**	**12**	**13**	**14**	**15**	**16**	**17**	**18**	**19**	**20**
1																				
2	5																			
3	4	3																		
4	6	5	4																	
5	5	8	7	7																
6	6	7	8	6	5															
7	5	8	7	7	6	7														
8	5	10	7	5	4	7	4													
9	7	6	5	7	8	7	6	8												
10	5	8	5	5	2	5	4	4	6											
11	6	9	10	8	9	6	7	7	7	9										
12	8	11	8	6	7	8	7	5	7	5	6									
13	9	10	9	7	8	7	8	6	4	8	7	7								
14	5	6	5	5	8	5	4	4	4	6	5	7	6							
15	9	10	9	7	8	7	8	6	6	8	7	5	4	6						
16	8	9	8	6	7	6	5	5	5	5	6	4	5	5	5					
17	5	8	5	3	6	5	6	4	6	4	7	5	4	4	6	5				
18	6	7	6	6	9	8	5	7	5	7	4	6	7	5	7	6	7			
19	8	7	6	6	9	8	7	9	3	7	6	6	5	7	7	6	7	2		
20	7	8	7	7	6	7	6	6	4	6	7	7	4	6	4	5	6	5	5	

On the other hand, equitability (J) was higher for neem (*Azadirachta indica*), green buttonwood (*Conocarpus erectus*), oleander (*Nerium oleander*) and Ashok tree (*Polyalthia longifolia*) and low for Indian rosewood (*Dalbergia sissoo*), wild almond (*Terminalia catappa*) and Golden- shower tree (*Cassia fistula*). It has been shown that size of resource unit affects the number of species that can co-occur (Sanders and Anderson, 1979;

Barlocher and Schweizer, 1983). Variances of diversity and equitability were not unexpectedly consistently low for almost all species as has previously been observed by Shaukat *et al*. (2013). Species richness (d_1) was high for neem (*Azadirachta indica*), curry leaf (*Murraya koenigii*) and *Eucalyptus* sp. and low for pepal tree (*Ficus religiosa*) and drumstick tree (*Moringa oleifera*). Species diversity may be important because of its possible role on the establishment and coexistence of species (Nicholson, 1972) though in some model systems it is found to play hardly any role on these processes (Stohr and Dighton, 2004). Dominance concentration (D) was found to vary inversely with the general diversity (H) which is in agreement with the results of Shaukat and Khan (1979) and Shaukat *et al*. (2013). Following Whittaker (1972) and Jost (2006) we also distinguished between alpha (α), beta (β) and gamma (γ) diversity. Gamma diversity of an area can be partitioned into alpha and beta diversity. The alpha diversity for each of the assemblage is given in last column of Table 2. The average alpha diversity was $\bar{\alpha}$ = 4.35. The matrix of beta diversity is presented in Table 3.

Table. 4. Similarity in the microfungal assemblages of the phylloplanes of 20 plant species. For the codes of plant species numbered 1-20 refer to Table 1.

	1	**2**	**3**	**4**	**5**	**6**	**7**	**8**	**9**	**10**	**11**	**12**	**13**	**14**	**15**	**16**	**17**	**18**	**19**	**20**
1																				
2	61.53																			
3	66.66	76.92																		
4	40	54.54	60.00																	
5	61.53	42.85	46.15	36.36																
6	40	36.36	20	25	54.54															
7	59.54	33.33	36.36	22.22	50	22.22														
8	54.54	16.66	36.36	44.44	66.66	22.22	60.00													
9	22.22	40	44.40	0	20	0	25	0												
10	54.51	33.33	54.54	44.44	83.33	44.44	60.00	60.00	25											
11	40	18.18	0	0	18.18	25	22.22	22.22	0	0										
12	20	0	20	25	36.36	0	22.22	44.44	0	44.44	25									
13	0	0	0	0	20	0	0	25	33.33	0	0	0								
14	44.44	40	44.44	28.57	20	28.57	50	50	33.333	25	28.57	0	0							
15	0	0	0	0	20	0	0	25	0	0	0	28.57	33.33	0						
16	0	0	0	0	22.22	0	28.57	28.57	0	28.57	0	33.33	0	0	0					
17	44.44	20	44.44	57.14	40	28.57	25	50	0	50	0	28.57	33.33	33.33	0	0				
18	40	36.36	40	25	18.18	0	44.44	22.22	28.57	22.22	50	25	0	28.57	0	0	0			
19	20	36.36	40	25	18.18	0	22.22	0	57.14	22.22	25	25	28.57	0	0	0	0	75		
20	22.22	20	22.22	0	40	0	25	25	33.33	25	0	0	33.33	0	33.33	0	0	28.57	28.57	

The average beta diversity $\bar{\beta}$ was found to be 6.26, while gamma (γ) diversity was 22. Despite low alpha diversity, beta diversity is relatively high accounting for greater gamma diversity. Likewise, Zhang *et al*. (2014) also found greater contribution of beta diversity to the gamma diversity in a regional vegetation of Mongolia. The qualitative similarity in the phylloplane mycobiota assemblages of the twenty plant species is represented in Table 4.

Computation of community similarity matrix disclosed a high range of similarity 0 to 83.32 percent. The highest similarity was found between the phylloplane mycobiota of *Eucalyptus camaldulensis* and *Salvadora persica* (83.32%). Whereas, a large number of pairs of assemblages (101 out of 190 comparisons) depicted zero percent similarity. The average similarity between phylloplane microfungal assemblages was 23.80 percent. A variety of environmental factors are known to affect fungal diversity (Stanwood, 2009). These factors include temperature, humidity, rainfall, dew, wind velocity and direction. In addition, intrinsic factors of the leaf also play an important role in fungal composition and diversity (Lindow and Brandl, 2003). Interestingly, the dominant phylloplane fungal species belonging to *Aspergillus*, *Fusarium*, *Cladosporium*, etc also occur as the dominant species of airborne mycospora (Afzal *et al*., 2005; Rao *et al*., 2009). Filamentous fungi are often regarded as transient inhabitants of phylloplane and colonize the phylloplane predominately as spore (Andrews and Harris, 2000; Lindow and Brandl, 2003). Based on the degree of correspondence between phylloplane and airborne mycobiota, it seems that the phylloplane microfungal assemblages contribute a great deal to the airborne mycobiota. In this context, Levetin and Dorsey (2006) have emphasized that the leaf-surface fungi are the major contributors to the airborne mycospora. Those taxa with an airborne dispersal are the major components in this respect. Based on the estimated concentrations of the two tree species *Ulmus* and *Quercus*, Levetin and Dorsey (2006) calculated that 19% of the airspora was contributed by the phylloplane fungi. However, the airspora is also contributed by the soil surface and the decaying or other organic waste lying on the land surface through winds and gale. Thus, further studies are needed to determine the role of phylloplane mycobiota to the airspora. Fungi are essential for nutrient mobilization, storage and release during the process of decomposition of plant parts, e.g., leaves in an ecosystem (Kjøller and Struwe, 2002). The examination of mycobiota associated with leaves would help to gain greater understanding of the role of fungi in litter decomposition and eventually nutrient cycling in the terrestrial ecosystems.

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