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Biodiversity of filamentous and yeast fungi of phyllosphere and phylloplane of citrus and grapevine plantations in Assiut area, Egypt

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Abstract: The present study is an extensive survey of mycobiota from phyllosphere and phylloplane of citrus and grapevine plantations in Sahel-Saleem City, Assiut Governorate, Egypt. The study was carried out during the period from April 2008 to February 2009. Identification of filamentous and yeast fungi was conducted using morphological and biochemical characteristics, and in many of them identification was confirmed using rDNA sequencing. A total of 186 species and 2 varieties were recovered from both phyllospheres (180 species and 2 varieties of filamentous fungi and yeast fungi) and phylloplanes (112 + 1) of both plantations. Phyllospheres of both plants registered higher number of fungal taxa than those of phylloplanes on both DYM and DRBC media. Also citrus phyllospheres registered higher number of genera and species and propagules than those of grapevine. The same trend was observed in phylloplanes of both plants. The peak of total phyllosphere and phylloplane fungi was regularly recorded in February (permanent mature leaves) in citrus and December (senescent leaves) in grapevine on both media. On the other hand, the trough of phyllosphere fungi of both plants was regularly recorded in June (mature and young leaves, respectively) on both media, while the trough of phylloplane fungi of citrus was observed in June and August on DYM and DRBC, respectively, and in June (young leaves) in grapevine on both media. Cladosporium, Alternaria, Aspergillus and yeasts were recovered in high frequency from both phyllospheres and phylloplanes of both plants on both isolation media. However, the counts of Cladosporium predominated on both phyllospheres followed by phylloplane of citrus, and those of Alternaria and yeasts were relatively higher on phyllosphere and phylloplane of grape on both media than those of citrus. Aspergillus percentage propagules were higher in phylloplanes of both plants than those of both phyllospheres. Phoma was generally of high frequency except in grape on DRBC where it was moderately encountered. The incidence of Quamballaria was variable being high on citrus phyllosphere and moderate, low or rare in the others cases. Many infrequent fungi were recorded from one or both plantations. The present study reveals that the dematiaceous fungi outnumbered the hyaline ones in phyllosphere and phylloplane. Basidiomyceteous yeasts were dominant over ascomyceteous yeasts in these environments.

Keywords: fungi, yeasts, citrus, grapevine, phyllosphere, phylloplane, Egypt.

Introduction

Plant tissues and surfaces are colonized by microbial communities consisting of filamentous fungi, yeasts, bacteria, actinomycetes, and algae (Last and Warren 1972, Dickinson 1976). The phyllosphere is the living leaf as a whole and includes the surface (phylloplane) and internal tissues colonized by a variety of epiphytic and endophytic microorganisms respectively, thereby occupying two distinct habitats on the leaf (Andrews 1996). The interest shown in the last few years in the study of phyllosphere microbes is due principally to their interactions with plants, herbivores and pathogens on living leaves which may be involved in the plant immunity system, reabsorption of organic and mineral matters from leachates, redistribution of nutrients prior to leaf fall and participation in the primary degradation of plant tissues (Lindow and Brandl 2003, Osono 2006, Võríšková and Baldrian, 2013). However, the phyllosphere, which lato sensu consists of the aerial parts of plants, and therefore primarily, of the set of photosynthetic leaves, is one of the most prevalent microbial habitats on earth. Phyllosphere microbiota are related to original and specific processes at the interface between

plants, microorganisms and the atmosphere (Bringel and Couée, 2015).

Another aspect of colonization ecology of phylloplane and/or phyllosphere fungi principally relates to the prevailing microenvironmental conditions on the leaf surfaces and their physical, chemical and phenological properties which affect the fungal establishment thereon (Andrews and Harris 2000). The phyllophane (surface of leaves) presents many peculiar features for microbial life (Rastogi et al. 2013; Müller and Ruppel, 2014). Leaf surfaces are by themselves a complex architecture of microenvironments showing bidimensionally and tridimensionally heterogeneous structures. The characteristics of upper or lower phylloplane (Reisberg et al. 2013) affect the interactions between epiphytic microorganisms, which live on plant surfaces, in particular by modulating the access to nutrients from leaf tissues (Bulgarelli et al. 2013), by providing more or less protection from incoming sunlight (Atamna-Ismaeel et al. 2012), or by presenting gateways for penetration within the plant endosphere (Schreiber et al. 2004).

The epiphytic (non-phytopathogenic) microbial communities of leaves are very diverse (Morris 2001, Lindow and Brandl 2003). Cuticle composition and topographic features (stomata, trichomes, veins, etc.) are also highly variable both within a leaf and among different plant species (Hallam and Juniper 1971) and may influence the composition and distribution of phylloplane communities (Kinkel 1997). Molecules leached from plant leaves include a variety of organic and inorganic compounds, such as sugars, organic acids, amino acids, methanol and various salts (Blakeman 1971, Morris 2001). The abundance of such nutrients varies with plant species, leaf age and growing conditions. Exogenous nutrient sources, such as aphid honeydew and pollen, have been associated with dramatic increases in the microbial carrying capacities of some leaves (Stadler and Muller 1996).

Cladosporium Alternaria alternata, cladosporioides, Fusarium oxysporum and Pestalotiopsis sp. were the dominant surface and interior colonizers of different trees (Alnus nepalensis, Castanopsis hystrix and Schima walichii) leaves. Alternaria raphani, Epicoccum purpurascens and Gliocladium roseum from Alnus nepalensis leaves and Scopulariopsis sp. and Trichoderma harzianum from Castanopsis hystrix were the species recovered specifically by washed disk method. Whereas, Gliocladium fimbriatum was isolated only from Schima walichii leaves as endophytic fungus in subtropical habitats of North Eastern India (Kayini and Pandey 2010).

Yeasts were isolated from leaf surfaces of five species of fruit trees located in southwest Slovakia. Fifteen yeast species were identified, but only three occurred regularly: Aureobasidium pullulans, Cryptococcus Metschnikowia pulcherrima. laurentii and Species such as Hanseniaspora uvarum, Pichia Rhodotorula anomala, glutinis and Saccharomyces cerevisiae were isolated in less frequently. Low incidence of R. glutinis was observed on leaves together with another two carotenoids-producing species (Rhodotorula mucilaginosa and R. minuta). The remaining five species: Candida tropicalis, Geotrichum candidum, Pseudozyma aphidis, P. fusiformata and Yarrowia lipolytica were isolated from the leaves in less frequently (Slavikova et al. 2009). In other study, the red yeast species Sporobolomyces roseus also belongs to the yeasts frequently occurring on leaf surfaces

(Phaff and Starmer 1987, Nakase 2000). Hanseniaspora sp., Pichia sp., Candida sp., Sporidiobolus sp., Meyerozyma sp., Symmetrospora sp., Rhodotorula sp., Starmerella sp. and Aureobasidium sp. were associated with leaves of Nanfeng mandarin (Citrus reticulata cv. Blanco) in China (Peng et al. 2018).

Phylloplane communities usually comprise deeply pigmented pink yeasts belonging to the genera Rhodotorula and Sporobolomyces and non-pigmented white Cryptococcus species (McCormack et al. 1994). Ascomyceteous yeasts are usually rare on the phylloplane but the species Debaryomyces hansenii was found with high frequency on plants from the Canary Islands (Middelhoven 1997), on sugarcane in Brazil (Azeredo et al. 1998) and on leaves of forest plants in Russia (Glushakova and Chernov 2004, Maksimova and Chernov 2004). The black yeast species of Aureobasidium are also found on the leaves of apple trees (Pennycook and Newhook 1981), leaves of Nanfeng mandarin (Citrus reticulata cv. Blanco) in China (Peng et al. 2018).

The present work was designed to investigate the diversity and seasonal fluctuations of filamentous and yeast fungi in the phyllosphere and phylloplane of two economically-important plants, citrus (orange) and grapevine (vitis) in Assiut area, Egypt.

Materials and Methods 1- Collection of samples

This study was carried out in Sahel-Saleem city at approximately 25 km south-east of Assiut city. Sampling was conducted bimonthly over one year from April 2008 - February 2009. Three different plantations of citrus in the suburbs of Sahel-Saleem city and three of grapevine in El-Khawaled village (about 6 Km to the east border of the river Nile), in the northeast of Sahel-Saleem city were selected.

A total of 33 leaf samples were collected from citrus (18) and grapevine plantations (15). Leaf samples were collected randomlly from different plants at each farm and put directly each into a clean plastic bag. Samples were brought into the laboratory and kept at 5°C till fungal analysis.

2- Isolation of phyllosphere fungi

Small pieces of leaves (approximately 1 cm²) were made using sterile scissors and 10 gm of each sample were placed in 250 ml sterile Erlenmeyer flask containing 90 ml sterile distilled water. Flasks were shaken on an orbital shaker for 15 minutes. Ten ml aliquots of the suspension were transferred into sterile Erlenmeyer flasks each containing 90 ml sterile distilled water, then were shaken for 5 minutes. One ml of the appropriate dilution was transferred into each sterile Petri-dish which was then poured with melted but cooled agar medium. Ten replicate plates were used for each sample (5 for each medium type).

3- Isolation of phylloplane fungi

The pieces of leaves after thorough shaking in a series of sterile distilled water were removed and dried using sterilized filter paper. Four pieces were placed on the surface of each agar plate. Five replicate plates were used for each type of medium and for each plant type.

The plates were incubated at 28°C for 1-2 weeks during which the developing fungi were counted and isolated for further identification and the number of colony forming units (CFU) was calculated. Isolates of different fungi were maintained on slants of yeast extract malt exract agar, YM (for yeasts), Czapek yeast agar, CYA (for filamentous fungi) and stored at 5°C till confirming the identification.

4- Media used for isolation of fungi

Two media were used for isolation. Yeast extract malt extract agar (YM, Wickerham 1951), modified by the addition of 1 ml/l of 2 mg of dichloran dissolved in 10 ml ethanol and designated as dichloran yeast extract malt extract agar, DYM (Moubasher et al. 2016), and dichloran rose bengal chloramphenicol agar, DRBC (King et al. 1979) were used.

5- Identification of filamentous fungi

The following references were used for the identification of fungal genera and species (purely morphologically, based on macroscopic and microscopic features): Raper and Fennell (1965), Rifai (1969), Ellis (1971), Pitt (1979), Sutton (1980), Sivanesan (1987), Moubasher (1993), Gams and Bissett (1998), De Hoog et al. (2000), Schroers (2001), Zare and Gams (2004), Leslie and Summerell (2006), Crous et al. (2007), Domsch et al. (2007), Simmons (2007), Seifert et al. (2011), Samson et al. (2011, 2014), Chen et al. (2016), Hubka et al. (2016).

6- Identification of yeasts

a- Morphological characters

the ability of yeasts to form pseudomycelium and true mycelium (Wickerham 1951) and to produce ascospores on three sporulation media (corn meal agar, potato glucose agar and yeast extract malt extract agar at 25°C) (Barnett et al. 2000) were tested.

b- Physiological characters

Fermentation of sugars and oxidative carbon utilization of compounds were performed according to Barnett et al. (2000). Assimilation of nine nitrogen compounds was also determined (Suh et al. 2008). Test for hydrolysis of urea, growth at high osmotic pressure, growth at different temperatures, growth in the presence of cycloheximide, diazonium blue B (DBB) test and production of extracellular starch-like compounds were

performed. Identification keys of Barnett et al. (2000) were followed to assign each isolate to its species level.

c- Molecular methods

Confirmation of identifications was carried out using the molecular techniques. The fungus was grown on CYA plates and incubated at 25° C for 7 days (for filamentous fungal isolates) and on YM plates and incubated at 25° C for 2 days (for yeast isolates). A small amount of fungal growth was scraped and suspended in 100 µl of distilled water and boiled at 100° C for 15 minutes. These preparaions were sent to SolGent Company, Daejeon, South Korea, for PCR and rDNA sequencing (refer to Moubasher et al. 2016). Contigs were created from the sequence data using CLCBio Main Workbench program. The sequence obtained from each isolate was further analyzed using BLAST from the National Center of Biotechnology Information (NCBI) website. Sequences obtained with those retrieved from GenBank database were subjected to Clustal W analysis using MegAlign (DNA Star) software version 5.05 for the phylogenetic analysis (Thompson et al. 1994). Sequence data were deposited in GenBank and accession numbers were given for them (Table 1).

Results and Disscusion

The present investigation focused on fungi on surfaces of citrus and grapevine leaves, in one year experiment, employing two media of isolation: yeast extract malt extract agar supplemented with dichloran (DYM) and dichloran rose-bengal chloramphenicol agar (DRBC). Identification of genera and species was performed using the morphological and microscopical characteristics in addition to the biochemical in case of yeasts. In suspected isolates, molecular techniques [Internal transcribed spacer (ITS) sequences of nuclear

ribosomal DNA were amplified using primers ITS1, ITS4] were also employed (Table 1).

The gross total counts of all fungi were much greater in citrus, than those in grapevine leaves, which were also richer in number of genera and species than those of grapevine. In terms of numbers, citrus phyllospheres registered higher number of genera and species (51 genera and 130 species + 1 varieties on DYM and DRBC) and propagules (1037584 and 1285916 CFU / all samples on both media, respectively) than those of grapevine (42 genera and 83 species + 1 variety) and propagules (320688 and 277244 CFU in all samples) and the same trend was observed in phylloplanes of both plants. Citrus plant has an advantage of being taller tree with dense evergreen foliage that may affect most environments around it more positively than grapevine. The number of genera and species was more on DRBC than that on DYM in both plantations.

A total of 180 species and 2 varieties belonging to 67 genera of filamentous and yeast fungi were recovered from the phyllosphere of both plants. Yeast fungi were represented by 14 genera and 23 species. A considerably higher number of genera and species (51 genera and 130 species + 1 varieties) were recovered from citrus phyllosphere compared with those recovered from grapevine (42 and 83+1 variety). Also, approximately 5-fold propagules were recovered from citrus phyllosphere (2312 colony forming units/mg fresh leaves in all samples on both isolation media) compared with those from grapevine (484) as shown in (Table 2).

The peaks of total fungal propagules were regularly recorded in February in citrus phyllosphere and in December in grapevine on both media, while their troughs were regularly recorded in June on both media in both plants (Figs 1-4).

The widest spectrum of fungal species in citrus phyllosphere was registered in August and October (43 species) on DYM and in October (38) on DRBC, and in grapevine phyllosphere in August (34 species on DYM and 37 + 1 variety on DRBC). The narrowest spectrum in citrus phyllosphere occurred in April (29 species + 1 variety on DYM) and in December (29 species on DRBC), and in grapevine in June (19 species on DYM) and in June and December (17 on DRBC) (Table 2).

A total of 113 species + 2 varieties belonging to 52 genera of filamentous and yeast fungi were recovered from the phylloplane of both citrus and grapevine. Yeast fungi were represented by 13 genera and 16 species. A higher number of genera and species (47 genera and 93 species + 1) were recovered from the phylloplane of citrus compared to those from grapevine (37 and 73 + 1). Also approximately 2-fold propagules were recovered from the citrus phylloplane (5275 colony forming units/360 fresh leaf pieces in all samples on both isolation media) compared with those isolated from grapevine phylloplane (2207 CFU/300 fresh leaf pieces) (Table 3). The peaks of total propagules of fungi were recorded in February from citrus phylloplane and in December (senescent leaf) from grapevine on both media, while their troughs were recorded in citrus plantations in June and August on DYM and DRBC respectively, and in June (young leaf) in grapevine on both media (Figs 5-8).

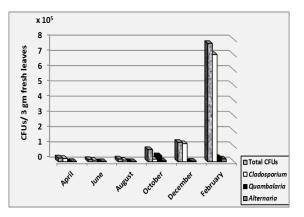


Figure 1: Bimonthly counts of common fungi in citrus phyllosphere on DYM, during the period from April 2008 to February 2009.

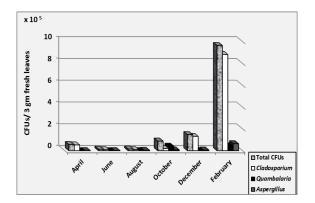


Figure 2: Bimonthly counts of common fungi in citrus phyllosphere on DRBC, during the period from April 2008 to February 2009.

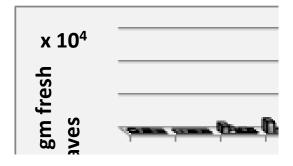


Figure 3: Bimonthly counts of common fungi in grapevine phyllosphere on DYM, during the period from April 2008 to December 2008.

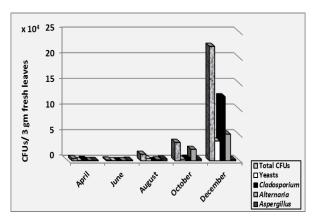


Figure 4: Bimonthly counts of common fungi in grapevine phyllosphere on DRBC during the period from April 2008 to December 2008

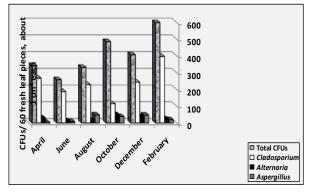


Figure 5: Bimonthly counts of common fungi in citrus phylloplane on DYM during the period from April 2008 to February 2009.

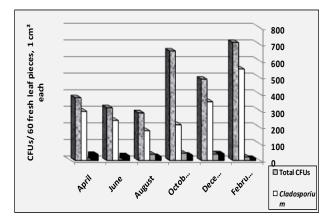


Figure 6: Bimonthly counts of common fungi in citrus phylloplane on DRBC, during the period from April 2008 to February 2009.

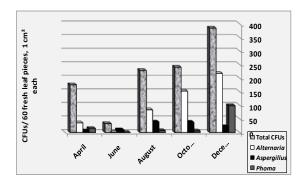


Figure 7: Bimonthly counts of common fungi in grapevine phylloplane on DYM, during the period from April 2008 to December 2008.

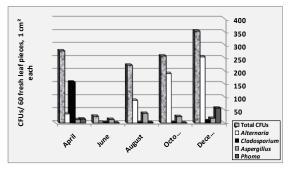


Figure 8: Bimonthly counts of common fungi in grapevine phylloplane on DRBC, during the period from April 2008 to December 2008.

The widest spectrum of species in citrus phylloplane was registered in August (31 speciess +1 variety on DYM and 35+1 on DRBC) as well as in grapevine phylloplane (26 species +1 variety on DYM and 28 species on DRBC), while the narrowest in citrus phylloplane in April (15 species and 18 on DYM and DRBC respectively), and in grapevine in April (19 species on DYM) and in June (13 on DRBC) (Table 3).

Cladosporium, Alternaria, Aspergillus and yeasts were recovered in high fequency in both phyllosphere and phylloplane of citrus and grapevine, while *Fusarium, Penicillium, Stemphylium* and *Phoma* in phyllolanes of both plants. Leaves (phyllosphere and phylloplane) of citrus were richer in *Cladosporium* and *Quamballaria* counts than those of grapevine, however grapevine phyllosphere was richer in *Alternaia* and yeasts, and phylloplane in *Alternaria, Aspergillus* and *Phoma*.

Citrus phyllosphere shared phyllosphere of grapevine in some highly frequent fungal genera on both media (*Cladosporium, Alternaria, Aspergillus, Fusarium, Penicillium, Pleospora*), or on one medium (*Cochliobolus* and *Phoma*) (Table 2).

The phylloplane of citrus shared the grapevine phylloplane in some highly encountered fungi on both media (*Alternaria, Aspergillus,* and *Phoma*) or on one medium (*Cladosporium, Penicillium* and *Stemphylium*) (Table 3).

Cladosporium was the most common genus in the phyllosphere and phylloplane of both plants, contributing higher percentage counts in citrus plantations than those in grapevine. *Cladosporium* gave its highest percentage counts in citrus phyllosphere (83.86 % - 85.99 % of total fungi), citrus phylloplane (59.17 % - 64.58 %). In this respect Moubasher (1995, 2010) adopted that fungal spores are dislodged from soil by air currents. A part of them remains suspended in air and the others alight or are sedimented on vegetation surface where a new substrate or niche is initiated. The conditions in this niche are substantially different from those in soil. Competition for the colonization of this substrate is less severe. Atmospheric conditions are more drastic, high light intensity, and deep fluctuations diurnal of temperature and humidity. Consequently, the mycobiota developing in this niche has a basically different pattern from that of soil. The dark-coloured melanin containing, fungi, or the are predominated over the hyaline ones, contrasting the pattern in the soil.

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Table 1: The Assiut University Mycological Centre accession number (AUMC) of **filamentous fungal** (all belonging to ascomycota) and **ascomyceteous and basidiomyceteous** yeast strains and their isolation sources with accession GenBank numbers given together with the closest match in the GenBank database and sequence similarity in percent to the match as inferred from Blastn searches of ITS sequences.

AUMC number	Isolation source	Accession number	Length (bp)	Closest Genbank match # ITS	Sequencing similarity (%)	Species	References
Filamento	ous fungi		•				•
6930	Citrus leaf	JQ425378	573	$AY373877 = CBS \ 261.67^{T}$	99	Aspergillus ustus	Haugland et al. 2004
6937	Citrus leaf	JQ425379	570	HQ285615 = KCCM60326	99	Emericella nidulans	
5798	Citrus leaf	JQ425380	566	GU219470 = NRRL 46124	99	Phialemonium curvatum	
Ascomyce	eteous yeasts						
7257	Citrus leaf	JQ425344	395	$GU246267 = CBS 565^{T}$	100	Candida catenulata	Groenewald & Smith 2010
7258	Citrus leaf	JQ425345	700	HQ396523 = CHY 1612 GU256755 = ATCC 60480	100	Kluyveromyces marxianus	Kang <i>et al.</i> 2010
7760	Citrus leaf	JQ425389	409	$GU246267 = CBS 565^{T}$ AJ853765 = WM 6	99 100	Candida catenulata	Groenewald & Smith 2010
7755	Citrus leaf	JQ425360	518	EU315767 $\overline{FM199964} = H5MandK14$	79	Issatchenkia terricola Pichia kudriavzevii	
Basidiom	yceteous yeas	sts					
7791	Grapevine leaf	JQ425362	603	AF417115 = CBS 484 $AY015429 = CBS 491^{T}$	99	Sporidiobolus pararoseus	Fell et al. 2002
7784	Grapevine leaf	JQ425387	590	AF145331 = ATCC 34633 $AF145325 = CBS 7711^{T}$	99	Cryptococcus albidosimilis	Scorzetti et. al. 2000, 2002
7772	Citrus leaf	JQ425367	623	$AF190008 = CBS 140^{T}$ EU480310 = CS11M5c59P	99 100	Filobasidium magnum	Fell <i>et al.</i> 2000
7797	Citrus leaf	JQ083438	520	$AF444473 = CBS 8641^{T}$ EU863543 = PUMCHBY27	100	Trichosporon japonicum	Scorzetti et al. 2002
7800	Citrus leaf	JQ425388	549	$AF444473 = CBS 8641^{T}$ EU863543 = PUMCHBY27	99	Trichosporon japonicum	Scorzetti et al. 2002
7795	Grapevine leaf	JQ425396	626	HQ702343 = UOA/HCPF 10538 AF444541 = CBS 316 ^T	99	Rhodotorula mucilaginosa	Scorzetti et al. 2002
7790	Grapevine leaf	JQ425398	538	EU149786 = CBS 10755 EU149785 = CBS 10634	99	Vishniacozyma carnescens	Connell <i>et al.</i> 2008, Arenz <i>et al.</i> 2006
7787	Citrus leaf	JQ425372	758	$AF294699 = CBS 517.83^{T}$ AF294697 = CBS 170.88	99	Pseudozyma aphidis	
7789	Grapevine leaf	JQ425404	614	AF444493 = CBS 6567 $AF444492 = CBS 6566^{T}$	99	Rhodosporidium paludigenum (Anamorph: Rhodotorula graminis)	Scorzetti et al. 2002
7780	Grapevine leaf	JQ425405	597	EU853846 = ATCC 66034 AF444541 = CBS 316 ^T	100 99	Rhodotorula mucilaginosa	Scorzetti et al. 2002

Table 2: Collective data of counts, percentage counts calculated to total fungi and frequency of occurrence of phyllosphere fungi recovered from the citrus and grapevine on DYM and DRBC agar media bimonthly during the period from April 2008-February 2009 (counts of CFU calculated per gm fresh leaves in each sample, collectively in 18 samples in case of citrus and 15 samples in grapevine).

		(Citrus pl	hyllosphere				G	rapevine	phyllosph	ere	
Таха		DYM			DRBC			DYM			DRBC	
	CFU	%CFU	F&O	CFU	%CFU	F&O	CFU	%CFU	F&O	CFU	%CFU	F&O
Filamentous fungi	1032184	99.48	18 H	1280560	99.58	18 H	25258	78.99	15 H	231860	83.63	15 H
Absidia	40	0.004	1 R	80	0.01	1 R						
A. cylindrospora	40	0.004	1 R									
Absidia sp.				80	0.01	1 R						
Acremonium	80	0.01	1 R							40	0.01	1 R
A. blochii	80	0.01	1 R									
A. chrysogenum										40	0.01	1 R
Alternaria	30272	2.92	18 H	25376	1.97	18 H	91708	28.69	13 H	76236	27.49	14 H
A. alternata	26296	2.53	17 H	20104	1.56	17 H	76052	23.79	13 H	61080	22.03	15H
A. botrytis										80	0.03	1 R
A. chlamydospora	680	0.07	8 M	856	0.07	6 M	4696	1.47	7 M	4156	1.49	8 H
A. citri	472	0.05	7 M	2360	0.18	7 M				440	0.16	2 R
Alternaria sp.	2824	0.27	6 M	2056	0.16	4 L	10960	3.43	4 L	10640	3.84	4 L
Apiospora mentagenii	80	0.01	2 R									
Arthrinium sacchari	440	0.04	2 R	1640	0.13	2 R						
Aspergillus	25720	2.48	18H	24648	1.92	18H	9296	2.907	15H	10664	3.85	15H
A. aculeatinus							180	0.06	2 R	780	0.28	4 L
A. aculeatus	5000	0.48	4 L	1800	0.14	4 L	3360	1.05	8 H	1420	0.51	6 H
A. aureoterreus										40	0.01	1 R
A. auricomus	200	0.02	1 R									
A. brasiliensis	5640	0.54	5 M	7200	0.56	6 M	1520	0.48	4 L	440	0.16	4 L
A. clavatus				64	0.005	2 R						
A. dimorphicus	160	0.02	2 R	200	0.02	2 R						
A. flavipes							80	0.02	2 R			
A, flavus var. flavus	1080	0.10	9 H	1232	0.09	11 H	364	0.11	8 H	344	0.12	7 M
A. flavus var. columnaris	152	0.01	3 L	720	0.06	5 M						
A. fumigatus	80	0.01	1 R							80	0.03	1 R
A. insulicola	80	0.01	1 R	160	0.01	2 R				20	0.01	1 R
A. kevei				80	0.01	1 R						

			Citrus pl	hyllosphere				G	rapevine	phyllosph	ere	
Таха		DYM			DRBC			DYM			DRBC	
	CFU	%CFU	F&O	CFU	%CFU	F&O	CFU	%CFU	F&O	CFU	%CFU	F&O
A. lacticoffeatus				40	0.003	1 R				40	0.01	1 R
A. latus	80	0.01	1 R									
A. montivedensis							20	0.006	1 R			
A. niger	8848	0.85	18 H	10480	0.81	16 H	3680	1.15	13 H	7092	2.56	13 H
A. ochraceus	2512	0.24	10 H	1648	0.13	12 H	256	0.08	9 H	252	0.09	7 M
A. oryzae				40	0.003	1 R						
A. ostianus	200	0.02	2 R									
A. parvathecia	40	0.004	1 R									
A. petrakii	40	0.004	1 R	40	0.003	1 R						
A. proliferans							80	0.03	2 R			
A. puniceus				40	0.003	1 R						
A. sclerotioniger							40	0.01	1 R	200	0.07	1 R
A. sclerotiorum				40	0.003	1 R						
A. speleneus				40	0.003	1 R						
A. stella-maris	240	0.02	1 R									
A. sellatus	1880	0.18	7 M	1120	0.09	6 M	4	0.001	1 R	68	0.02	2 R
A. sulphureus	40	0.004	1 R									
A. sydowii	40	0.004	1 R				20	0.006	1 R			
A. terreus	480	0.05	5 M	776	0.06	6 M	36	0.01	2 R	92	0.03	4 L
A. tubingensis	80	0.01	1 R	40	0.003	1 R	100	0.03	2 R	40	0.01	1 R
A. ustus				120	0.01	2 R						
Aspergillus sp. (sect.	8	0.001	1 R				20	0.006	1 R	180	0.06	2 R
nidulantes)												
Beltrania querna	40	0.004	1 R									
Botryodiplodia theobromae	2600	0.25	6 M	2440	0.19	6 M	20	0.006	1 R	40	0.01	1 R
Ceratocystis radicicicola	80	0.01	1 R									
Chaetomium globosum							20	0.006	1 R			
Chuppia sarcinifera										20	0.01	1 R
Cladosporium	870096	83.86	18 H	1105720	85.99	18 H	128404	40.16	11 H	132224	47.69	12 H
C. cladosporioides	694096	66.89	18 H	921056	71.63	18 H	127132	39.77	10 H	129628	46.76	12 H
C. herbarum				1840	0.14	1 R						
C. oxysporum	53944	5.19	14 H	40184	3.12	12 H	40	0.01	1 R	404	0.15	3 L
C. sphaerospermum	121896	11.75	14 H	142600	11.09	17 H	1232	0.38	4 L	2192	0.79	6 M

			Citrus p	hyllospher	·e			G	rapevine	phyllosp	here	
Taxa		DYM			DRBC			DYM		Ī	DRBC	
	CFU	%CFU	F&O	CFU	%CFU	F&O	CFU	%CFU	F&O	CFU	%CFU	F&O
C. spongiosum	160	0.02	1 R	40	0.003	1 R						
Curvularia	2488	0.25	13 H	160	0.013	2 R	1974	0.62	10 H	776	0.28	9 H
C. clavata	120	0.01	2 R				820	0.26	2 R	160	0.06	2 R
C. lunata	600	0.06	5 M				212	0.07	5 M	464	0.17	4 L
C. penneseti	80	0.01	1 R									
C. spicifer	80	0.01	2 R									
C. subpapendorfii	200	0.02	1 R	40	0.003	1 R						
C. tsudae	1408	0.14	7 M	120	0.01	1 R	932	0.29	5 M	152	0.05	4 L
Dichocladosporium	3600		1 R	7080	0.55	3 L						
chlorocephalum		0.35										
Exserohilum rostratum	584	0.06	6 M	360	0.03	3 L	1044	0.33	5 M	548	0.19	7 M
Fuscoannellis carbonaria	160	0.02	2 R	120	0.01	1 R				20	0.01	1 R
Fusarium	2096	0.20	11 H	1832	0.14	11 H	8072	2.52	12 H	2588	0.93	11 H
F. capmtoceras				40	0.003	1 R						
F. chlamydosporum							180	0.06	3 L	20	0.01	1 R
F. equiseti	800	0.08	1 R	1000	0.08	1 R						
F. incarnatum	480	0.05	8 M	456	0.04	8 M	7112	2.22	10 H	1984	0.72	8 H
F. lactis							140	0.04	1 R	120	0.04	1 R
F. lateritium							200	0.06	1 R	160	0.06	1 R
F. phyllophilum	200	0.02	1 R									
F. proliferatum	320	0.03	2 R	120	0.01	1 R	80	0.03	1 R	80	0.03	1 R
F. scirpi							160	0.05	1 R			
F. solani	40	0.004	1 R	96	0.01	2 R	20	0.006	1 R	60	0.02	3 L
F. thapsinum										80	0.03	1 R
F. verticillioides	256	0.02	3 L	120	0.01	2 R	180	0.06	3 L	84	0.03	3 L
Geosmithia lavendula	280	0.03	1 R									
Gibellulopsis nigrescens							4	0.001	1 R	32	0.01	2 R
Humicola fuscoatra	280	0.03	4 L	120	0.01	3 L	140	0.04	2 R			
Lichtheimia corymbifera				40	0.003	1 R						
Leptodontidium elatius				120	0.01	1 R						
Microascus brevicaulis	120	0.01	1 R	160	0.01	3 L						
Mortierella alpina	240	0.02	2 R	240	0.02	2 R						
Mucor	80	0.01	1 R	8	0.001	1 R				40	0.01	1 R

			Citrus pl	hyllospher	e			G	rapevine	phyllosp	here	
Таха		DYM			DRBC			DYM		1	DRBC	
	CFU	%CFU	F&O	CFU	%CFU	F&O	CFU	%CFU	F&O	CFU	%CFU	F&O
M. circinelloides	80	0.01	1 R	8	0.001	1 R						
M. hiemalis										40	0.01	1 R
Neurospora crassa	40	0.004	1 R				4	0.001	1 R			
Nigrospora	2984	0.29	13 H	480	0.04	6 M	580	0.18	5 M	680	0.25	5 M
N. oryzae	2864	0.28	11 H	480	0.04	6 M	580	0.18	5 M	680	0.25	5 M
N. sphaerica	120	0.01	2 R									
Penicillium	6736	0.65	17 H	6914	0.54	18 H	1348	0.42	10 H	1364	0.49	8 H
P. aurantiogriseum	184	0.011	4 R	112	0.013	3 L						
P. bilaii	80	0.01	1 R	80	0.01	1 R						
P. brevicompactum	8	0.001	1 R	66	0.01	2 R						
P. citrinum	1136	0.11	6 M	2352	0.18	9 H	40	0.01	1 R	4	0.001	1 R
P. crustosum	400	0.04	1 R	400	0.03	1 R						
P. expansum	16	0.002	2 R	208	0.02	2 R						
P. glandicola	200	0.02	1 R									
P. griseofulvum	40	0.004	1 R	80	0.01	1 R	80	0.03	1 R			
P. italicum	400	0.04	1 R									
P. ochrochloron	40	0.004	1 R									
P. olsonii	1136	0.11	7 M	536	0.04	6 M						
P. oxalicum	1800	0.17	10 H	1424	0.11	8 M	1048	0.33	8 H	1036	0.37	8 H
P. roquefortii	56	0.01	1 R	8	0.001	1 R						
P. variabile							20	0.006	1 R	60	0.02	1 R
Phoma	5776	0.56	15 H	10624	0.83	14 H	7920	2.48	11 H	3588	1.29	7 M
P. epicoccina	5776	0.56	15 H	10624	0.83	14 H	7800	2.44	11 H	3588	1.29	7 M
P. eupyrena							120	0.04	2 R			
Pithomyces atro-olivaceus				8	0.001	1 R						
Pleurodesmospora sp.	40	0.004	1 R									
Pochonia sp.	200	0.02	1 R	200	0.02	1 R				20	0.01	1 R
Quambalaria cyanescens	69320	6.68	9 H	86800	6.75	9 H	240	0.08	3 L	1200	0.43	4 L
\tilde{z} Rhizopus arrhizus							96	0.03	5 M	64	0.02	3 L
Sagenomella diversispora										8	0.003	2 R
Sarocladium	40	0.004	1 R	48	0.004	2R						
S. kiliense				8	0.001	1 R						
S. strictum	40	0.004	1 R	40	0.003	1 R						

		(Citrus ph	yllosphere				Gi	rapevine	phyllosphe	ere	
Таха		DYM	•		DRBC			DYM	•	1	DRBC	
	CFU	%CFU	F&O	CFU	%CFU	F&O	CFU	%CFU	F&O	CFU	%CFU	F&O
Schizocillium tetrasperma				40	0.003	1 R						
Scytalidium	80	0.01	2 R	120	0.01	2 R						
S. japonicum	40	0.004	1 R	80	0.01	1 R						
S. lignicola	40	0.004	1 R	40	0.003	1 R						
<i>Stachybotrys</i> sp. (synnematous)							48	0.01	2 R	28	0.01	2 R
Stemphylium	5632	0.55	16 H	2954	0.23	12 H	636	0.20	11 H	1176	0.42	11 H
S. botryosum	3552	0.34	10 H	1874	0.15	8 M	492	0.15	8 H	944	0.34	8 H
S. sarciniforme				40	0.001	1 R				180	0.06	1 R
S. vesicarium	1280	0.124	4 L	880	0.07	2 R	24	0.007	1 R			
Stemphylium spp.	800	0.08	5 M	160	0.01	2 R	120	0.04	3 L	52	0.02	2 R
Talaromyces	1240	0.12	11H	1728	0.14	13H	200	0.07	3L	264	0.09	4L
P. duclauxii	240	0.02	3 L	488	0.04	7 M	80	0.03	1 R	200	0.07	1 R
P. funiculosus	40	0.004	1 R									
T. pinophilus				120	0.01	2 R						
T purpurogenus	960	0.09	8 M	1040	0.08	7 M	80	0.03	2 R	64	0.02	4 L
T. helicus							40	0.01	1 R			
Talaromyces sp.				80	0.01	1 R						
Trichoderma	400	0.04	6 M	520	0.04	3 L						
T. atroviride	40	0.004	1 R	80	0.01	1 R						
T. aureoviride	80	0.01	2 R									
T. harzianum	200	0.02	1 R	400	0.03	1 R						
T. longibrachiatum				40	0.003	1 R						
T. paraceramosum	40	0.004	1 R									
Trichoderma sp.	40	0.004	1 R									
Trichothecium roseum	400	0.04	1 R	400	0.03	1 R						
Verticillium fungicola							480	0.15	1 R	45384	16.37	12 H
Yeasts	7200	0.69	13 H	5352	0.42	13 H	68160	21.01	12 H	45384	16.37	12 H
Candida catenulata	1880	0.18	4 L	1560	0.12	5 M						
Cryptococcus	3670	0.35	6 M	1592	0.13	8 M	31288	9.79	10H	14220	5.09	4L
C. albidosimilis										184	0.07	3 L
C. albidus	3592	0.35	5 M	1552	0.12	6 M	31288	9.79	10H	13936	5.03	10H
C. luteolus	80	0.008	1 R	40	0.003	1 R						

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		(Citrus ph	yllosphere				Gr	apevine	phyllosph	ere	
Таха		DYM			DRBC			DYM			DRBC	
	CFU	%CFU	F&O	CFU	%CFU	F&O	CFU	%CFU	F&O	CFU	%CFU	F&O
Filobasidium	8	0.001	1R	136	0.011	3R				36	0.011	3L
F. floriforme	8	0.001	1 R	120	0.01	1 R				4	0.001	1 R
F. magnum				16	0.001	2 R				32	0.01	2 R
Geotrichum citri-aurentii	280	0.03	1 R	760	0.06	1 R						
Kluyveromyces marxianus	840	0.08	1 R	320	0.02	1 R	320	0.10	2R	440	0.16	2 R
Papiliotrema laurentii	56	0.005	2 R	96	0.01	2 R	164	0.05	3L	768	0.28	5 M
Pichia	40	0.004	1 R	240	0.02	2 R	60	0.016	2 R	772	0.274	4L
P. gluillermondiil							20	0.006	1R	760	0.27	3L
P. kudriavzevii	40	0.004	1 R	240	0.02	2 R	40	0.01	1R	12	0.004	1R
Pseudozyma	64	0.006	2 R	136	0.01	1 R						
P. aphidis	40	0.004	1 R									
P. rugulosa	24	0.002	1 R	16	0.001	1 R						
<i>Pseudozyma</i> sp.				120	0.01	1 R						
Rodosporidium												
paludigenum							820	0.26	4L	660	0.24	3 L
Rhodotorula	160	0.02	2 R	360	0.03	3 L	33492	10.48	8M	27876	10.05	9 H
R. glutinis	120	0.01	1 R	360	0.03	3 L	540	0.17	1R	8	0.003	1 R
R. mucilaginosa	40	0.004	1 R				32952	10.31	8M	27868	10.05	9 H
Sporidiobolus	24	0.002	1 R				460	0.146	5M	468	0.17	5M
S. mearoseus	24	0.002	1 R				20	0.006	2R	24	0.009	2R
S. pararoseus							440	0.14	3L	400	0.14	2R
S. ruineniae										44	0.02	2R
Trichosporon japonicum	120	0.01	1 R									
Vishniacozyma carnescens	16	0.002	1 R	32	0.002	1 R	4	0.001	1R	140	0.05	5 M
Yeast sp. (black)	40	0.004	1 R	120	0.01	1 R						
Total CFU	1037584	100	18 H	1285916	100	18 H	320688	100	15 H	277244	100	15 H
No. of genera (58)		46			41			28			32	
No. of species (138)		107+1			95+1			66			72+1	

*F = Frequency of occurrence out of 18 samples in case of citrus and 15 samples in grapevine. *O = Occurrence remarks for citrus: H = high, 9-18; M = moderate, 5-8; L = Low, 3-4; R = rare, 1-2 samples

= For grapevine: H, 8-15; M, 5-7; L, 3-4; R= 1-2 samples

Table 3: Collective data of counts, percentage counts calculated to total fungi and frequency of occurrence of phylloplane fungi recovered from citrus and grapevine plantations on DYM and DRBC agar media bimonthly during the period from April 2008- February 2009 (counts of CFU calculated per 20 fresh leaf pieces in each sample, collectively in 18 samples in case of citrus and 15 samples in grapevine).

Таха		(Citrus p	hyllopla	ne			(Frapevin	e phyllo	plane	
		DYM			DRBC			DYM			DRBC	
	CFU	%CFU	F&O	CFU	%CFU	F&O	CFU	%CFU	F&O	CFU	%CFU	F&O
Filamentous fungi	2273	93.46	18 H	2766	97.29	18 H	1003	94.27	15 H	1076	94.14	15 H
Acremonium fusidioides	1	0.04	1 R									
Alternaria	165	6.78	17 H	170	5.98	18 H	492	46.24	13 H	572	50.04	15 H
A. alternata	122	5.02	17 H	128	4.50	18 H	433	40.69	13 H	440	38.49	15 H
A. atra										1	0.09	1 R
A. botrytis										1	0.09	1 R
A. chlamydospora	24	0.99	7 M	14	0.49	9 H	25	2.35	10 H	59	5.16	8 H
A. citri	12	0.49	7 M	23	0.81	7 M	2	0.19	1 R	8	0.69	3 L
A. limoniasperae	4	0.16	1 R	3	0.11	1 R						
A. longipes				2	0.07	1 R						
A. tenussima										4	0.35	1 R
Alternaria sp.	3	0.12	2 R				32	3.01	5 M	59	5.16	7 M
Apiospora montagonii	1	0.04	1 R									
Arthrinium sacchari	1	0.04	1 R	1	0.04	1 R						
Aspergillus	174	7.15	18 H	156	5.48	18 H	129	12.12	15 H	123	10.06	15 H
A. aculeatinus							2	0.19	1 R	2	0.17	2 R
A. aculeatus	26	1.07	3 L	23	0.81	3 L	35	3.29	6 M	26	2.27	6 M
A. brasiliensis	22	0.90	5 M	19	0.67	5 M	34	3.19	6 M	21	1.84	6 M
A. dentatus										1	0.09	1 R
A. dimorphicus	3	0.12	2 R									
A. flavipes							5	0.47	2 R			
A. flavus var. flavus	11	0.45	6 M	6	0.21	6 M	7	0.66	5 M	9	0.79	6 M
A. flavus var. columnaris	17	0.69	3 L	7	0.25	2 R	1	0.09	1 R			
A. fumigatus	2	0.08	2 R				1	0.09	1 R	2	0.17	1 R
A. insulicola				1	0.04	1 R						
A. japonicus							3	0.28	1 R			
A. lacticoffeatus	1	0.04	1 R	1	0.04	1 R						
A. montivedensis	1	0.04	1 R	4	0.14	1 R						
A. niger	61	2.51	12 H	64	2.25	10 H	33	3.10	11 H	48	4.19	11 H

Taxa		(Citrus pl	hyllopla	ne			G	Frapevin	e phyllo	plane	
		DYM			DRBC			DYM	-		DRBC	
	CFU	%CFU	F&O	CFU	%CFU	F&O	CFU	%CFU	F&O	CFU	%CFU	F&O
A. ochraceus	24	0.98	8 M	22	0.77	7 M	3	0.28	3 L	7	0.61	4 L
A. ostianus	1	0.04	1 R									
A. petrakii	2	0.08	2 R									
A. proliferans							1	0.09	1 R			
A. stellatus	11	0.45	5 M	5	0.18	3 L						
A. sydowii	1	0.04	1 R							1	0.09	1 R
A. terreus	1	0.04	1 R	1	0.04	1 R	3	0.28	2 R	2	0.17	2 R
A. tubingensis	1	0.04	1 R									
A. ustus				2	0.07	1 R						
Aspergillus spp. (Sect. Nidulantes)				1	0.04	1 R	1	0.09	1 R	3	0.26	2 R
Basidiomycete sp.							1	0.09	1 R			
Beltrania querna				1	0.04	1 R						
Bipolaris papendorfii										1	0.09	1 R
Botryodiplodia theobromae	3	0.12	2 R	8	0.28	3 L	1	0.09	1 R			
Chaetomium				1	0.04	1 R	1	0.09	1 R			
C. globosum							1	0.09	1 R			
C. erectum				1	0.04	1 R						
Chrysosporium keratinophilum				1	0.04	1 R						
Cladosporium	1439	59.17	17 H	1836	64.58	18 H	93	8.74	8 H	167	14.61	8 H
C. cladosporioides	881	36.23	17 H	1082	38.06	18 H	47	4.42	6 M	43	3.76	6 M
C. herbarum							40	3.76	3 L	117	10.24	3 L
C. oxysporum	177	7.28	7 M	193	6.79	9 H	1	0.09	1 R	5	0.44	3 L
C. sphaerospermum	381	15.67	13 H	561	19.73	14 H	5	0.47	2 R	2	0.17	2 R
Curvularia	19	0.69	8M	10	0.35	7 M	36	3.94	12 H	21	1.83	6 M
C. clavata	2	0.08	2 R				16	1.50	5 M	3	0.26	1 R
C. lunata	4	0.16	3 L	3	0.11	3 L	9	0.85	6 M	3	0.26	3 L
C. pallescense				2	0.07	1 R						
C. tsudae	11	0.45	4 L	5	0.18	4 L	17	1.59	5 M	15	1.31	4 L
C. tuberculata										1	0.09	1 R
Dichocladosporium chlorocephalum	31	1.27	3 L	78	2.74	4 L	1	0.09	1 R	3	0.26	1 R
Embellisia didymospora				1	0.04	1 R						
Exserohilum rostratum	5	0.21	2 R	3	0.11	2 R	34	3.19	8 H	21	1.84	5 M
Fusarium	5	0.21	3 L	2	0.07	2 R	11	1.03	5 M	4	0.35	2 R

Таха		(Citrus p	hyllopla	ne			6	Frapevin	e phyllo	plane	
		DYM			DRBC			DYM			DRBC	
	CFU	%CFU	F&O	CFU	%CFU	F&O	CFU	%CFU	F&O	CFU	%CFU	F&O
F. camptoceras	1	0.04	1 R									
F. incarnatum	2	0.08	2 R	1	0.04	1 R	7	0.66	5 M	3	0.26	1 R
F. proliferatum	1	0.04	1 R	1	0.04	1 R						
F. scripi							1	0.09	1 R			
F. solani	1	0.04	1 R				1	0.09	1 R			
F. verticillioides							2	0.19	1 R	1	0.09	1 R
Fuscoannellis carbonaria				1	0.04	1 R	3	0.28	1 R	2	0.17	1 R
Geosmithia lavendula				2	0.07	1 R						
Humicola fuscoatra	5	0.21	1 R	4	0.14	1 R				1	0.09	1 R
Macrophomina phaseolina	8	0.33	1 R	1	0.04	1 R						
Memmnoniella echinata	2	0.08	1 R	1	0.04	1 R						
Mucor circinelloides							1	0.09	1 R	1	0.09	1 R
Myrothecium	2	0.08	2 R	5	0.18	3 L	3	0.28	1 R			
M. roridum							3	0.28	1 R			
M. verrucaria	2	0.08	2 R	5	0.18	3 L						
Nigrospora oryzae	44	1.81	14 H	10	0.35	7 M	12	1.13	6 M	13	1.14	7 M
Paecillium lilacinum				1	0.04	1 R						
Penicillium	36	1.48	10 H	33	1.15	11 H	14	1.32	5 M	19	1.66	8 H
P. aurantiogriseum	8	0.33	2 R	7	0.25	2 R						
P. citrinum	12	0.49	4 L	8	0.28	4 L						
P. crustosum	3	0.12	2 R	2	0.07	1 R						
P. digitatum	1	0.04	1 R	2	0.07	2 R						
P. griseofulvum	1	0.04	1 R	2	0.07	1 R	1	0.09	1 R	1	0.09	1 R
P. italicum	2	0.08	2 R									
P. olsonii	1	0.04	1 R	4	0.14	2 R						
P. oxalicum	6	0.25	3 L	8	0.28	4 L	10	0.94	4 L	18	1.57	7 M
P. roquefortii	2	0.08	1 R									
P. variabile							3	0.28	1 R			
Phialomonium curvatum	1	0.04	1 R									
Phoma	38	1.56	14 H	48	1.69	13 H	127	11.94	10 H	74	6.47	8 H
P. epicoccina	38	1.56	14 H	48	1.69	13 H	124	11.65	10 H	72	6.29	6 M
P. eupyrena							3	0.28	1 R	2	0.17	2 R
Pochonia sp.							3	0.28	1 R			

Таха		(Citrus pl	hylloplai	ne			G	rapevin	e phyllo	plane	
		DYM			DRBC			DYM			DRBC	
	CFU	%CFU	F&O	CFU	%CFU	F&O	CFU	%CFU	F&O	CFU	%CFU	F&O
Quambalaria cyanescens	241	9.91	5 M	354	12.45	6 M	2	0.19	1 R	2	0.17	1 R
Rhizopus arrhizus	1	0.04	1 R	1	0.04	1 R	13	1.22	4 L			
Sarocladium strictum	2	0.08	1 R	1	0.04	1 R						
Scytalidium japonicum	1	0.04	1 R	3	0.11	1 R						
Stachybotrys							2	0.19	1 R			
S. chartarum							1	0.09	1 R			
Stachybotrys sp. (Synnematous)							1	0.09	1 R			
Stemphylium	29	1.19	11 H	22	0.77	11 H	17	1.59	5M	31	2.71	9H
S. botryosum	13	0.53	6 M	9	0.32	6 M	10	0.94	4 L	25	2.19	6 M
S. sarciniforme	1	0.04	1 R				6	0.56	1 R	3	0.26	1 R
S. vesicarium	2	0.08	1 R	8	0.29	3 L	1	0.09	1 R	1	0.09	1 R
Stemphylium spp.	13	0.54	5 M	5	0.18	3 L				2	0.18	2 R
Talaromyces	6	0.25	4 L	10	0.36	8M	1	0.09	1 R	20	1.75	5M
T. duclauxii				2	0.07	2 R				1	0.09	1 R
T. pinophilus				1	0.04	1 R				2	0.17	2 R
T. purpurogenus	6	0.25	4 L	7	0.25	6 M	1	0.09	1 R	17	1.49	3 L
Trichoderma sp.										1	0.09	1 R
Trichothecium roseum				1	0.04	1 R						
Yeasts	159	6.54	13 H	77	2.71	15 H	61	5.73	10 H	67	5.86	12 H
Candida catenulata	36	1.48	5 M	19	0.67	5 M						
Cryptococcus	49	2.02	5 M	15	0.53	5 M	10	0.94	4 L	19	1.66	8 H
C. albidus	41	1.69	4 L	9	0.32	4 L	10	0.94	4 L	19	1.66	8 H
C. luteolus	8	0.33	1 R	6	0.21	1 R						
Filobasidium	2	0.08	1 R	7	0.25	1 R	2	0.19	1 R	4	0.35	3L
F. floriforme	2	0.08	1 R							1	0.09	1 R
F. magnum				7	0.25	1 R	2	0.19	1 R	3	0.26	2 R
Geotrichum citri-aurantii	8	0.33	2 R	5	0.18	1 R						
Kluyveromyces marxianus	48	1.97	1 R	10	0.35	1 R						
Papiliotrema laurentii	4	0.16	2 R	2	0.07	2 R	5	0.47	1 R	4	0.35	1 R
Pichia kudriavzevii				1	0.04	1 R						
Pseudozyma aphidis				2	0.07	1 R						
Rhodosporidium paludigenum	2	0.08	1 R	2	0.07	1 R	13	1.22	2 R	14	1.22	2 R
Rhodotorula muclaginosa	5	0.21	2 R	6	0.21	4 L	27	2.54	7 M	17	1.49	7 M

Таха		(Citrus pl	hylloplaı	ne			G	rapevin	e phylloj	olane	
		DYM			DRBC			DYM			DRBC	
	CFU	%CFU	F&O	CFU	%CFU	F&O	CFU	%CFU	F&O	CFU	%CFU	F&O
Sporidiobolus							4	0.37	2R	8	0.70	3L
S. metaroseus							1	0.09	1 R	3	0.26	2 R
S. pararoseus							3	0.28	1 R	5	0.44	1 R
Trichosporon japonicum	3	0.12	1 R									
Vishniacozyma carnescens	2	0.08	1 R	9	0.32	1 R				1	0.09	1 R
Total CFU	2432	100	18 H	2843	100	18 H	1064	100	15 H	1143	100	15 H
No. of genera (52)		35			41			28		24		
No. of species $(113 + 2 \text{ varieties})$		73+1			73+1			58+1		56		

*F = Frequency of occurrence out of 18 samples in case of citrus and 15 samples in grapevine.

*O = Occurrence remarks for citrus: H = high, 9-18; M = moderate, 5-8; L = Low, 3-4; R = rare, 1-2 samples

= For grapevine: H, 8-15; M, 5-7; L, 3-4; R= 1-2 samples.

Five species of Cladospoium were collected of which C. cladosporioides was regularly the most frequent species in both plants followed by C. sphaerospermum. C. oxysporum was recorded from both plantations while. C. spongiosum was isolated from only phyllosphere of citrus.

In the phyllosphere, Cladosporium yielded more percentage counts than those recorded in the phylloplane, also in citrus more than those in grapevine. In citrus phyllosphere, it was the most dominant genus, contributing 83.86 % -85.99 % of total fungi. In grapevine phyllosphere, it yielded about half the total percentage counts in citrus phyllosphere, contributing 40.16 % and 47.69 % of grapevine phyllosphere fungi. In citrus phyllosphere, C. cladosporioides was also the most common species, contributing higher percentages of total fungi (66.89 % - 71.63 %) than those in grapevine phyllosphere (39.77 % - 46.76 %). C. sphaerospermum was recorded in high frequency in citrus phyllosphere on both media, contributing relatively high percentage counts (11.75 % - 11.09 %) but in grapevine phyllosphere, it was isolated in low and moderate frequencies, donating small percentage counts (0.38 % - 0.79 %). C. oxysporum was recorded in high frequency in citrus pyllosphere while it was recorded in rare frequency on one medium and low frequency on the other in grapevine phyllosphere. C. herbarum and C. spongiosum were isolated only from citrus phyllosphere.

Cladosporium cladosporioides was one of the most dominant endogenous contaminants on the leaf surface of Sorbus domestica (Labuda et al. 2005, Kačániová and Fikselová 2007), phyllosphere of leaves from different plants cultivated in four reclaimed areas,

Assiut, Egypt (Elkhateeb et al. 2016); leaves of Acanthus ilicifolius var. xiamenen, China (Chi et al. 2019).

In the phylloplane, Cladosporium was also the most common genus, contributing more than half of fungi (59.17 % - 64.58 % in citrus), while in grapevine phylloplane it constituted markedly smaller percentages (8.74 % and 14.61 % of fungi). C. cladosporioides was the most common species, contributing more than one-third of total fungi (36.23 % -38.06 %) in citrus phylloplane, while it yielded markedly smaller percentage counts (3.76 % -4.42 %) in grapevine phylloplane. С. sphaerospermum was recorded in high frequencies in citrus phylloplane on both media, contributing 15.67 % - 19.73 %. In grapevine phylloplane, it was isolated in rare frequency on both media, yielding small percentages (0.47 % and 0.17 %). С. oxysporum was recorded in moderate and high frequencies in citrus phylloplane while in grapevine phylloplane, it was recorded in rare frequency on one medium and low frequency on the other. C. herbarum was isolated in low frequency from grapevine phylloplane only, contributing relatively more percentage than C. sphaerospermum.

Cladosporium, Fusarium, and Alternaria have been described as predominent in phylloplanes (Dickinson 1976, Breeze and Dix 1981, Koizumi and Kuhara 1984, Farr et al. 1989, Fenn et al. 1989, Caretta et al. 1999, Andrews and Harris 2000, Araújo et al. 2001, de Jager et al. 2001, Pereira et al. 2002, Guimarães et al. 2011, Elkhateeb et al. 2016).

Cladosporium cladosporioides, Alternaria alternata, Fusarium oxysporum and Pestalotiopsis sp. were extensively reported as common on leaf surfaces of wide variety of plants throughout the world (Breeze and Dix 1981, Mishra and Dickinson 1981, Andrews 1996, Osono 2006, Elkhateeb et al. 2016) which can withstand adverse conditions such as desiccation, UV radiation and microbial lysis by producing thick walled pigmented multicellular spores and microsclerotia (Hudson 1968, Sadaka and Ponge 2003). These fungi are normally encountered as epiphytes, but some can also occur as endophytes (Petrini 1991). Reports showed that С. sphaerospermum is a typical phylloplane microbe on various plant families and is almost worldwide in distribution (Ouf 1993).

Phytopathologically, *Cladosporium* species are the incitants of serious plant diseases e.g. pod rot and blight of pea and southern pea caused by *C. cladosporioides* (Agrios 2005).

Alternaria was more prevalent in the phyllosphere and phylloplane in citrus than their respectives in grapevine, despite its contribution of higher percentage counts in grapevine than those in citrus. The highest percentage of *Alternaria* propagules was recorded from grapevine phylloplane (46.24% - 50.04 % of total fungi) and grapevine phyllosphere (27.49 % - 28.69 %).

Nine species (6 species from citrus and 7 species from grapevine) were identified from all sources in both plantations. In the phyllosphere, Alternaria yielded less percentage counts than those recorded in the phylloplane, also from citrus than those from grapevine although it was more common in citrus phyllosphere than in grapevine. In citrus phyllosphere, it was the most common genus, despite its small percentage counts (1.97 % -2.92 %). In grapevine phyllosphere, it yielded about one fourth of the total fungal phyllosphere counts, contributing 27.49 % -

28.69 %. Its peak was recovered in February in citrus phyllosphere and in December in grapevine on both media.

In the phyllosphere of both plants, A. alternata was also the most common species of this genus. In grapevine phyllosphere, it contributed higher percentages of total fungi (22.03 % 23.79 %) than in citrus phyllosphere (1.56 % - 2.53 %). Α. chlamydospora was recorded in moderate frequency in citrus phyllosphere on both media, but in grapevine phyllosphere, it was isolated in high or moderate frequency. A. citri was recorded in moderate frequency on both media in citrus pyllosphere while it was recorded in rare on DRBC only in grapevine phyllosphere.

A. alternata was the dominant species in the *Triticum vulgare* phyllosphere in Saudi Arabia (Abdel-Hafez 1981), *Quercus rotundifolia* phyllosphere in a holm oak forest, high Atlas Morocco (Sadaka and Ponge 2003), three forest tree leaves of *Alnus nepalensis, Castanopsis hystrix* and *Schima walichii* in North East India (Kayini and Pandey 2010), leaves of different plants cultivated in four reclaimed areas, Assiut, Egypt (Elkhateeb *et al.* 2016).

In the phylloplane, *Alternaria* was the most common genus, contributing about half of the grapevine phylloplane fungi (46.24 % - 50. 04 % of the total phylloplane propagules in grapevine on DYM and DRBC respectively). However its count was declined so much in citrus phylloplane to 6.78 % and 5.98 % respectively. Its peak in the phylloplane of citrus was recorded in October and December on DYM and DRBC respectively, while in grapevine in December (senescent leaf) on both media.

In the phylloplane of both plants, A. alternata was the most common species of this genus, contributing 38.49 % - 40.69 % in grapevine phylloplane while it yielded relatively small percentage counts (4.50 % -5.02 %) in citrus phylloplane. Α. chlamydospora was recorded in high frequencies in grapevine phylloplane on both media, contributing 2.35 % - 5.16 % of total fungi. In citrus phylloplane, it was isolated in high or moderate frequency, giving small percentages (0.99 % and 0.49 %). A. citri was recorded in moderate frequencies on both media in citrus pylloplane while in grapevine phylloplane it was recorded in rare frequency on one medium and low on the other. A. limonisporae and A. longipes were isolated in rare frequency from citrus phylloplane only, while A. tenuissima was rarely isolated from grapevine phylloplane only.

Alternaria has been described as prominent in the phylloplane of Acer campestre, A. platanoides, A. pseudoplatanus, Quercus petraea, Q. pubescens, Q. robur and Q. rubra (Guimarães et al. 2011), Acer platanoides phylloplane (Breeze and Dix 1981), phylloplane of grassland vegetation, Kenya (Caretta et al. 1999), mango phylloplane (de Jager et al. 2001), and phylloplane of plants growing in a Mediterranean ecosystem (Pereira et al. 2002).

Alternaria sp. was found to be present on the leaves of Ulmus americana and Quercus palustris trees on Tulsa campus, USA (Levetin and Dorsey 2006) and on the leaf surface of tea plantation area of Cachar District, Assam, India (Dutta *et al.* 2010), from Artemisia lavandulifolia, A. tangutica, A. brachyloba, A. subulata, A. argy and A. scoparia in two Chinese localities, Qichun and Wuhan (Cosoveanu *et al.* 2016), from healthy leaves of ten medicinal plants, India (Jariwala and Desai 2018), leaves of *Acanthus ilicifolius* var. *xiamenensis*, China (Chi *et al.* 2019).

Phytopathologically, Alternaria species are the incitants of serious plant diseases e.g. Alternaria brown spot of mandarin and tangerines, caused by A. citri, first was described on emperor mandarin in Australia in 1903 (Cobb 1903, Doidge 1929, Ruehle 1937, Kiely 1964, Pegg 1966, Whiteside 1976, Peever et al. 2004) and A. alternata (Dewdney and Timmer 2011). Black rot of citrus also has been attributed to A. citri and symptoms include a stem-end decay of mature fruit in storage, which can occur on all commercial citrus cultivars (Doidge 1929, Ruehle 1937, Bliss and Fawcett 1944, Kiely, 1964, Pegg 1966, Whiteside 1976). Alternaria leaf spot of rough lemon, originally described from South Africa in 1929 (Doidge 1929). A. citri was also described as one of the citrus moulds (Ritenour et al. 2003).

A. alternata causes black rot of olive and citrus, black point of small cereals, and black mould of several vegetables (Logrieco *et al.* 2003).

The genus *Alternaria* produces 71 known mycotoxins and phytotoxins (Montemurro and Visconti 1992). Mycotoxins include alternariol, alternariol monomethyl ether, alternuene, altertoxins, L-tenuazonic acid and other less toxic metabolites (Liu *et al.* 1992, Wild and Hall 1997, Brandwagt *et al.* 2001, Ito *et al.* 2004, Ostry *et al.* 2007).

The genus *Aspergillus* was almost the most common fungus in the two plants giving higher gross total counts in grapevine compared with those of citrus. Forty-five species and two species varieties of *Aspergillus* (41 species and 2 varieties from citrus and 28 species & 2 species varieties from grapevine) were recorded in both plantations.

In the phyllosphere, this genus yielded lower percentage counts than those recorded in the phylloplane, also from citrus than those from grapevine. It accounted for 1.02 % - 2.48 % of total fungi recovered from citrus phyllosphere, and 2.91 % - 3.85 % of those recorded from grapevine phyllosphere, whereas it contributed 5.48 % - 7.15 % of total fungi from citrus phylloplane and 10.06 % -12.12 % of those recorded from grapevine phylloplane.

The greater percentage counts of *Aspergillus* in the phylloplane than those in the phyllosphere may be attributed to its ability to penetrate the leaf surface cells and cannot be easily washed, and remain attached to them, thus expressing themselves more in the former than in the latter habitat.

In the phyllosphere, Aspergillus (33 + 2)varieties) accounted for 2.48 % and 1.92 % of total fungi on DYM and DRBC respectively in the phyllosphere of citrus and grapevine (2.91 % and 3.85 % respectively). Its peak was recorded in October on DYM and in February on DRBC in citrus phyllosphere and in October and August in DYM and DRBC respectively in grapevine phyllosphere. A. niger was common in the phyllosphere of both plants on both media, while A. aculeatus was recorded in high frequency in grapevine phyllosphere but in low frequency in citrus phyllosphere. A. ochraceus and A. flavus var. flavus were recorded in high frequency on both media in citrus phyllosphere and in high or moderate frequency in grapevine phyllosphere. A. brasiliensis, A. stellaus, and A. terreus were recorded in moderate frequency in citrus

phyllosphere, but in low or rare frequencies in grape phyllosphere. Fifteen species and one species variety of *Aspergillus* were recorded from citrus phyllosphere only while 6 species from grapevine only (Table 2).

In the phylloplane, Aspergillus (22 species + 2 varieties in the phylloplane of both plantations) accounted for 7.15 % and 5.48 % of total fungi on DYM and on DRBC respectively in citrus phylloplane, and constituted relatively more proportions of propagules in grapevine phylloplane (12.12 % and 10.06 % respectively). Its peak was recorded in August on DYM and in October on DRBC in citrus phylloplane and in August (mature leaf) in grapevine phylloplane on both media. Only A. niger was common in both plants on both media, A. flavus var. flavus was recorded in moderate frequency in he phyllosphere of both plants. A. aculeatus was recorded in moderate frequency on both media in grapevine phylloplane but in low frequency in citrus phyllosplane while A. ochraceus was recorded in moderate frequency on both media in citrus phylloplane but in low frequency in grapevine phylloplane. Ten species of Aspergillus were recorded from citrus phylloplane only while 5 species from grapevine only (Table 3).

A. niger was the commonest on citrus leaf in citrus plantations in upper Egypt (Moubasher *et al.* 1971), the phyllosphere and phylloplane of different plants of three different plant families, namely Labiatae, Solanaceae, and Umbelliferae (El-Kady *et al.* 1997), phyllosphere of chili, datura, khella, margoram, peppermint and rosemary (Abdel-Gawad 1984), but came second in the phylloplane of broad bean cultivated in the Oasis, Western Desert (Abdel-Fattah *et al.*

1977). A. niger and A. ochraceus were regularly isolated from the leaves of Citrus sinensis, Gossypium barbadense, Prunus persica, and Punica granatum (Wahab 1975), phyllosphere of broad bean cultivated in the Oasis, Western Desert (Abdel-Fattah et al. 1977), Triticum vulgare phyllosphere, and phylloplane in Saudi Arabia (Abdel-Hafez 1981). However, these two species were not recorded from leaves of Valencia orange in California (Fenn et al. 1989). Aspergillus flavus, A. japonicus, and A. versicolor Aspergillus were the most common species of phylloplane fungi from different plants cultivated in four reclaimed areas at Assiut Governorate in Egypt (Elkhateeb et al. 2016). Aspergillus japonicus was isolated from wild plant Euphorbia indica (Ismail et al. 2018).

Aspergillus sp. was found to be present on the leaves from Artemisia lavandulifolia, A. tangutica, A. brachyloba, A. subulata, A. argy and A. scoparia in Qichun and Wuhan, China (Cosoveanu *et al.* 2016), from healthy leaves of ten medicinal plants, India (Jariwala and Desai 2018).

Phytopathologically, *Aspergillus* species are the incitants of serious plant diseases e.g. *A. niger* aggregate, were the most frequent pathogenic species isolated from the grapes from Dao causing black rot of grapes. Toxigenic strains of *A. carbonarius* and ochratoxin A were often found associated with black rot of grapes (Logrieco *et al.* 2003).

Aspergillus species are also well known to be toxigenic. They are responsible for Ochratoxin A (OTA) production. The most frequent OTA-producing strains found in grapes belong to the species *Aspergillus carbonarius* (Battilani *et al.* 2003, Serra *et al.* 2003) and less frequent *A. niger* and *A. ochraceus* (Serra *et al.* 2005).

A. flavus, Botryodiplodia theobromae, Colletotrichum sp., Mucor sp. and Rhizopus stolonifer were isolated from the skin surface and pulp of sour-sop (Anona muricata. L.) fruits in Abia State, Nigeria (Okwulehie and Alfred 2010), from the leaves of Calotropis procera in Taif region, Saudi Arabia (Gherbawy and Gashgari 2014). Some strains of A. flavus are well known as aflatoxinproducing (Logrieco et al. 2003).

A. fumigatus which, was recovered in rare frequency from both grapevine and citrus plantations was of moderate occurrence in soils collected in the sixties in Egypt (Moubasher and Moustafa 1970), but was the second most frequent fungal species in the seventies (Moubasher and Abdel-Hafez 1978) and was recorded in high frequency in the eighties (Moubasher et al. 1985) while it was recorded in rare frequency in the present and other investigations accomplished in the nineties of last century and first decade of the twenty-first century (refer to Moubasher 2010). The seventies and eighties of the last century witnessed the maximum application of which pesticides might induce basic environmental selective conditions that may be hypothetically responsible for the remarkable abundance during that period.

It is worthy to mention that *Aspergillus* stella-maris was isolated in this study from the phyllosphere of citrus plantations as a second world record (Moubasher & Zeinab Soliman 2011) after its first description in 2008 in the Mediterrenean region from *Eucalyptus* leaf litter in Tunisia and hypersaline saltern water in Slovenia (Zalar *et al.* 2008), and *Aspergillus carlsbadensis* and *Aspergillus* porphyreostipitatus (Moubasher et al. 2018) after their first description from soil of Galapagos Islands, Ecuador, and Carthage, Tunesia by Samson et al. (2011) and from dust from a church, Sayulita, Mexico, and from house dust, Songkhla, Thailand by Visagie et al. (2014).

Fusarium (12 species) was common in phyllosphere of both plants, while it was infrequently isolated from the phylloplane of both plants.

In the phyllosphere, *Fusarium* was of high frequency, yielding more percentage counts than those recorded in the phylloplane, also in grapevine than those in citrus. In citrus phyllosphere, it recorded smaller percentage counts (0.14 % - 0.20 % of total fungi on DYM and DRBC respectively) than those in grapevine phyllosphere (0.93 % - 2.52 %).

In citrus phyllosphere, Only *F. incarnatum* was of high frequency of occurrence in grapevine yielding 0.72 % - 2.22 % of total fungi and of moderate frequency in citrus contributing small percentages (0.04 % - 0.05 %). *F. camptoceras, F equiseti,* and *F. phyllophilum* were isolated from citrus phyllosphere only while *F. chlamyosporum, F. lactis, F. lateritium, F. scripi,* and *F. thapsinum* were isolated from grapevine phyllosphere only.

Fusarium dominated in the phyllosphere of the tea plantation area of Cachar District, Assam, India (Dutta *et al.* 2010). *Fusarium oxysporum* was the dominant colonizer of *Triticum vulgare* phyllosphere in Saudi Arabia (Abdel-Hafez 1981), from fresh and healthy leaves of two young trees of *Otoba gracilipes* in La Carolina, Cali, Colombia (Caicedo *et al.* 2010), from endemic medicinal plants of *Tirumala hills* (Anitha *et al.* 2013), leaves of Calotropis procera in Taif region, Saudi Arabia (Gherbawy and Gashgari 2014), leaves of different plants cultivated in four reclaimed areas, Assiut, Egypt (Elkhateeb *et al.* 2016), the leaves of *Otoba gracilipes*, a medicinal tree from a tropical rainforest in Colombia (Caicedo *et al.* 2019). three forest trees, *Alnus nepalensis, Castanopsis hystrix* and *Schima walichii* leaves, in North East India and *F. equiseti* was recovered from *Alnus nepalensis* phyllosphere (Kayini and Pandey 2010).

In the phylloplane, *Fusarium* was isolated in low or rare frequency in citrus, contributing small percentage counts (0.07 % - 0.21 % of total fungi). In grapevine phylloplane, it was recorded in moderate or rare frequency contributing 0.35 % - 1.03 % of total fungi respectively.

Fusarium has been described as prominent in the phylloplane of Acer campestre, A. platanoides, A. pseudoplatanus, Quercus petraea, Q. pubescens, Q. robur and Q. rubra (Guimarães et al. 2011), Acer platanoides phylloplane (Breeze and Dix 1981), phylloplane of grassland vegetation, Kenya (Caretta et al. 1999), mango phylloplane (de Jager et al. 2001), and phylloplane of plants growing in a Mediterranean ecosystem (Pereira et al. 2002). It was identified from the leaves of Ulmus americana and Quercus palustris trees in Tulsa campus, USA (Levetin and Dorsey 2006). It was isolated from the leaf segments of two medicinal plants Elaeocarpus sphaericus and Myristica fragrans (Deepthi et al. 2018), from healthy leaves of ten medicinal plants, India (Jariwala and Desai 2018).

F. incarnatum was isolated in rare frequency on both media from citrus phylloplane while in moderate or rare frequency from grapevine phylloplane. *F.*

camptoceras and *F. proliferatum* were isolated from citrus phylloplane only while *F. scripi* and *F. verticillioides* were isolated from grapevine phylloplane only.

F. moniliforme (F. verticillioides) was the prevalent species in the phylloplane of fresh water plants, Upper Egypt (El-Hissy *et al.* 1990).

Phytopathologically, Fusarium species are the incitants of serious plant diseases. Fusarium solani is well documented as a pathogen of a number of legumes and other tropical plants where it often is associated with cankers and dieback problems of trees (Nelson et al. 1983), and with twig gum disease of citrus (El-Helaly et al. 1966). F. semitectum (=F. incarnatum) has been reported to cause a canker of walnut, pod and seed rot of beans, reduced seed germination and seedling growth of sorghum, corky dry rot of melons, and storage rot problems of bananas and other fruits (Leslie and Summerell 2006). Fusarium species are also well known to be toxigenic with most common Fusarium mycotoxins are trichothecenes, zearalenones, fumonisins, In addition, moniliformin, beauvericin and fusaproliferin may occasionally present problems (Logrieco et al. 2003).

Fifteen species of *Penicillium* (14 species from citrus and 4 species from grapevine) were recorded from the phyllosphere and phylloplane in both plantations. Eighteen species were isolated from citrus plantations only, while six species were isolated from grapevine plantations only.

In the phyllosphere, *Penicillium* was one of the most common genera yielding lower percentage counts than those recorded in the phylloplane, also from grapevine phyllosphere than those from citrus. It accounted for 0.54 % - 0.65 % of total fungi recovered from citrus phyllosphere on DYM and on DRBC respectively, and 0.42 % - 0.49 % of those recorded from grapevine phyllosphere. P. oxalicum, P. olsonii and P. citrinum were the most frequent species in citrus phyllosphere, but only P. oxalicum predominated in grapevine phyllosphere. It is worthy to mention that 10 species were recorded only from the citrus phyllosphere compared to one species only (*P*. *variabile*) from grapevine phyllosphere (Table 2).

In citrus phyllosphere, *P. citrinum* was recovered in high or moderate frequency but was isolated in rare frequency in grapevine. *P. olsonii* was recorded in moderate frequency on both media from citrus phyllosphere while it was missed in grapevine. *P. oxalicum* was recorded in high frequency on both media in grapevine phyllosphere but in high and moderate frequency on DYM and DRBC in citrus phyllosphere.

Penicillium was found to be present on the phyllosphere of Ulmus americana and Quercus palustris trees on Tulsa campus, USA (Levetin and Dorsey 2006). P. citrinum was found to be present on the tea leaf surface in Barak Valley, Assam, India (Dutta, et al. 2010), from Artemisia lavandulifolia, A. tangutica, A. brachyloba, A. subulata, A. argy and A. scoparia leaves in two Chinese localities, Qichun and Wuhan (Cosoveanu et al. 2016). P. chrysogenum and P. corylophilum were isolated from the phyllosphere of fresh water plants, Upper Egypt (El-Hissy et al. 1990), from the leaves of Calotropis procera in Taif region, Saudi Arabia (Gherbawy and Gashgari 2014). Penicillium chrysogenum, Ρ. corylophilum, Ρ. funiculosum, Р. brevicompactum, P. citrinum were the most common species of phyllosphere fungi from different plants cultivated in four reclaimed areas at Assiut Governorate in Egypt (Elkhateeb *et al.* 2016).

In the phylloplane, *Penicillium* (10 species) was recorded in high frequency in citrus and in high or moderate frequency in grapevine phylloplane. It contributed 1.15 % - 1.48 % of total fungi from citrus phylloplane and 1.32 % - 1.66 % of those recorded from grapevine phylloplane. As explained earlier, it is possible that its cells can penetrate the leaf surface cells and stick strongly to them gaining higher counts in the phylloplane than in the phylloplane in April and October on DYM and on DRBC respectively, while in August (mature leaf) in grapevine phylloplane on both media.

In citrus phylloplane, *P. aurantiogriseum*, *P. citrinum*, *P. crustosum*, *P. digitatum*, *P. italicum*, *P. olsonii* and *P. roquefortii* were isolated in low or rare frequency while they were missed in grapevine phylloplane. *P. oxalicum* was encountered in moderate or low frequency in grapevine phylloplane while in low frequency on both media in citrus phylloplane. *P. variabile* was recorded from grape phylloplane only.

Penicillium was reported in moderate occurrence in the phylloplane of Quercus robur, Q. rubra and Acer platanoides (Guimarães *et al.* 2011), the mango phylloplane (de Jager et al. 2001), phylloplane of plants growing in a Mediterranean ecosystem (Pereira et al. 2002), and leaf litter of a lowland rain forest in Costa Rica (Bills 1994), and Polishook from Artemisia lavandulifolia, A. tangutica, A. brachyloba, A. subulata, A. argy and A. scoparia in two Chinese localities, Qichun and Wuhan (Cosoveanu *et al.* 2016).

Phytopathologically, *Penicillium* species are the incitants of serious plant diseases e.g. *Penicillium brevicompactum* was the most frequent pathogenic species in Madeira grapes. *P. expansum* causes blue mould rot of apple and pear. *P. italicum* and *P. digitatum* which are the incitants of blue and green citrus rot respectively. A few species are among the most common and destructive agents of postharvest diseases, affecting most kinds of fruits and vegetables (Logrieco *et al.* 2003).

Phoma (2 species) was recorded in high frequency from phyllosphere but it was of high moderate incidence in or grapevine phyllosphere. In phylloplane, it constituted high proportions of propagules in grapevine phylloplane (11.94 % and 6.47 % of total fungi on DYM and DRBC respectively) compared with its low count in citrus phylloplane (1.56 % and 1.69 % of total fungi). Its peak in citrus phylloplane was recorded in August and December on DYM and DRBC respectively, and in December (senescent leaf) in grapevine phylloplane on both media. The most common Phoma species was Р. epicoccina (=Epicoccum nigrum) in both plantations. P. recorded eupyrena was in grapevine plantations only. Phoma sp. was the most common in the phyllosphere of Ulmus americana and Quercus palustris trees (Levetin and Dorsey 2006), leaf surfaces (Mishra and Dickinson 1981, Osono 2002, Osono et al. 2004), several varieties of grapevines, Madrid, Spain (González and Tello 2010). (=P.Epicoccum purpurascens epicoccina) was isolated from Alnus nepalensis phyllosphere (Kayini and Pandey 2010). Epicoccum nigrum was one of the most fungal

species isolated from *Triticum vulgare* phyllosphere (Abdel-Hafez 1981). *Phoma eupyrena* was recorded from mature and healthy leaves of *Coffea arabica* in two systems (conventional and organic coffee) in the municipality of Garanhuns, Pernambuco, Brazil (Oliveira *et al.* 2014).

Stemphylium was isolated from in high frequency in both phyllospheres and in moderate frequency in both phylloplanes of both plantations. S. botryosum was more frequent than S. vesicarium and S. sarciniforme. S. botryosum was isolated from leaf and citrus fruit rind (Moubasher et al. 1971), and phyllosphere of broad bean, Oases, Egypt (Abdel-Fattah et al. 1977). Epicoccum nigrum, Stemphylium botryosum, Rhizopus stolonifera, Eurotium amstelodami, and Scopulariopsis brevicaulis were isolated in different frequencies from phyllosphere of different plants cultivated in four reclaimed areas at Assiut Governorate in Egypt (Elkhateeb et al. 2016). .

Curvularia (8 species) was recorded in high frequency in the phyllosphere of both plantations and in high and moderate in grapevine phylloplane and in moderate frequency in citrus phylloplane. The most prevalent species was C. lunata followed by C. tsudae was isolated from all sources in both plantations. Curvularia spp. were isolated from leaves of Artemisia lavandulifolia, A. tangutica, A. brachyloba, A. subulata, A. argy and A. scoparia in two Chinese localities, Qichun and Wuhan (Cosoveanu et al. 2016), from healthy leaves of ten medicinal plants from Gujarat region, India (Jariwala and Desai 2018).

Nigrospora (N. oryzae and N. sphaerica) was recorded in high frequency from citrus

phyllosphere and phylloplane, in moderate frequency from grapevine phyllosphere and phylloplane. N. oryzae represented the genus in phyllospheres and phylloplanes of both plantations while N. sphaerica was recorded from citrus phyllosphere only. Curvularia, Epicoccum, Pithomyces, Drechslera, and, Nigrospora were identified from Ulmus americana and Quercus palustris leaves, USA (Levetin and Dorsey 2006). Drechslera, Curvularia, Phoma and Chaetomium were the most frequent genera in the Qat (*Catha edulis*) phyllosphere, Yemen (Alhubaishi and Abdel-Kader 1991). Nigrospora was isolated from leaves of Artemisia lavandulifolia, Α. tangutica, A. brachyloba, A. subulata, A. argy and A. scoparia in two Chinese localities, Qichun and Wuhan (Cosoveanu et al. 2016). Nigrospora sphaerica was recovered with moderate occurrence from different plants cultivated in four reclaimed areas at Assiut Governorate in Egypt (Elkhateeb et al. 2016).

Botryodiplodia theobromae was recorded in moderate frequency from citrus phyllosphere, low from citrus phylloplane, but rare from both grapevine phyllosphere and phylloplane. *B. theobromae* was recorded as causal agent of mango and banana fruit-rot (El-Helaly *et al.* 1966). *Botryodiplodia theobromae* causing leaf blight on the orchid *Catasetum fimbriatum* in Brazil (Lopes *et al.* 2009).

Exserohilum rostratum was recorded in high frequency from grapevine phylloplane and in rare frequency from citrus phylloplane, while in moderate frequency from the phyllosphere of both plantations. *Exserohilum* was the most common fungus isolated from phyllosphere and phylloplane of banana plants cultivated in Upper Egypt (El-Said 2001). *E. rostratum* was isolated from phyllosphere and phylloplane of broad bean in Oases, Egypt (Abdel-Fattah *et al.* 1977), phyllosphere of different plants cultivated in four reclaimed areas at Assiut Governorate in Egypt (Elkhateeb *et al.* 2016).

Quambalaria (Q. cyanescens) was recorded in high and moderate frequency from citrus phyllosphere and phylloplane, but in low and rare frequency from grapevine phyllosphere, and phylloplane respectively. Its count was relatively higher in cirus (6.68 % - 12.45 %) than in grape leaves (0.08 – 0.43 %). Q. cyanescens was isolated in low frequency from the leaves, and the branches of *Betula pendula* in Moscow city (Antropova *et al.* 2014). *Quambalaria* spp. cause leaf and shoot dieback diseases on young Eucalyptus trees in Australia, Thailand, South America and in both temperate and sub-tropical areas of South Africa (Roux *et al.* 2006).

Talaromyces (7 species) was isolated from both phyllosphere and phylloplane of both plantations. *T. duclauxii* and *T. purpurogenus* were isolated from phyllosphere and phylloplane of both plants.

Trichoderma (5 species and 1 unidentified) was encountered in moderate frequency from citrus phyllosphere and in rare frequency from grapevine phylloplane. All species reported were rare. Trichoderma was isolated from Quercus and Acer phylloplanes, Czech Republic (Guimarães al., 2011). et Trichoderma viride and T. koningii were isolated from the phyllosphere of Alnus nepalensis, Castanopsis hystrix and Schima walichii in a subtropical forest of North East India (Kayini and Pandey, 2010). T. koningii and T. harzianum were recorded as constantly isolated species on well-decomposed leaf litters of Quercus myrsinaefolia (Shirouzu et *al.*, 2009). *T. koningii* was isolated in different frequencies from phyllosphere of different plants in Assiut, Egypt (Elkhateeb *et al.* 2016).

Some other filamentous fungi were recorded but in low or rare frequency from one or both substrates of plantations (Tables 2 & 3).

Yeasts were represented by 21 species and 2 unidentified species belonging to 14 genera from phyllospheres and phylloplanes of both plantations (18 species belong to 14 genera from citrus plantations and 15 species belong to 9 genera from grapevine plantations). They were recovered in high frequency from the phyllospheres and phylloplanes of both plants. Of these species only 5 were ascomycetous and 18 were basidiomycetous.

Yeast fungi of citrus phyllosphere showed their peak of total propagules in February, while those of grapevine in December (senescent leaf), while their trough occurred in August in the phyllosphere of citrus and in April (juvenile leaf) in the phyllosphere of grapevine. However, it showed their peak of total propagules recovered from citrus phylloplane in October and June on DYM and DRBC respectively, and from grapevine in August (mature leaf) on both media, while their trough occurred in April and February in the phylloplane of citrus, and in October (mature leaf) and June (young leaf) in grapevine phylloplane on DYM and DRBC respectively. Wang et al., 2016 found that ascomycete yeasts represented 6.3% of total yeasts and 93.7% were basidiomycete in the Arabidopsis thaliana phyllosphere.

Candida (represented by C. catenulate) wasrecoveredinfrequentlyfromcitrusphyllosphereandphylloplanecontributing

minute percentage anount of CFU, while it was missed from grapevine. *Candida* sp. was isolated from the phyllosphere of *Bauhinia forficate*, *Tabebuia* sp. and *Terminala catappa*, southeastern Brazil (Valarini *et al.*, 2007). *Candida sp.* was presented in the tea phyllosphere, Barak Valley, Assam, India (Dutta *et al.*, 2010), healthy leaves of ten medicinal plants from Gujarat region, India (Jariwala and Desai 2018), leaves of Nanfeng mandarin (*Citrus reticulata* cv. Blanco) in China (Peng *et al.* 2018).

The genus *Cryptococccus* (3 species) was was recovered in high frequency in the phyllosphere of grapevine contributing 9.84 % and 5.43 % of total fungi on DYM and DRBC respectively while in moderate frequency and low counts in citrus. It was recovered in moderate frequency from both phylloplanes, contributing 2.26 % and 1.16 % of total fungi from citrus phylloplane and 1.59 % and 2.36 % from grapevine.

C. albidus was recovered in high frequency on both media in grapevine phyllosphere and in moderate frequency in citrus phyllosphere, contributing the greatest component of the genus counts (0.12 % - 0.35 % and 5.03 % -9.79 % of total fungi in citrus and grapevine phyllospheres respectively. From phylloplanes it was recorded in low frequency in citrus and in low and high in grapevine on DYM and DRBC media, respectively. *C. luteolus* was recovered from only citrus phyllosphere and phylloplane and *C. albidosimilis* from grapevine phyllospere only.

Cryptococcus species were prevalent in pineapple leaves, Rio de Janeiro, Brazil (Robbs *et al.* 1989), phylloplane communities (McCormack *et al.* 1994). *C. albidus* (Fonseca *et al.* 2000; Sugita *et al.*, 2001) was deemed to be ubiquitous phylloplane colonists regardless of plant type or geography (Inácio et al. 2002; Maksimova and Chernov 2004) and it was prevalent from sugarcane leaves, Rio de Janeiro, Brazil (Azeredo et al. 1998), the phyllosphere of Bauhinia forficata, Tabebuia sp. and Terminala catappa, southeastern Brazil (Valarini et al. 2007), apple, plum, and cherry leaves, southwest Slovakia (Slavikova et al. 2009). Cryptococcus spp. are widely distributed and were the dominant species in the phyllosphere of many plants, such as carnivorous plant Drosera indica (Sun et al., 2014), Arabidopsis thaliana phyllosphere, accounting over half of all isolates (Wang et al. 2016).

Filobasidium (represented by *F. floriforme* and *F. magnum*) was in rare frequency on one or boh media from the phyllosphere and phylloplane of both plants.

Geotrichum citri-aurantii was recovered in rare frequency on both media from phylosphere and phylloplane of citrus only, yielding less percentage counts in phyllosphere (0.03 % - 0.06 % of total fungi) than those in the phylloplane (0.18 % - 0.33 %). *Geotrichum* spp. were isolated from healthy leaves of ten medicinal plants from Gujarat region, India (Jariwala and Desai 2018). The fungal pathogen *Geotrichum citri-aurantii* is causing the sour rot; a major postharvest disease of citrus fruit (Pompeo *et al.* 2016).

Klyuveromyces was represented by *K. marixianus* only. It was recorded infrequently from the phyllosphere of both plantations, citrus phylloplane. Its highest percentage count was gained from citrus phylloplane (0.35 % - 1.97 % of total fungi).

Kluyveromyces was found in elm phylloplane in California (Phaff and Starmer

1987). *Candida kefyr* (anamorph of *K. marxianus*) is occasionally involved in superficial candidiasis (Hernandez-Molina *et al.* 1994), was described from a cardiac transplant patient with pulmonary infection (Lutwick *et al.* 1980).

Papiliotrema laurentii was recorded in moderate or low frequency of occurrence in grapevine phyllosphere while, it was recorded in rare frequency in both phylloplanes of both plants and citrus phyllosphere. *P. laurentii* (as *Cryptotococcus laurentii*) was seemed to be ubiquitous phylloplane colonists regardless of plant type or geography (Sugita *et al.* 2000; Takashima *et al.* 2003, Inácio *et al.* 2002; Maksimova and Chernov 2004) and was prevalent species from sugarcane leaves, Rio de Janeiro, Brazil (Azeredo *et al.* 1998), apple, plum, and cherry leaves, southwest Slovakia (Slavikova *et al.* 2009).

Pichia (represented by P. guilliermondii and P. kudriavzevii (anamorph: Candida krusei) was isolated in rare frequency from both phyllospheres on both media but in low frequency on DRBC from both plantations, contributing 0.004 % - 0.02 % of total fungi in citrus phyllosphere and 0.016 % - 0.274 % in grapevine phyllosphere. In the phylloplane, it (as P. guilliermondii) was recorded in rare frequency in citrus phylloplane on DRBC, contributing 0.04 % of total fungi. It was missed in grapevine phylloplane. Pichia guillermondii isolated from was the phyllosphere of Bauhinia forficate, Tabebuia sp. and Terminala catappa, southeastern Brazil (Valarini et al. 2007), apple, plum, and peach leaves, southwest Slovakia (Slavikova 2009). Pichia sp., Hanseniaspora sp., and Meyerozyma sp. were associated with leaves of

Nanfeng mandarin (*Citrus reticulata* cv. Blanco) in China (Peng *et al.* 2018).

Pseudozyma was recorded infrequently from from phyllosphere and phylloplane of only citrus plantations. In the phyllosphere, it was encountered in rare frequency in citrus phyllosphere on both media contributing 0.006 % - 0.01 % of total fungi. *P. aphidis*, *P. rugulosa*, and *Pseudozyma* sp. were recorded from citrus phyllosphere. It was missed in grapevine phyllosphere. In the phylloplane, it was recovered in rare frequency from citrus phylloplane on DRBC only contributing 0.07 % of total fungi, represented by *P. aphidis*. It was missed in grapevine phylloplane.

Pseudozyma aphidis was isolated from apple, cherry, and apricot leaves, Southwest Slovakia (Slavikova *et al.* 2009). It was a common taxon in the phylloplane of seven ferns in Baton Rouge, Lousiana (Sebastian 2012).

Rhodosporidium was isolated infrequently from grapevine phyllosphere and the phylloplane of both plantations on both media. Its highest percentage count was recorded from grapevine phylloplane (1.22 % of total fungi on each medium).

Rhodotorula (2 species) was isolated from both phyllospheres and phylloplanes of both plants but more frequent in grapevine than citrus. Its highest percentage count was also recorded from grapevine phyllosphere (10.05 % - 10.48 % of total fungi) followed by grapevine phylloplane (1.49 % - 2.54 %) but minor proportions from citrus phyllospere and phylloplane. *R. mucilaginosa* was isolated from phyllospheres and phylloplanes of both plants, it was the main component of *Rhodotorula* in grapevine phyllosphere and phylloplane while relatively small proportions of propagules in citrus plantations. *R. glutinis* was recovered from phyllospheres only.

Rhodotorula was prevalent in pineapple leaves, Rio de Janeiro, Brazil (Robbs *et al.*, 1989), the leaf surfaces of *Banksia collina* and *Callistemon viminalis* (Shivas and Brown, 1986), phyllosphere of *Arabidopsis thaliana* (Wang *et al.* 2016).

Rhodotorula mucilaginosa was prevalent in the sugarcane leaves, Rio de Janeiro, Brazil (Azeredo et al., 1998). Rhodotorula glutinis and Rhodotorula mucilaginosa were isolated from apple and plum leaves southwest Slovakia (Slavikova et al., 2009). Phylloplane communities usually comprise deeply pigmented species belonging to the genera Rhodotorula and Sporobolomyces (McCormack et al., 1994). R. glutinis and R. mucilaginosa appear to be prevalent regardless of plant type or geography (Inácio et al., 2002; Maksimova and Chernov 2004).

Sporidiobolus was recorded infrequently on both media from the grapevine phyllosphere and phylloplane only. It was represented by S. metaroseus S. pararoseus and S. ruineniae, contributing contributing minor percentages of propagules. S. metaroseus (as Sporobolomyces roseus) was isolated from the phylosphere of Bauhinia forficata, Tabebuia sp. and *Terminalia* catappa, southeastern Brazil (Valarini et al. 2007). Also, it appears to be prevalent in the phylloplane regardless of plant type or geography (Bai et al. 2002, Fell et al. 2002, Inácio et al. 2002, Maksimova and Chernov 2004). S. pararoseus was isolated in high frequency from phylloplane of seven ferns in Baton Rouge, Lousiana (Sebastian 2012). Sporidiobolus sp. and Rhodotorula sp. were associated with leaves of Nanfeng mandarin

Citrus reticulata cv. Blanco) in China (Peng *et al.* 2018).

Trichosporon (T. japonicum) was isolated only from citrus phyllosphere and phylloplane. *Trichosporon* was one of the predominant yeasts found on sugarcane leaves in Rio de Janeiro, Brazil (Azeredo *et al.*, 1998), plant surfaces (Phaff and Starmer, 1987; Santos *et al.*, 1996).

Vishniacozyma carnescens was isolated in rare frequency from phyllospheres and phylloplane of both plants.

Black yeasts were isolated in rare frequency on DYM from citrus phyllosphere contributing 0.004 % of total fungi. Black yeast isolates were prevalent in pineapple fruit and leaves in Rio de Janeiro, Brazil (Robbs *et al.* 1989). The black yeast *Aureobasidium pullulans* was the dominant species in with leaves of Nanfeng mandarin (*Citrus reticulata* cv. Blanco) in China (Peng *et al.* 2018).

Conclusion: The present study reveals that the dematiaceous fungi outnumbered over the hyaline ones in phyllosphere and phylloplane. Basidiomyceteous yeasts were dominant over ascomyceteous yeasts in these environments. In phyllosphere and the phylloplane, the environmental factors may induce selective effects for the advantage of the dematiaceous fungi over the hyaline ones. The melanincontaining fungi are more adapted to survive the injurious effects of the atmospheric conditions.

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