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Diversity of terrophilous mycobiota of Sinai.

Abdel-Azeem, A. M. [1*] and Ibrahim, M. E. [2]

1. Department of Botany, Faculty of Science, Suez Canal University, Ismailia 41522, Egypt.
2. Department of Biological and Geological Sciences, Faculty of Education, Suez Canal University, Al-Arish, Egypt.

ABSTRACT

After more than eighty years of mycological studies in Egypt, our information about soil mycobiota of Sinai is still mysterious, fragmentary and limited. These fungi have never been the sole target of any comprehensive study before concerning their ecology, taxonomy and diversity. The main objective of this study is to shed some light on the structure, ecology and diversity of mycobiota in the Sinai Peninsula. Samples were collected from eighteen different sites representing six different ecological districts. Fifty species belonging to five classes, seven orders, ten families and twenty-seven genera were recorded from different localities in Sinai. Comparison between the present results and those of other studies on soil fungi of deserts in Egypt shows that there is no specific mycobiota characteristic of Sinai soils.

Keywords: Mycobiota, Sinai, diversity, ecology, taxonomy, terrophilous

INTRODUCTION

The Sinai Peninsula occupies a unique location on the map in the north east of Egypt, since it is considered to be the Asian part of African Egypt. The Sinai desert belongs to the Saharan type (MacGinnies *et al.* 1968). The total length of Sinai's coastlines is about 870 km, nearly 30% of Egypt's 2400-km coastline. The area of the peninsula is about 61.000 km², i.e. almost 6% of the total area of Egypt and three times as much as the area of the Nile Delta. The climate of Sinai belongs to the "Arabian Type", characterized by aridity, winter precipitation and moderate temperature (Danin 1978). It has three main divisions: the low northern part is covered with sand; the central division is predominantly limestone hills and gravel plains; while the southern division is mountainous magnetic and metamorphic rocks. According to Danin (1983), the Sinai desert is divided into twelve districts (Fig. 1), each with its own characteristic climatic and edaphic features.

In 1935, Sabet started his pioneer study on the soil mycobiota of Egypt, followed by a number of studies on the mycobiota of cultivated, desert and salt marshes soils (Abdel-Azeem 1991, 2003; Abdel-Hafez 2000; Abdulla *et al.* 1987; Abdul Wahid 1990; Aboul-Nasr 1981; Ezz El-Din 1988; Ibrahim 1999; Moubasher 1993; Moubasher *et al.* 1990). Up to the present, the mycology of Sinai has been given little attention, and as a result information concerning the mycology of this area is scarce and fragmentary. The main object of the present work is to throw some light on the structure and diversity of the fungal mycobiota of many localities of Sinai.

* Address for Correspondence

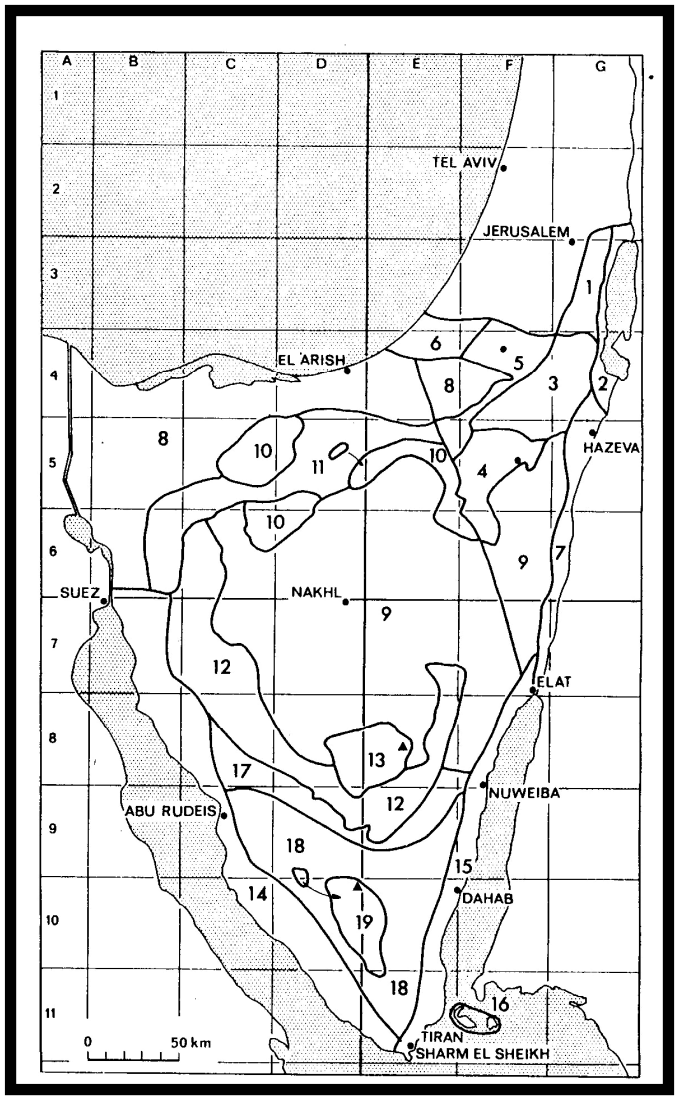


Figure 1: Geomorphological districts of the deserts in Sinai and Israel (occupied Palestine) (after Danin 1983).

MATERIALS AND METHODS

A range of soil samples collected between May 2002 and September 2003 were screened in order as completely as possible to give a preliminary picture of the mycobiota structure of Sinai. Six districts out of the twelve were selected for soil sampling (see Table 1). 162 soil samples were collected under some of the dominant plants from 18 different sites throughout Sinai Peninsula (Fig. 2). These were placed in sterilized polyethylene bags closed by a rubber band, and transferred to the laboratory until plated out. From each site, nine soil samples were collected and mixed thoroughly to form three composite samples.

Mechanical analysis of soil samples was applied using the sieving method (Fathy *et al.* 1975). The pH values of water extracts of soil samples were determined with a pH meter (model 201, Orion Research Co.) using a water-to-soil ratio of 1: 5 (wt/v) to avoid errors of too-high dilution (Jackson 1967). The total soluble salts of soil samples were

determined using an electric conductivity meter (EC-meter) as described by Jackson (1967). Gravimetric methods were used to determine moisture content; organic content was determined by loss-on-ignition (LOI), where loss was calculated as a percentage of the oven-dried sample (Wilde *et al.* 1972).

Table 1: Sites locations and districts numbers.

Site No.	Site Name	District No.
1	El-Kantara -Arish Road	8
2	Al-Arish	8
3	Gebel Halal	10
4	Gebel Lubna	10
5	Hammam Faraon	14
6	El-Tour	14
7	Ras Mohamed	15
8	Sharm El-Sheikh	15
9	Nuweiba	15
10	Taba	15
11	Nabaq	15
12	Dahab	15
13	Wadi Mandar	18
14	Wadi Lithi	18
15	Gebel Catherine	19
16	Wadi El-Deir	19
17	Wadi El-Arbaen	19
18	Wadi Kid	19

The fungal flora of the soil samples was isolated by the dilution plate technique as described by Johnson *et al.* (1960) and modified by Moubasher & Abdel-Hafez (1978). Czapek's agar supplemented with 0.5 % yeast extract (CYA), amended with Rose bengal (1/15000) and chloramphenicol (50 ppm), was used for primary isolation. Fifteen plates were used for each sample. The plates were incubated at 28 ºC for 10 days and the developing fungi counted. For maintaining cultures and for proper identification, pure cultures of isolated fungi were grown on standard media such as Vegetable Agar (V8), Oatmeal Agar (OA), Malt Extract Agar (MEA) Potato Dextrose Agar (PDA) and Potato Carrot Agar (PCA).

Fungal isolates were identified using various sources: e.g. *Aspergillus* (Raper & Fennell 1965); *Penicillium* (Raper & Thom 1949, Pitt 1979); *Chaetomium* (Arx *et al.* 1986, Cannon 1986); *Fusarium* (Booth 1971); for dark-coloured Hyphomycetes (Ellis 1971, 1976); Samson (1974) for *Paecilomyces* and some allied *Hyphomycetes*; and other general taxonomic works (Domsch *et al.* 1980, Arx 1981). The system proposed by Hawksworth *et al.* (1995) was used for higher classification.

Various parameters pertaining to the degrees of occurrence, richness, diversity, and similarity were used. The abundance of fungi is presented in terms of frequency i.e. as number of cases of isolation. To estimate the similarity of species composition among

different sites, we used the similarity coefficient suggested by Sorenson (1948). Species richness is calculated after Barbour *et al.* (1987), while species diversity is calculated as Simpson's diversity index (Bakus 1990).

The data were manipulated using Excel (Microsoft Office-XP Package, 2002), and subjected to statistical analyses with Minitab (version 9.2, 1993) and Statistica (version 5.1 F, 1997).

RESULTS

The organic matter of the soils ranged between 1.24-5.7 %. Moisture content ranged between 0.25 (Wadi El-Arbaein) and 7.5 % (Gebel Katherine): the majority of sites were between 0.95 and 5.5 %. Soil pH ranged between 7.17 and 8.20. Soil textures were assessed as sandy to gravelly sandy. Total dissolved salts ranged mainly between 84 and 585 ppm.

We recorded twenty-seven genera and fifty species from eighteen sites and six districts (Table 2). The most prevalent species were *Aspergillus niger*, *Aspergillus flavus*, *Aspergillus alutaceus*, *Emericella nidulans*, *Chaetomium globosum*, *Aspergillus terreus*, *Aspergillus versicolor* and yeast. These occurred in 50-100% of the samples, and formed 2.0-40.7 % of total fungi in the eighteen sites.

The species richness at the generic level shows that *Aspergillus* is the most diverse (represented by 13 species), followed by *Penicillium* (8 species), *Paecilomyces* and then *Chaetomium*, *Eurotium*, and *Acremonium* (2 species each). All the remaining genera were represented by only one species each. At the family level, the Moniliaceae had the highest contribution (30 species), followed by Mucoraceae (4 species), Chaetomiaceae and Trichocomaceae (3 species), Agonomycetaceae, Sordariaceae and Dematiaceae (2 species) and Microascaceae, Tuberculariaceae and Syncephalastraceae (1 species each). The species-to-genus ratio per family shows that family Moniliaceae was the most diverse taxonomical rank (3.3), followed by Chaetomiaceae and Trichocomaceae.

The species isolated were assigned to five classes, seven orders, and ten families. Mitosporic fungi had fifteen genera, thirty-five species and 88.7 % of the total fungi isolated; Ascomycota had seven genera, nine species and 9.6 %; Zygomycota had only 1.7 % of the total fungi isolated.

Biodiversity has three important aspects: ecological or habitat diversity covers the concepts of population, community, ecosystem and biome; species diversity addresses both the number of species (species richness) and their relative abundance; and genetic diversity, since individuals will not be genetically identical. The species diversity of fungi was estimated using two indices: species richness and Simpson's diversity index.

Species richness was highest at Hammam Faraon (25 species), and a number of other sites also had high species richness (sites 14, 8, 7, 17, etc). Figure 4 shows the species richness among different districts, with district 19 (the high mountains) having the highest at 33 species. Hammam Faraon also showed the highest Simpson diversity (0.878885), followed by other sites in southern Sinai. The lowest Simpson diversity was recorded at Ismailia (site 1).

Based on the presence\absence of fungal species in the studied sites, the distribution pattern (Fig. 3) shows that recorded taxa could be tentatively classified into three groups. Group 1 comprises taxa restricted to a single site e.g. *Achaetomium macrosporum*, *Acremonium strictum*, *Gelasinospora seminude* and *Fusarium solani*. Group 2 consists of species occurring in two or more sites but with higher prevalence in one, e.g. *Eurotium chevalieri*, *Microascus cinereus* and *Penicillium citrinum*. Group 3

contains species common to almost all sites, e.g. *Aspergillus niger*, *A. flavus*, *Chaetomium globosum* and *Emericella nidulans*.

Table 2: Total counts (colonies per g dry soil), frequency of occurrence, number of cases of isolation (NCI, out of 18 sites), total count percentage out of 17380.

Species	Total CFU	Freq. (%)	NCI	Total count %
Absidia corymbifera (Cohn) Sacc. & Trotter	50	1.85	5	0.29
Achaetomium macrosporum Rai, Wadhwani & Tewari	40	1.11	1	0.23
Acremonium rutilum Gams	190	2.59	1	1.09
Acremonium strictum Gams	120	1.85	1	0.69
Acrophilaophora nainiana Edward	10	0.37	1	0.06
Actinomucor elegans (Eidam) Benj. & Hesseltine.	30	1.11	1	0.17
Agonomycete	20	0.74	1	0.12
Alternaria alternata (Fr.) Keissler	10	0.37	1	0.06
Aspergillus alutaceus Berk. & Curt.	390	9.63	11	2.24
Aspergillus candidus Link ex. Link	30	1.11	3	0.17
Aspergillus clavatus Desm.	40	1.11	2	0.23
Aspergillus flavus Link ex Fr.	3210	50.74	18	18.47
Aspergillus fumigatus Fres.	360	6.67	4	2.07
Aspergillus japonicus Saito	140	3.7	6	0.81
Aspergillus niger van Tieghem	7080	90.74	18	40.74
Aspergillus sydowii (Bain. & Sart.) Thom & Church	150	4.07	7	0.86
Aspergillus tamarii Kita	120	3.33	6	0.69
Aspergillus terreus Thom	370	8.52	9	2.13
Aspergillus ustus (Bain.) Thom & Church	80	1.85	1	0.46
Aspergillus versicolor (Vuill.) Tiraboschi	390	8.89	9	2.24
Aspergillus wentii Wehmer	50	1.85	4	0.29
Auerobasidium pullulans (de Bary) Arnaud	40	1.48	1	0.23
Bipolaris spicifera (Bain.) Subram.	70	2.22	3	0.4
Chaetomium brasilense Batista & Pontual	120	2.59	2	0.69
Chaetomium globosum Kunze	860	16.67	12	4.95
Chrysosporium xerophilum Pitt	100	1.85	3	0.58
Emericella nidulans (Eidam) Vuillemin	440	11.11	13	2.53
Eurotium amstelodami Mangin	20	0.37	1	0.12
Eurotium chevalieri Mangin	40	1.48	2	0.23
Fusarium solani (Mart.) Appel & Wollenw	90	2.59	1	0.52
Gelasinospora seminuda Cailleux	20	0.74	1	0.12
Humicola fuscoatra Traaen	220	4.44	4	1.27
Macrophomina phaseolina (Tassi) Goidanich	150	4.07	3	0.86
Microascus cinereus (Emile-Weil & Gaudin) Curzi	80	1.85	2	0.46
Mucor circinelloides van Tieghem	10	0.37	1	0.06
Paecilomyces farinosus (Holm ex Gray) Brown & Sm.	10	0.37	1	0.06
Paecilomyces variotii Bainier	60	2.22	5	0.35
Penicillium brevi-compactum Dierekx	200	4.81	8	1.15
Penicillium canescences Sopp	150	2.96	5	0.86
Penicillium chrysogenum Thom	490	5.93	4	2.82
Penicillium citrinum Thom	50	1.11	2	0.29

Penicillium cyclopium Westling	170	3.33	4	0.98
Penicillium purpurogenum Stoll	50	1.48	3	0.29
Penicillium raciborskii Zaleski	40	1.11	2	0.23
Penicillium variable Wehmer	260	2.22	5	1.5
Rhizopus stolonifer Ehrenb. ex Fr.	180	6.3	7	1.04
Sordaria fimicola (Roberge) Cesati & de Notaris	70	1.85	3	0.4
Syncephalastrum racemosum Cohn ex Schroet	20	0.74	2	0.12
Trichoderma psuedokoningii Rifai	140	4.44	6	0.81
Yeast	350	8.15	9	2.01
Gross Total Count (CFU)	17380			

An important question needs to be answered: are mycobiota the same in all districts? or does each district has a characteristic group of species?. To find an answer, we clustered the sites using Sorensen's similarity coefficient (Fig. 4). The lagomorph mycotas (species in common) formed two main subgroups. Four sites (16, 17, 18 and 6) were rather differnet from the rest, which were divided into two noticeable clusters, a large one (sites 1, 2, 3, 11, 13, 9, 12, 15, 4, 10 and 14) and a small one (sites 5, 7 and 8). We interpret these clusters as showing that interrelations between sites are based on edaphic features and vegetation structure.

DISCUSSION

Sinai desert soils showed relatively low counts of fungal populations (an average count of 330.18 CFU/ g). Most of the taxa have been isolated previously from Egyptian desert soils, but this is the first report concerning ecology, taxonomy and diversity of these fungi in Sinai. Similar observations of low counts associated with a narrow spectrum of species have been also reported by other investigators (Abdel-Azeem 1991, 2003; Moubasher *et al.* 1985, Abdel-Hafez *et al.* 2000). The soil moisture content is usually low because of reduced water-holding capacity and the high rate of evapotranspiration resulting from strong solar radiation. These factors together may account for the relatively low water activity, which greatly affects microbial diversity and activity.

Based on the value of frequency of occurrence, the distribution pattern of fungal species among different types soils and districts indicated that while some species were of restricted occurrence on certain types of soils, others were of common occurrence in almost all types of soils. Such observations have been reported before among soil fungi (Abdel-Azeem 2003) and coprophilous fungi (Lundqvist 1972; Angel & Wicklow 1975; Parker 1979; Richardson 2001). These authors advocated that the physical and chemical properties of substrates differ from one site to another, and accordingly soil from a particular district would favour colonization by certain fungi. The lagomorph mycotas produced by using similarity coefficient showing that the geomorphological, climatic and edaphic factors of each district behave as key characters (Danin 1983) in the distribution of their mycobiota.

Comparison of the taxonomic list isolated in the present study with those obtained in these other studies on desert soils indicates that the Hyphomycetales probably constitute the main core of mitosporic fungi of desert habitats.

Our list of ascosporic species by comparison with those obtained from other Egyptian studies shows that the following species are common to desert habitats: *Eurotium amstelodami*, *E. chevalieri*, *Emericella nidulans*, *Chaetomium globosum* and *Microascus*

cinereus. The xerophilic potentiality of the first three taxa has already been documented (Hocking & Norton 1983) while the latter two require further investigations.

A comparison between the present results and those of other studies on soil fungi of deserts in Egypt shows that there is no specific mycobiota characteristic of Sinai soils.

Figure 2: Map of locations sampled

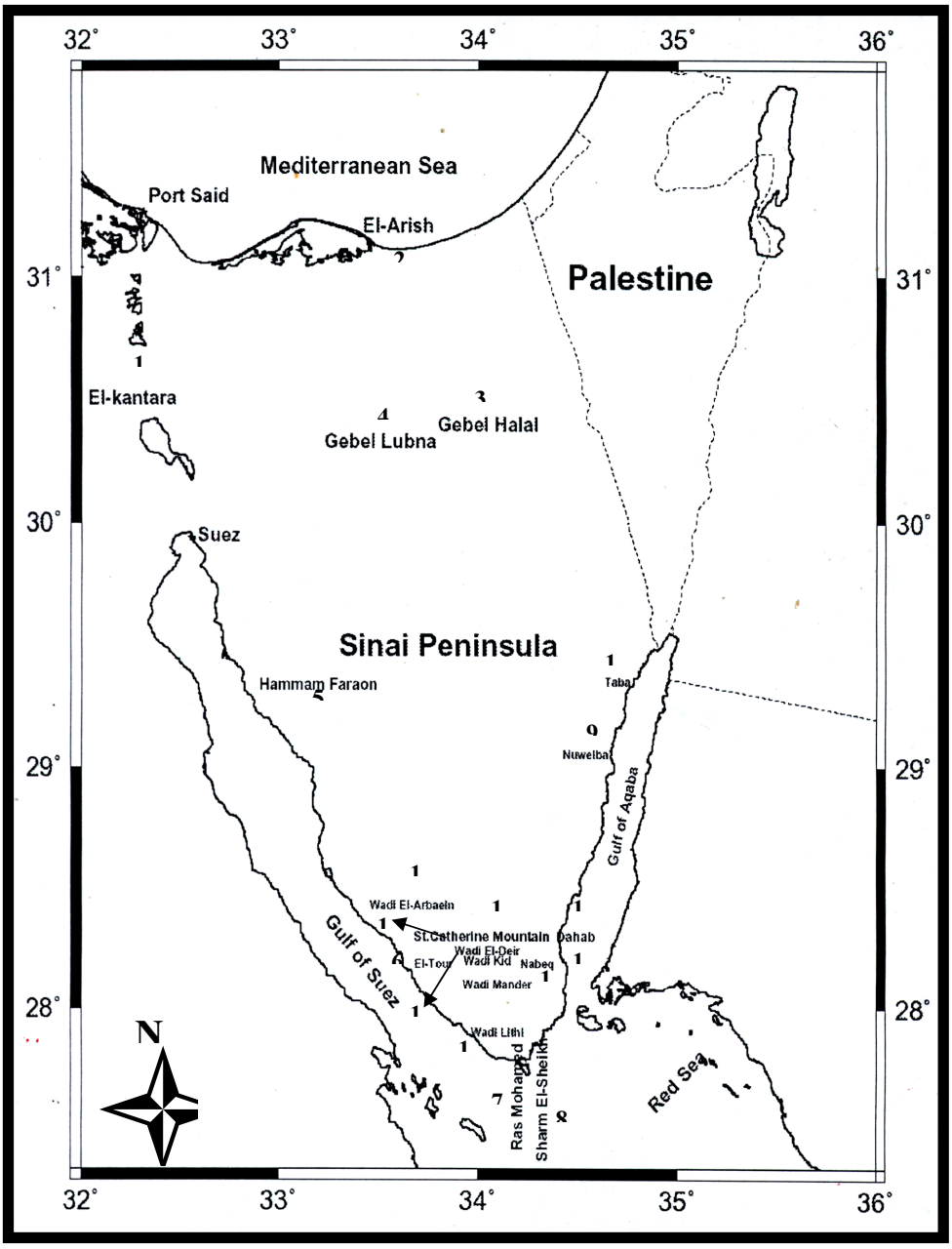
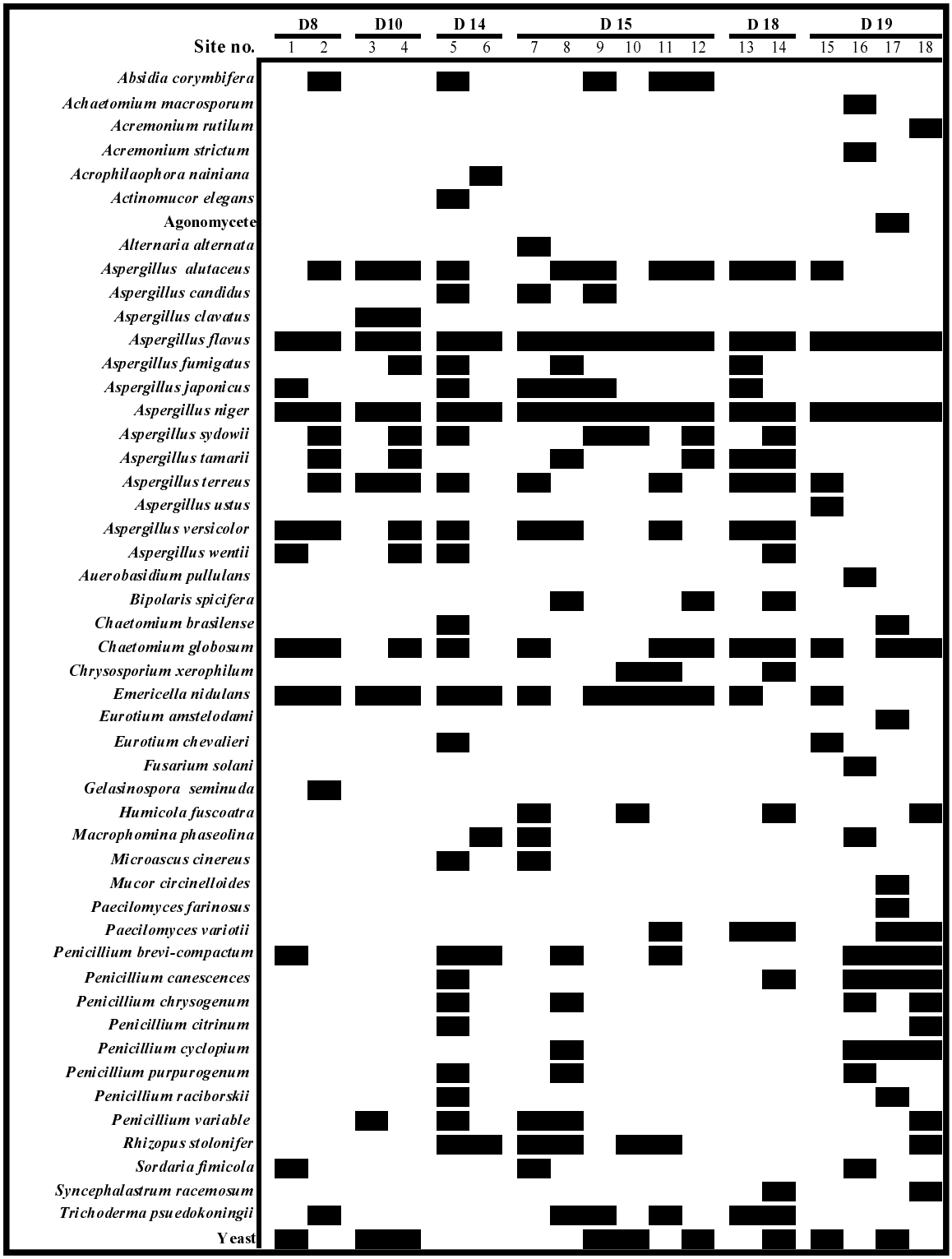


Figure 3: Occurrence of isolated fungi in different sites and districts.



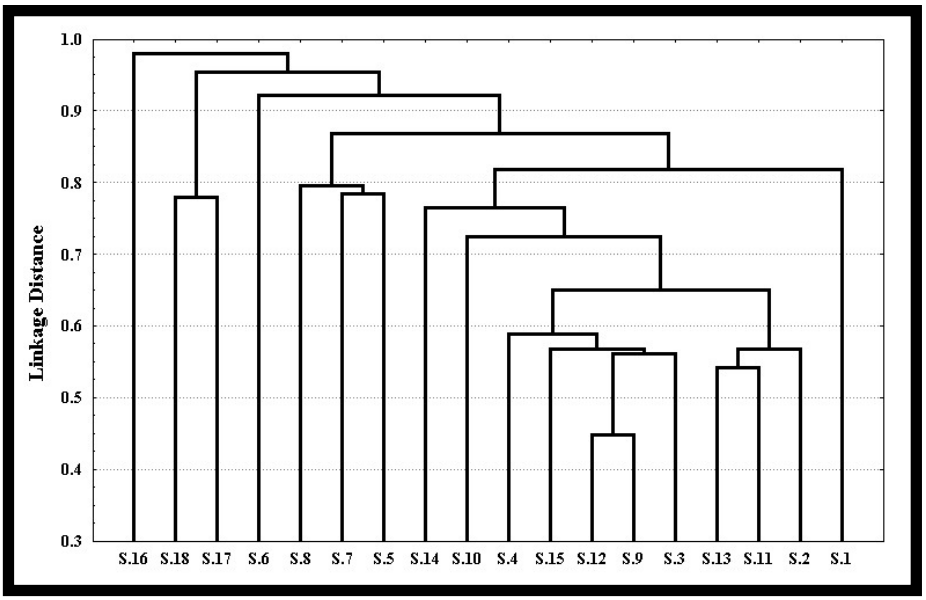


Figure 4: Investigated sites dendogram based on Sorenson's similarity coefficient.

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الملخص العربى

التنوع البيولوجى لفطريات التربة فى سيناء

أحمد محمد أحمد عبد العظيم [١] – محسن السيد إبراهيم [٢]

١- قسم النبات –كلية العلوم -جامعة قناة السويس، الإسماعيلية ٤١٥٢٢- مصر.
٢- قسم العلوم الجيولوجيّة و البيولوجيّة- كلية التربية - جامعة قناة السويس- العريش - مصر.

خلال أكثر من ثمانين عاما من دراسة الفطريات في مصر لاتزال معلوماتنا عن المحتوى الفطرى لتربة سيناء غامضة ومحدودة إلى حد كبير. هذه الفطريات لم تكن أبدا هدفا لأيّ دراسة شاملة من قبل تهتم ببيئتهم وتصنيفهم و تنوعهم البيولوجى. والهدف الرئيسى لهذه الدراسة هو إلقاء بعض الضوء على تركيب وبيئة والتنوع البيولوجى لفطريات التربة فى شبه جزيرة سيناء. تم تجميع عينات من ثمانية عشر موقعا تمثل ستّ مناطق مُختلفة بيئيا. وقد اوضح المسح المبدئى وجود خمسين نوعا من الفطريات تنتمى لسبع وعشرين جنسا سُجِّلتْ من اماكن مختلفة من سيناء. وتصنيفيا وجد أن الانواع المعزولة تنتمى إلى خمسة طوائف وسبعة رتب وعشرة فصائل. وبمقارنة نتيجة هذه الدراسة والدراسات الأخرى التى اجريت على فطريات الصحارى فى مصر أن سيناء لاتتميز بانواع خاصة من الفطريات.