

## THE EFFECT OF CEMENT DUST EMITTED FROM GAZIANTEP CEMENT PLANT ON MICROFUNGUS FLORA OF SURROUNDINGS SOILS, TURKEY

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**Abstract:** The microfungi flora of the soil polluted by the Gaziantep Cement Plant was examined and compared to nearest unpolluted soils. One hundred and sixteen microfungi isolates have been obtained from both two areas. *Penicillium*, *Aspergillus*, *Ulocladium*, and *Cladosporium* genera are the most ones in terms of richness of species. As a result of the quantitative analysis, average of 61.525 CFU/g has been found in the fresh soil, which is equivalent to 1 g oven dried soil. microfungi propagules is 71.500 CFU/g at 5 cm depth and 33.350 CFU/g at 15 cm polluted areas, 117.900 CFU/g at 5 cm and 23.350 CFU/g at 15 cm depth where pollution is not detected. *Rhizopus oryzae*, *Aspergillus fumigatus*, *Penicillium expansum*, *Penicillium humuli*, *Penicillium fagi*, *Embellisia chalydospora* are most common species.

**Key words:** Microfungi, soil, pollution, cement plant, cement dust, Turkey.

### Gaziantep Çimento Fabrikasının Kirlettiği Toprakların Mikrofungus Florası

**Özet:** Bu çalışmada, Gaziantep Çimento Fabrikasının kirlettiği toprakların mikrofungi florası incelenmiş ve bu topraklara en yakın kirlenmemiş toprakların florası ile karşılaştırılmıştır. Her iki alandan toplam 116 mikrofungus izolatu elde edilmiştir. *Penicillium*, *Aspergillus*, *Ulocladium* ve *Cladosporium* tür zenginliği bakımından en fazla bulunan cinslerdir. Kantitatif analiz sonucu, 1 g kurutulmuş toprağa karşılık gelen taze toprakta ortalama 61.525 CFU/g fungus bulunmuştur. Kirlenmiş alanlarda 5 cm derinlikte 71.500 CFU/g ve 15 cm derinlikte 33.350 CFU/g mikrofungus propagülü, kirlenmemiş alanlarda ise 5 cm derinlikte 117.900 CFU/g ve 15 cm derinlikte 23.350 CFU/g bulunmuştur. *Rhizopus oryzae*, *Aspergillus fumigatus*, *Penicillium expansum*, *Penicillium humuli*, *Penicillium fagi*, *Embellisia chalydospora* en yaygın türlerdir.

**Anahtar kelimeler:** Mikrofungi, toprak, kirlilik, çimento fabrikası, çimento tozu, Türkiye.

### Introduction

Environmental pollution is serious problem in Turkey as well as the world. Especially soil pollution may influence profoundly the biology of soil microorganism as well as other organisms. Microorganism activity in the soil is important for the biogeochemical cycles in nature. If microorganism activity affected negatively from pollution and the other environmental condition, it will also have negative effect on the other values of the ecosystem.

The soil microorganism, fungi, actinomycetes, and bacteria, differ in their respective abilities to decompose organic matter, tolerate drought and other forms of stress, their numbers and biomass in the soil, and in the other functions that they perform in the soil. Fungi can breakdown lignin and cellulose and also begin the decomposition of organic matter. They are more resistant to drought than the other microorganisms. Fungi are composed of long strings cells called hyphae that create mycells. Fungi are the least numerous of the soil mi-

croorganisms. Actinomycetes look like fungus but their cells are more like bacteria. Their numbers are intermediate between fungi and bacteria. They can digest some of the hard to digest organic compounds and are somewhat drought resistant. Actinomycetes isolated from soil have provided a number of antibiotics that we use like streptomycin. Bacteria are the most numerous soil organisms. They are not drought tolerant and cannot decompose complex organic compounds. They are very important in N cycling, sulfur chemistry in mine spoils, and global climate change.

The diversity of mycoflora may be affected by certain pollutants. One of the effects of inputs of nitrogen to the soil is to restrict the capacity of mycorrhizal fungi to form their characteristic associations. However, such mechanisms have only been investigated for a small number of pollutants. Other toxic substances that today may not even be recognized as such or may be difficult to detect also represent a potential threat to fungi, particularly with regard to their long-term effects. Pollutants may not only affect mycoflora directly, through the soil, but also - particularly in the case of mycorrhizal fungi - have an indirect influence, via the host plant.

Although individual heavy metals such as cadmium, mercury, lead or even radioactive cesium scarcely affect the growth of fungi, they accumulate in fungal fruiting bodies. Consequently, frequent consumption of such mushrooms may be hazardous to human health.

Singh et al. (1990) have investigated experimentally the affect of cement dust on some wheat phylloplone fungus in India. The percentage frequency and number of colonies per cm<sup>2</sup> leaf area of all of the test fungi decreased at higher doses of cement dust during both pre and post inoculation treatment. Conversely, the population of some fungi only increased at low dose.

Bagy (1992) have investigated content of saprophytic and keratinophilic fungi in cultivated and desert soils, exposed continuously to cement dust, in Egypt. The most common isolated fungi were *Aspergillus niger*, *A. fumigatus*, *A. japonicus*, *A. terreus*, *A. flavus* and *Penicillium funiculosum*.

Hemida (1992) have isolated thermophilic and thermotolerant fungi from cultivated and desert soils, exposed continuously to cement dust particles in Egypt. Ten genera, 16 species and two varieties of *Aspergillus flavus* and *Malbranchia pulchella* were recovered from the soil samples.

Hasenekoğlu and Sülün (1991) have investigated unpolluted and polluted soils, exposed to cement dust, for the first time in Turkey. In this study, it was determined that microfungal flora was affected substantially in a negative way by the dust. An average 36.450 CFU/g have been found in the fresh soil equivalent to 1 g oven-dried soil in two different areas. Comparing the studies near the research area, this value was determined as fairly low. Unpolluted soils were found richer than the polluted soils at both percentage frequency and composition of species. 37 species belong to 10 genera and 11 sterile microfungi were isolated with the soil dilution technique

The first studies on soil microfungi in Turkey were carried out by Öner (1970 and 1974). Studies on soil microfungi in Turkey have primarily concentrated on northeast Anatolia (Hasenekoğlu, 1982; Hasenekoğlu and Azaz, 1991; Hasenekoğlu and Sülün, 1991), the vicinity of İzmir (Ekmekçi, 1974 a,b, 1975; Öner, 1974; Türker, 1979), and Thrace (European part of Turkey)(Asan, 1997 a,b; Asan and Ekmekçi, 1994).

The dust emitted from cement plant may also affect the fungi of soil. The purpose of this study is to determine the negative result of cement dust as an environmental problem on soil fungi and then to compare this with unpolluted soil.

## Material and Methods

### *Description of the research area:*

Gaziantep province lies between latitudes 36° 38" N and 37° 32" N, longitudes 38° 28" and 38° 0" E. The province of Gaziantep is located at the place where the Mediterranean region joins the Southeastern Anatolia and it borders Syria. Gaziantep is taken part in the intermediate zone between Mediterranean and terrestrial climate. Summers are generally hot and dry, but the nights are rather cool. Winter months are cold, with precipitation. The annual average temperature is 14.5° C. The average relative humidity is %60, and the average amount of rain is 556.2 mm. The dominant wind is south or southwest wind and its direction is northwest. Higher parts of the province are covered partly with plantation forests of pine, fir and cedar, and lower zones with shrubs and steppe or semi-steppe flora. Gaziantep cement plant is located about 5 km from east of Gaziantep province centre. It was built in 1961. The soil near the cement plant is not suitable for agriculture. Land

of surrounding of plant is rough and covered with seasonal plant and rarely naturally growing trees. There are plantation pine, cypress trees and bushes in the cement plant borders.

#### *Collection of Samples*

The samples were collected was the region within 7 km from Cement plant. Beyond this distance, it was determined that the effect of the pollution was negligible. Therefore, the twenty samples unaffected by the pollution were collected beyond 7 km from plant from 5 cm and 15 cm depths and (while) twenty the sample affected by pollution were collected from the close cement plant from same depths. The sampling soils are barren and their characters were taken into accounts. Twenty soil samples were collected from 5 cm and 15 cm depths from near the plant and another twenty samples about 7 km distance at which pollution is not appreciable from Gaziantep cement plant. The stations were chosen randomly. For taking of samples, first, a soil profile was dug and then the surface of profile was cleaned and the samples were taken from 5 and 15 cm depths with a disinfected spatula. Samples were kept in cool during the transfer to the laboratory and then refrigerated until they were plated on microbiological growth media.

#### *Culture techniques*

The culture technique used was based on "Soil Dilution Plate Method" described by Waksman 1922, in using this technique, moisture content of a certain amount of soil was determined and fresh soil quantities corresponding to 25 g of oven-dried soil were calculated. Then  $10^{-3}$  dilutions of the samples were prepared and 20 plates of Dextrose-Peptone agar were inoculated from dilution of each sample. 30 mg/L streptomycin and 30 mg/l rose bengal were added to Dextrose-Peptone agar for inhibition bacterial growth and restriction fungal colony.

The plates were kept at 25 °C for 10 days. Culture plates were examined macroscopically and then colonies were enumerated. Different colonies were isolated to Potato-Dextrose Agar and Czapek Dox Agar and identified on these media at ambient temperature.

#### *Physical and chemical characterization of the soil samples*

Soil composition (such as clay, silt, etc.) was characterized and chemical characteristics of the soil (pH, total salt, available phosphorus, lime (CaCO<sub>3</sub>), organic matter) were determined at Laboratory of Soil Department of Agriculture Faculty, Atatürk University. Soil composition was determined with hydrometer of Bouyoucos (Bouyoucos, 1962). Lime content according to volumetric calcimeter methods (Sağlam, 1978), total salt value with Wheatstone bridge (U.S. Salinity Lab.Staft, 1954), soil reaction (pH) using a pH meter with a glass electrode in a mixed soil-water 1:1 ratio (Jackson, 1958), organic matter content with Smith-Weldon method (Hocaoğlu, 1966) and available phosphorus content according to Olsen et al. (1954) was determined.

#### *Identification of microfungi*

Identification of the isolates was performed according to the Raper and Thom (1949), Raper and Fennell (1965), Simmons (1967), Dickinson (1968), Rifai (1969), Zycha et al. (1969), Booth (1971), Ellis (1971), Hanlin (1973), Samson (1974), Bertoldi (1976), Tulloch (1976), Arx (1981) and Hasenekoglu (1991).

#### *Statistical analysis*

Using Mann-Whitney U test in SPSS 11.01 version, the average of the results of the quantitative analysis of microfungi propagules of the polluted soil and unpolluted soil were compared.

## **Results**

The results of pH, organic matter, lime analysis, available phosphorus, total salt and soil composition are given in Table.1. 116 different microfungi were isolated. From those, 81 species belong to the *Moniliales*, 3 species of *Mucorales* and 2 species of *Sphaeriales* and 30 were sterile which have no any fructification. Among these, *Penicillium* spp., *Aspergillus* spp., *Rhizopus* spp., *Polyscytalum* spp. and *Cladosporium* spp. were the most common ones. However, *Penicillium* was found more than the other for both species richness and propagule frequency. *Aspergillus*, *Cladosporium* and *Ulocladium* followed to *Penicillium* from the point of view species richness.

Numbers of colonies and isolates for individual species were illustrated in the Table 3.

In all the samples, an average 61.525 CFU/g have been found in the fresh soil equivalent to 1 g oven-dried soil in two different areas and their two depths. The numbers of average microfungi propagules are 71.500

CFU/g at 5 cm depth and 33.350 CFU/g at 15 cm polluted areas, 117.900 CFU/g at 5 cm and 23.350 CFU/g at 15 cm depth where pollution is not detected.

Characteristics of soil were illustrated in Table 1.

Statistical results of the quantitative analysis of microfungi propagules of the polluted soil and unpolluted soil were given Table 2.

**Table 1.** Physical and chemical characterizations of soil samples (as mean of related samples).

Soil	PH (1:1)	Org. Matter (%)	CaCO <sub>3</sub> (%)	Avail. P (%)	Total salt (%)	Physical properties
Polluted (5 cm)	8.54	3.95	2.80	0.69	0.11	Clay-silt
Polluted (15 cm)	8.75	2.38	2.80	0.69	0.11	Clay-silt
Unpolluted (5 cm)	7.84	2.44	2.50	1.04	0.06	Clay-silt
Unpolluted (15 cm)	7.54	0.8	2.50	1.04	0.06	Clay-silt

**Table 2.** Mann-Whitney U test results of comparison of the quantitative analysis of microfungi propagules of the polluted and unpolluted soils of

	N	X	U	Z	P*
Polluted (5 cm)	10	9,55	40,5	-,718	>0,05
Unpolluted (5 cm)	10	11,45			
Polluted (15 cm)	10	13,05	24,5	1,92	>0,05
Unpolluted (15 cm)	10	7,95			
Polluted (5 cm)	10	14,04	14,5	-2,685	<0,05
Polluted (15 cm)	10	6,95			
Unpolluted (5 cm)	10	14,20	13	-2,797	<0,05
Unpolluted (15 cm)	10	6,80			
Polluted	20	27,8	54	-3,95	> 0,05
Unpolluted	20	13,20			

\* P< 0.05, (there is a significant difference between average of the results of the quantitative analysis of microfungi propagules of the polluted and unpolluted soils)

## Discussion

The number of microfungi propagules was found as average 61.525 CFU/g both in polluted and unpolluted area in the fresh soil, which is equivalent to 1 g oven-dried.

The number and diversity of microfungi of polluted soils by cement dust was low than unpolluted soils (Table 3). However, it was not significant statistically (Table 2). The main cause of this may be the fact that pH of polluted soil was higher than the pH of unpolluted soil. Cement dust affects pH of the soil and increases it. The increase in the amount lime is related to the increase of pH of the polluted soil (Table 1) (Bayhan et al., 2002; Fabbri et al. 2004). As indicated by Bayhan et al. 2002, high value of pH of the soils is an indicator of the pollution with cement dust. According to these authors the increase in the amount of lime is related to the increase in the pH of the polluted soil. The increase in pH causes phosphorous compounds to have a form that plant is unable to absorb them. Consequently, these changes negatively affect the microbial activities of the soil.

Griffin (1972), pointed out that rates of endogenous respiration in fungi are little affected by the hydrogen ion concentration of the external medium over the range pH 5 to 8. Exogenous respiration and growth, however, are affected by change in external pH and are presumably influenced primarily by system located at the cell surface and by changes in the permeability of membranes. pH of the investigated soils was found as over 8 in polluted soils (high alkaline), whereas over 7 in unpolluted soils (low alkaline)(Table 1). Because growth of microfungi occurs better in some acidic soils on the contrary bacteria, probably it may be an adverse affect of this high alkalinity on microfungi growth. But species richness is more in the samples taken from 15 cm depth of polluted soils than in samples taken same depth of unpolluted soils. From this fact it may be said that the species richness is affected from pH variations.

**Table 3.** Number of colonies and isolates for individual species.

Taxa	Colony Number			
	a	b	c	d
<i>Acremonium</i> sp. Link ex Fr.	-	3	-	-
<i>Alternaria alternata</i> (Fr.) Keissl.	-	1	-	-
<i>Alternaria pluriseptata</i> (P. Karst & Har. ex Peck) JØrst.	23	5	-	-
<i>Arhrinium</i> state of <i>Apiospora montagnei</i> Sacc.	5	-	-	-
<i>Aspergillus allahabadii</i> B.S. Mehrotra & Agnihotri	-	1	-	-
<i>Aspergillus alliaceus</i> Thom & Church	-	6	-	-
<i>Aspergillus candidus</i> Link	-	3	-	2
<i>Aspergillus carneus</i> Blochwitz	32	-	-	8
<i>Aspergillus ficuum</i> (Reichardt) Henn.	1	33	81	-
<i>Aspergillus flavus</i> Link	-	2	-	-
<i>Aspergillus foetidus</i> var <i>pallidus</i> (Nakaz., Simo & A.Wat.) Raper & Fannell	-	-	-	2
<i>Aspergillus fumigatus</i> Fresen.	12	243	374	27
<i>Aspergillus kanagawaensis</i> Nehira	-	36	1	2
<i>Aspergillus nidulans</i> (Eidam) G.Winter, Rabenh.	-	5	-	4
<i>Aspergillus niger</i> Tiegh.	4	-	-	-
<i>Aspergillus oryzae</i> (Ahlb.) Cohn var. <i>effusus</i> (Tiraboschi) Ohara	-	-	-	2
<i>Aspergillus reptans</i> Samson & W. Gams	-	-	7	1
<i>Aspergillus sulphureus</i> (Fresen.) Wehmer	-	-	-	51
<i>Aspergillus terreus</i> Thom	-	7	-	1
<i>Aspergillus terricola</i> E.J.Marchal	-	1	-	-
<i>Aspergillus ustus</i> (Bainier) Thom & Church	15	-	-	-
<i>Aspergillus versicolor</i> (Vuill.) Tirab.	10	-	1	-
<i>Chaetomium homopilatum</i> Omvik	1	-	-	-
<i>Chaetomium</i> sp. Kunze ex Fr.	53	-	-	-
<i>Cladosporium cladosporioides</i> (Fresen.) G. A. de Vries	-	-	4	7
<i>Cladosporium elatum</i> (Harz) Nannf.	-	-	-	1
<i>Cladosporium herbarum</i> (Pers) Link	-	7	2	2
<i>Cladosporium oxysporium</i> Berk. & M.A.Curtis	-	7	42	3
<i>Coccidioides immitis</i> G.W. Stiles	-	4	-	-
<i>Drechslera</i> state of <i>Cochliobolus spicifer</i> R.R. Nelson	-	1	-	-
<i>Curvularia sorghina</i> R.G. Shivas & Sivan.	-	1	-	-
<i>Drechslera australiensis</i> (Bugnic.) Subram.& B.L. Jain	2	6	-	-
<i>Embellisia chalmydospora</i> (Hoes, G.W. Bruehl & C.G. Shaw) E.G. Simmons	153	10	-	-
<i>Fusarium</i> sp. Link ex Fr.	-	3	-	-
<i>Geomyces pannorum</i> (Link) Sigler & J.W. Carmich. var. <i>pannorum</i> Oorschot	-	5	2	-
<i>Humicola grisea</i> Traaen	7	-	-	-

Taxa	Colony Number			
	a	b	c	d
<i>Humicola insolens</i> Cooney & Emers.	1	-	-	-
<i>Humicola</i> sp. Traen	-	-	-	1
<i>Mortierella antarctica</i> Linnem.	-	-	-	3
<i>Mucor circinelloides</i> Tiegh. f. <i>lusitanicus</i> (Bruderl.) Schipper	-	-	-	1
<i>Myrothecium roridum</i> Tode	-	-	1	2
<i>Paecilomyces lilacinus</i> (Thom) Samson	-	1	-	9
<i>Penicillium aeneum</i> G.Sm.	-	3	-	-
<i>Penicillium alicantinum</i> C.Ramirez & A.T.Martinez	-	-	43	2
<i>Penicillium atramentosum</i> Thom	-	-	-	8
<i>Penicillium brevicompactum</i> Dierckx	2	19	9	7
<i>Penicillium canescens</i> Sopp.	--	-	1	5
<i>Penicillium chermesinum</i> Biourge	-	3	-	-
<i>Penicillium chrysogenum</i> Thom	141	-	-	1
<i>Penicillium corylophium</i> Dierckx	43	2	74	-
<i>Penicillium echinulatum</i> Raper and Thom ex Fassat.	-	-	1	-
<i>Penicillium expansum</i> Link	28	6	202	8
<i>Penicillium fagi</i> C.Martinez & A.T.Ramirez	180	5	-	8
<i>Penicillium frequentans</i> Westling	10	-	-	-
<i>Penicillium humuli</i> J.F.H. Beyma	27	7	722	-
<i>Penicillium implicatum</i> Biourge	3	-	-	-
<i>Penicillium islandicum</i> Sopp.	-	-	-	2
<i>Penicillium italicum</i> var. <i>italicum</i> Wehmer	-	-	-	1
<i>Penicillium janthinellum</i> Biourge	-	1	3	79
<i>Penicillium jensenii</i> K.M.Zalessky	-	-	-	2
<i>Penicillium kojigenum</i> S. G.Sm.	-	1	-	-
<i>Penicillium miczynski</i> K.M.Zalessky	-	2	-	-
<i>Penicillium olsonii</i> Bainier & Sartory	1	-	-	-
<i>Penicillium oxalicum</i> Currie & Thom	-	5	3	3
<i>Penicillium purpurogenum</i> Stoll	-	-	-	10
<i>Penicillium roqueforti</i> Thom	6	-	-	-
<i>Penicillium sartoryi</i> Thom	-	3	-	-
<i>Penicillium simplicissimum</i> (Oudem.)Thom	12	19	-	-
<i>Penicillium stoloniferum</i> Thom	61	-	-	-
<i>Penicillium velutinum</i> J.F.H. Beyma	-	-	-	10
<i>Penicillium verrucosum</i> Dierckx var. <i>corymbiferum</i> (Westling) Samson, Stolk & Hadlok	-	1	-	16
<i>Penicillium verrucosum</i> Dierckx var. <i>cyclopium</i> (Westling) Samson, Stolk & Hadlok	90	3	4	-
<i>Penicillium verrucosum</i> Dierckx var. <i>ochraceum</i> (Bainier) Samson, Stolk & Hadlok	-	11	139	2
<i>Penicillium waksmanii</i> K.M.Zalessky	4	5	-	-
<i>Phialophora</i> sp. Medlar	-	-	-	1
<i>Polyscytalum fecundissimum</i> Riess	39	2	7	4
<i>Polyscytalum pustulans</i> (M.N. Owen & Wakef.) M.B. Ellis	-	-	-	2
<i>Polyscytalum</i> sp. Riess	29	-	-	-
<i>Rhizopus oryzae</i> Went & Prins. Geerl.	97	83	449	37
<i>Stachybotrys microspora</i> (B.L. Mathur & Sankhla) S.C. Jong & E.E. Davis	1	-	-	-
<i>Trichoderma harzianum</i> Rifai	1	-	-	-
<i>Ulocladium atrum</i> Preuss	9	-	18	-
<i>Ulocladium chartarum</i> (Preuss) E.G. Simmons	8	-	-	-
<i>Ulocladium chlamydosporum</i> Mouch.	1	-	-	-

Taxa	Colony Number			
	a	b	c	d
<i>Ulocladium tuberculatum</i> E.G. Simmons	2	-	-	-
Sterile 1	38	1	-	-
Sterile 2	22	-	-	-
Sterile 3	1	-	-	-
Sterile 4	1	-	-	-
Sterile5	125	1	-	1
Sterile 6	2	6	-	1
Sterile 7	1	-	1	2
Sterile 8	27	3	80	-
Sterile 9	6	1	42	7
Sterile 10	70	-	-	2
Sterile 11	23	-	-	1
Sterile 12	-	-	2	1
Sterile 13	-	-	2	-
Sterile 14	-	-	1	2
Sterile 15	-	12	1	15
Sterile 16	-	2	2	-
Sterile 17	-	-	18	-
Sterile 18	-	10	1	-
Sterile 19	-	17	10	74
Sterile 20	-	1	7	1
Sterile 21	-	13	-	1
Sterile 22	-	-	-	-
Sterile 23	-	17	1	-
Sterile 24	-	3	-	-
Sterile 25	-	3	-	-
Sterile 26	-	3	-	-
Sterile 27	-	-	-	3
Sterile 28	-	-	-	4
Sterile 29	-	-	-	8
Sterile 30	-	2	-	7

(a: The soils, exposed to cement dust (5cm depth), b: The soils, exposed to cement dust (15 cm depth), c: The soils, pollution is not evident (5cm depth), d: The soils, pollution is not evident (15cm depth)

The number of microfungi propagules of polluted soil was lower than the unpolluted soil sampled at 5.0 cm. It was no statistically significant (Table 2). This may be due to the high pH value of polluted soil.

Conversely, the number of microfungi propagules of polluted soil sampled at 15 cm was more than unpolluted soil sampled from the same depth that there was no statistically significant (Table 2). However the cause of this may be due to the fact that the amount of organic matter of unpolluted soil was quite low (Table 1). Polluted areas have more species than unpolluted ones at both depths (Table 3).

In our study, average number of microfungi propagules of the polluted soils at 5 cm was fairly high in comparison with that of 15 cm depth. This difference was found as statistically significant (Table 2). The reasons of this may be due to the soils of 5 cm depth nearer to the surface and consequently they were well aerated and organic matter content of them was slightly higher than the deeper soils layers. There was a quite distinction between the numbers of microfungi propagule at 5 and 15 cm depths of soils samples taken from the area which pollution is not evident. The average of microfungi number of the soils sampled from 15 cm of these areas was highly lower than the microfungi number of samples of 5 cm depth of the same areas. It may be due to low organic matter content of 15 cm depth of this soil (Table 1). Statically, this was significant (Table 2). But species richness of 15 cm depths of both polluted and unpolluted areas were more than 5 cm depths. Actually 38 taxa from 5 cm depth, 42 taxa were from 15 cm in the polluted soils, and 24 different microfungi from 5 cm depth and 39 taxa from 15 cm depth in the unpolluted soils were isolated.

Soil dilution plate method, which has developed for bacterial isolation from soil, is regarded also as the best method for the determination of mycoflora of soil. However, this method is not convenient to use to determine the activity of microfungi in soil. When used for the microfungi there are serious objections to the method. The most important of these is that this method will favor the species, which produce abundant spores whereas in this method some microfungi such as Basidiomycetes taxa are less evaluated than its actual value.

To decrease the mistakes during the application of DPT (dilution plate technique), the number of the Petri dishes has been increased in our study. The number of Petri dishes inoculated was found directly proportional to the number of the species obtained (Hasenekoğlu and Sülün, 1991). Therefore, parallel inoculations have been performed as 20 Petri dishes for each soil sample.

*Penicillium*, *Aspergillus*, *Rhizopus* and *Polysctalum* were the most common genera both in the areas and depths. Furthermore, *Penicillium* was most common as both frequency and species richness (Table 3).

There were inequalities in the composition of microfungi between polluted soils and unpolluted soils. Actually, *Aspergillus niger*, *A. ustus*, *Penicillium frequentans*, *P. implicatum*, *P. restrictum*, *P. olsonii*, *P. stoloniferum*, *Trichoderma harzianum*, *Ulocladium tuberculatum*, *U. chartarum*, *Arhrinium* state of *Apiospora montagnei*, *Stachybotrys microspora*, *Humicola grisea*, *H. insolans*, *Chaetomium homopilatum*, *Chaetomium* sp., *Polyschatulum* sp. were isolated only from polluted soils which was sampled from 5 cm depth, *Aspergillus alliaceous*, *A. flavus*, *A. terricola*, *A. nidulans*, *A. allahabadii*, *Penicillium aeneum*, *P. chermesinum*, *P. sartoryi*, *P. miczynski*, *P. kojigenum*, *Fusarium* sp., *Acremonium* sp., *Curvularia sorghina*, *Alternaria alternata*, *Drechslera* state of *Coeliobolus spicifer*, *Coccidioides immitis* from only polluted soils and 15 cm depth (Table 3). The *Alternaria alternata* *Penicillium frequentans* and *P. stoloniferum* species were obtained also by Hasenekoğlu and Sülün(1991) from only polluted soils. Furthermore *Alternaria alternata* were isolated from polluted air (Schoenlin-Crusius et al. 2001). This may be interpreted that these species have tolerance the pollution.

*Penicillium echinulatum* from only 5 cm depth in unpolluted soil, *Aspergillus sulphureus*, *A. oryzae* var. *effusus*, *A. foetidus* var. *pallidus*, *Penicillium velutinum*, *P. jensenii*, *P. purpurogenum*, *P. atramentosum*, *P. italicum* var. *italicum*, *P. islandicum*, *Cladosporium cladosporioides* *C. elatum*, *Humicola* sp., *Phielophora* sp., *Mucor circinelloides* f. *lusitanicus*, *Mortierella antarctica*, *Polyschatulum pustulans* from only 15 cm in unpolluted soils (Table 3).

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