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# Fungi isolated from the initial industrial soil planted with a mixture of alfalfa and grasses

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Research were conducted during the years 1988-1990 on the "Machów" industrial waste dump. It was determined, that a mixture of alfalfa with grasses, introduced as first culture on the raw substrate of industrial wastes, was helpful for the development of soil fungi communities.

## INTRODUCTION

The studies on the reclamation soil-less lands in the industrial waste dumps of sulphur surface mines showed that the plantations of alfalfa with grasses improve the physical and chemical properties of the initial in industrial soil by initiating and stimulating soil development processes (K o w a l i k, 1989 a, b).

In the current mycological literature only few studies concern the communities of soil fungi of reclaimed soil-less lands (K o w a l i k, K o w a l i k, 1982; K o w a l i k, 1990, 1993; M a d e j, 1992). Nevertheless, the activity of soil microorganisms such as bacteria and fungi reclaimed industrial waste lands determines the course and dynamics of the soil development processes. Moreover, it also determines the productivity of the reclaimed indusrial waste lands, during the first years of cultivation. Therefore, the changes in the species composition and diversity of soil fungi reflect the changes which occur in the initial soil (R i c h a r d s, 1977; K o w a l i k, 1990). These processes may also help in improving the system of land managment and have a positive effect on the development of fertile and highly productive soils.

The aim of the present study was to determine fungal communities isolated from the initial soil planted with a mixture of alfalfa and grasses on the "Machów" dumping ground and assess the changes occurring in the qualitative and quantitative composition of soil fungi depending on the time of application of the mixture.

## DESCRIPTION OF THE STUDY AREA

Studies were carried out in the outer industrial waste dumps of "Machów" surface sulphur mine near Tarnobrzeg.

The soil of the „Machów" industrial waste dump are characterized by high soil compaction (65-85 % of silt and clay, mean specific gravity of 2.75 g/cm$^3$, high bulk density of 1.75 g/cm$^3$, low porosity of 36.0 % and plasticity index of 32.7 %. Their mechanical composition is the same as of loam or clay loam. They are impermeable and poorly aerate, cold and expandable. Mechanical cultivation is very difficult there. They have the character of „minute" soils and are distinguished by neutral or slightly alkaline pH, a considerable content of $CaCO_3$, K and Mg, and a poor content of P and N. The content of the 3 forms of sulphur is:
S og. – 1.46 %, S el. – 0.03 % and S-$SO_4$ – 0.20 %. The content of all the macro- and microelements varies in the range regarded as normal or admissible for natural soils.

The investigated area was described on the basis of works by F u k et al. (1981), K o w a l i k (1989 b), K r z a k l e w s k i et al. (1989).

The studies were carried out on three experimental fields of the industrial waste dump, differing in age and period of agricultral use:

Field III – the ledge of an escarpment formed in 1986 and planted with a mixture of alfalfa and grasses in spring 1987.

Field VI – a plateau formed in 1976 and cultivated since 1977; planted with a mixture of alfalfa and grasses in spring 1986.

Field IX – the ledge of an escarpment formed in 1984 and palnted with a mixture of alfalfa and grasses in spring 1985.

## MATERIAL AND METHODS

Mycological studies of the initial soils of the "Machów" waste dump were conducted in the years 1988-1990 using sand samples (M a ń k a, 1974). Fungi from the investigated fields on the dumping ground planted with a mixture of alfalfa and grasses were isolated at 5 dates:

A – at the beginning of the vegetation season – 1st or 2nd decade of April,
B – after the first harvest of alfalfa and grasses – 1st decade of June,
C – after the second harvest of alfalfa and grasses – the 3rd decade of June in 1988 and 2 nd decade of July in 1989 and 1990,
D – after the third harvest of alfalfa and grasses – 1st decade of September,
E – after the first ground frosts – 1st and 2nd decade of November.

At each isolation period the mixture of soil and sand (mass of 28 mg in weigh) was placed on 30 Petri dishes and a cooled Martin-Johnson medium was added.

Fungi were incubated at room temperature. Subsequently, colonies of fungi were isolated and transferred to test-tubes containing obligue solid 2 % PDA medium.

For the determination of fungi species the mycological keys and the monography given by K o w a l i k (1993) were employed.

## RESULTS

In fungal communities isolated from soils of 3 plantations the occurrence of 40 species of fungi of 23 genera was ascertained (Table 1).

The most abundantly occuring species were: *Aspergillus oryzae* – 534 colonies, *A. versicolor* – 297, *Chaetomium olivaceum* – 892, *Ch. globosum* – 228, *Doratomyces stemonitis* – 148, *Paecilomyces farinosus* – 147, *Penicillium granulatum* – 174, *Phoma eupyrena* – 218, *Scopulariopsis brumptii* – 382, *Trichoderma viride* – 113 and *Verticillium albo-atrum* – 81.

Fungal communities of the soil environment differed by the number of colonies of various species.

The most numerous fungal community (2844 colonies and 88 species) occurred in the soil where the mixture of alfalfa with grasses was cultivated for 2-4 years (field VI – in agricultural use for several years). The smallest number of colonies and species of fungi (1253 and 64, respectively) was isolated from the soil which had been cultivated for 3-5 years (field IX). An intermediate number of 1590 colonies and 82 species of fungi was isolated from soils of 1-3 years old plantations (field III).

In the soils covered with a mixture of alfalfa and grasses for 1-3 years, the greatest number of colonies and fungi species was noted in field III in the first year of the study (1988). In the following years, a considerable decline in the number of colonies of fungi was accompanied by a decrease in the number of species which remained stable afterwards.

The following species of fungi previled in this type of soil environment: *Aspergillus* (mainly *A. oryzae* and *A. versicolor*) – 19.4 %, *Penicillium* (*P. expansum*, *P. granulatum* i *P. sclerotium*) – 13.5 %, *Scopulariopsis* (*S. brumptii* i *S. chartarum*) – 7.5 %, *Doratomyces* (*D. stemonitis*) – 6.6 %, *Phoma* (*P. eupyrena*, *P. medicaginis*) – 6.5 %.

In the group of sub-dominants the most frequently occurring species were: *Chaetomium* (mainly *Ch. olivaceum*) – 4.9 %, *Mortierella* (*M. isabellina*, M. *parvispora* and *M. vinacea*) – 4.8 %, *Verticillium* (*V. albo-atrum* and *V. lecanii*) – 4.5 %, *Gliocladium* (*G. catenulatum*) – 4.4 %, *Fusarium* (*F. culmorum*, *F. oxysporum* and *F. tabacinum*) – 3.1 %, *Trichoderma* (*T. viride* and *T. piluliferum*), *Acremonium murorum* – 2.6 %, *Cladosporium herbarum* – 2.4 %, and *Torula graminis* – 1.8 %.

Table 1

Fungi isolated from the soil under the plantations of alfalfa with grasses in the fields III, VI, IX

| Species | Number of isolates in years | | | | | | | | |
|---|---|---|---|---|---|---|---|---|---|
| | 1988 | 1989 | 1990 | 1988 | 1989 | 1990 | 1988 | 1989 | 1990 |
| | Field No. | | | | | | | | |
| | III | | | VI | | | IX | | |
| *Absidia glauca* Hagem | . | 3 | . | . | 6 | 3 | . | 1 | . |
| *Acremoniella atra* (Corda) Sacc. | 22 | . | . | . | . | . | 4 | . | . |
| *Acremonium furcatum* (F. et M. Moreau) ex W. Gams. | . | . | 7 | . | 12 | . | . | . | . |
| *A. kiliense* Grütz | . | . | 9 | . | . | . | . | . | . |
| *A. murorum* (Corda) W. Gams | 6 | 17 | 3 | 8 | 53 | 31 | 1 | 4 | 24 |
| *Acropialophora fusispora* (Saksena) Samson | 4 | . | . | 2 | . | . | 3 | . | . |
| *Alternaria alternata* (Fr.) Keissler | 5 | 18 | . | 9 | . | 3 | 2 | . | . |
| *A. tenussima* (Kunze ex Pers.) Wilts. | . | . | . | . | . | 6 | . | . | . |
| *Arachniotus terrestris* Raillo | 1 | . | . | . | . | . | . | . | . |
| *Arthrinium phaeospermum* (Corda) M. B. Ellis | 2 | . | . | 4 | . | . | . | 1 | . |
| *Aspergillus candidus* Link ex Link | . | . | . | . | . | . | . | 2 | . |
| *A. clavatus* Desm. | . | 4 | . | . | 63 | . | . | 2 | . |
| *A. niger* v. Tiegh. | 4 | . | . | 3 | . | . | 4 | . | . |
| *A. oryzae* (Ahlb.) Cohn | . | . | 207 | . | . | 122 | . | . | 205 |
| *A. versicolor* (Vuill.) Tirab. | 64 | 17 | 12 | 36 | 18 | 32 | 31 | 51 | 36 |
| *Chaetomium cochlioides* Pall. | 8 | 2 | . | 2 | . | . | . | . | . |
| *C. globosum* Kunze et Steud. | 6 | . | . | 7 | 98 | 81 | 16 | 2 | 18 |
| *C. olivaceum* Cooke et Ellis | 26 | . | 36 | 301 | 116 | 407 | 6 | . | . |
| *Chrysosporium asperatum* Carm. | . | . | . | 2 | . | . | . | . | . |
| *C. pannorum* (Link) Hughes | 8 | . | . | . | . | . | . | . | . |
| *Cladosporium cladosporioides* (Fres.) de Vries | . | . | . | . | 3 | 3 | . | . | . |
| *C. herbarum* (Pers.) Link ex S. F. Gray | 17 | 3 | 12 | 25 | 5 | 24 | 10 | 8 | . |
| *C. macrocarpum* Preuss | . | . | 7 | 17 | . | . | . | . | 1 |
| *C. sphaerospermum* Penz | . | . | . | . | . | . | 7 | 16 | 3 |
| *Coniothyrium minitans* Campbell | . | . | . | . | . | . | 1 | . | . |
| *Cylindrocarpon destructans* (Zins.) Scholten | . | . | 5 | 3 | 58 | 2 | . | . | . |
| *Dendryphion nanum* (C. G. Nees ex S. F. Gray) Hughes | . | . | . | . | . | 2 | . | . | . |
| *Doratomyces microsporus* (Sacc.) Morton et G. Smith | . | 1 | . | 2 | . | . | . | . | . |
| *D. nanus* (Ehrenb. ex Link) Morton et G. Smith | 1 | . | . | . | . | 3 | 1 | . | . |
| *D. stemonitis* (Pers. ex Steut) Morton et G. Smith | 88 | . | 15 | 10 | 10 | 24 | 1 | . | . |
| *Fusarium avenaceum* (Fr.) Sacc. | . | . | . | 8 | . | 15 | 4 | . | . |
| *F. concolor* Reinking | 4 | . | . | 10 | . | . | . | 7 | . |
| *F. culmorum* (W. G. Smith) Sacc. | 3 | . | 21 | 5 | . | 13 | . | . | . |
| *F. equiseti* (Corda) Sacc. | . | . | . | 2 | . | . | . | . | . |
| *F. heterosporum* Nees. et Fr. | . | . | . | 2 | . | . | . | . | . |
| *F. nivale* (Fr.) Ces. | . | . | . | . | . | 9 | 6 | . | . |
| *F. oxysporum* Schlecht. emend. Snyd. et Hans. | 8 | . | 2 | 28 | . | 6 | 1 | . | . |
| *F. poae* (Peck) Wollenw. | . | . | . | 6 | 23 | 3 | . | . | . |

cont. Tab. 1

| | | | | | | | | | | |
|---|---|---|---|---|---|---|---|---|---|---|
| *F. semitectum* Berk et Rav. | . | . | . | . | . | 8 | . | . | . |
| *F. solani* (Mart.) Sacc. | . | . | . | 1 | 5 | . | . | . | . |
| F. tabacinum (Beyma) W. Gams | 11 | . | . | 7 | 2 | . | . | 11 | . |
| *Gliocladium catenulatum* Gilman et Abbott | 52 | . | 3 | . | 9 | . | . | . | . |
| *G. roseum* Bain. | 6 | . | . | 4 | . | . | . | . | . |
| *G. virens* Miller, Ciddens et Foster | 9 | . | . | . | . | . | . | . | . |
| *Gymnoascus reessii* Baran | . | . | 1 | . | . | . | . | . | . |
| *Humicola fuscoatra* Traaen var. *fuscoatra* | . | . | 2 | . | 1 | 6 | . | 28 | . |
| *Mammaria echinobotryoides* Cesati | . | . | 3 | . | . | 2 | . | . | . |
| *Metarrhizium anisopliae* (Metschn.) Sorok. | . | 6 | . | . | . | . | . | . | . |
| *Microdochium bolleyi* (Sprague) de Hoog and Hermanides-Nijhof | . | 10 | . | . | . | . | . | . | . |
| *Minimedusa polyspora* (Hotson) Weresub et Le Clair | . | . | . | . | . | . | . | 8 | . |
| *Monilia geophila* Oudem. | . | . | 13 | . | . | 25 | . | 24 | 7 |
| *M. implicata* Gilman et Abbott | 1 | . | . | . | . | . | 10 | . | . |
| *Mortierella alpina* Peyronel | . | . | 4 | 6 | 1 | . | . | . | . |
| *M. isabellina* Oudem. | 20 | 5 | 11 | 22 | 3 | 9 | 16 | . | 3 |
| *M. parvispora* Linnem. | 13 | 8 | . | 6 | . | . | 38 | 2 | . |
| *M. stylospora* Dixon-Stewart | 3 | . | . | . | . | . | . | . | . |
| *M. vinacea* Dixon-Stewart | 12 | . | . | 1 | 6 | . | 6 | . | . |
| *Mucor hiemalis* Wehmer f. *hiemalis* | . | . | . | . | . | 8 | . | . | . |
| *M. racemosus* Fres. f. *sperosporus* (Hagem) Schipper | . | . | . | 9 | . | . | . | . | . |
| *Nectria inventa* Pethybr. | . | 2 | 6 | 17 | 4 | 22 | . | . | . |
| *Paecilomyces farinosus* (Holme ex Gray) A. H. S. Brown and G. Sm. | . | 47 | 2 | . | 36 | . | . | 36 | 26 |
| *P. lilacinus* (Thom) Samson | . | . | . | . | 2 | 2 | 1 | 23 | . |
| *Papulaspora irregularis* Hotson | 2 | . | . | . | . | . | 11 | . | . |
| *Penicillium chrysogenum* Thom | . | . | . | . | . | . | . | . | 6 |
| *P. claviforme* Bain. | . | 22 | . | . | . | . | . | . | . |
| *P. echinulatum* Fassatiová | 2 | . | . | . | . | . | . | . | . |
| *P. expansum* (Link ex Gray) Thom | 66 | . | 4 | 26 | . | . | . | . | . |
| *P. frequentans* Westl. | 10 | . | 38 | . | 14 | . | . | 26 | . |
| *P. funiculosum* Thom | . | . | . | . | . | . | . | 1 | . |
| *P. granulatum* Bain. | . | 22 | . | . | 4 | . | . | 148 | . |
| *P. hordei* Stolk | . | . | . | . | . | . | . | . | 3 |
| *P. janthinellum* Biourge | . | . | . | . | 12 | 3 | . | . | . |
| *P. jensenii* Zaleski | 3 | . | 5 | 6 | . | . | 1 | 1 | . |
| *P. odoratum* Christensen et Backus | . | . | 9 | . | . | . | . | . | . |
| *P. oxalicum* Currie et Thom | . | 6 | . | . | . | . | . | . | . |
| *P. purpurogenum* Stoll | . | . | . | . | 18 | . | . | . | . |
| *P. restrictum* Gill. et Abbott | 4 | . | . | . | . | . | . | . | . |
| *P. sclerotiorum* v. Beyma | . | . | 10 | . | . | . | . | . | . |
| *P. spinulosum* Thom | . | 4 | . | . | 2 | . | . | . | . |
| *P. steckii* Zaleski | . | . | . | 1 | . | 1 | 22 | 30 | . |
| *P. verrucosum* Dierckx var. *cyclopium* (Westl.) Samson | . | . | . | 2 | . | . | 4 | . | . |
| *P. verucosum* Dierckx var. *verrucosum* Samson | 4 | . | . | 15 | . | . | . | . | . |
| *P. verruculosum* Peyronel | . | 1 | . | . | . | 8 | . | . | . |
| *P. waksmanii* Zaleski | . | . | 4 | 1 | 1 | 14 | . | . | . |
| *Pestalotia hartigii* Tubeuf | . | . | . | 6 | 18 | 2 | . | . | . |
| *Phoma chrysanthemicola* Hollós | . | 10 | . | . | . | . | . | . | 18 |

cont. Tab. 1

| Species | Number of isolates in years | | | | | | | | |
|---|---|---|---|---|---|---|---|---|---|
| | 1988 | 1989 | 1990 | 1988 | 1989 | 1990 | 1988 | 1989 | 1990 |
| | Field No. | | | | | | | | |
| | III | | | VI | | | IX | | |
| *P. eupyrena* Sacc. | 2 | 14 | 14 | 2 | 150 | . | 6 | 24 | 6 |
| *P. leveillei* Boerma and Bollen. | 17 | . | 3 | 10 | . | . | . | . | 5 |
| *P. medicaginis* Malbr. et Roum. | 26 | 17 | . | . | 2 | . | 17 | . | 10 |
| *P. putamium* Speg. | . | . | . | . | . | . | . | 5 | . |
| *Phymatotrichopsis omnivora* (Duggar) Henneb. | 1 | 3 | . | . | . | . | . | . | . |
| *Plectosphaerella cucumerina* (Lindf.) W. Gams | . | 1 | . | . | . | . | . | . | . |
| *Pseudeurotium zonatum* v. Beyma | 1 | 3 | 19 | . | 22 | 10 | 5 | . | 7 |
| *Pseudogymnoascus roseus* Raillo | 6 | . | . | 8 | . | . | 4 | . | 2 |
| *Pythium oligandrum* Drechsler | . | . | . | 3 | . | . | . | . | . |
| *Rhizopus stolonifer* (Ehrenb. ex Link) Lind | . | . | . | . | . | 1 | . | 13 | . |
| *Sclerotnia sclerotiorum* (Lib.) de Bary | . | . | . | . | . | . | . | 23 | . |
| *Scopulariopsis brevicaulis* (Sacc.) Bain. | . | 2 | . | 7 | 3 | 6 | . | . | . |
| *S. brumptii* Salvanet-Duval | 23 | 34 | 35 | 74 | 129 | 32 | 6 | 36 | 15 |
| *S. chartarum* (G. Smith) Morton et G. Smith | 20 | 6 | . | 12 | . | . | 2 | . | . |
| *S. fusca* Zach. | . | . | . | . | 3 | . | . | . | . |
| *Sordaria fimicola* (Rob.) Ces. ex de Not. | . | . | . | . | 3 | . | . | . | . |
| *Sporormia ambigua* Niessl | 4 | . | . | 54 | 6 | 7 | . | . | 3 |
| *Stachybotrys chartarum* (Ehrenb. ex Link) Hughes | . | . | . | 12 | . | 6 | . | . | . |
| *Staphylotrichum coccosporum* J. Meyer and Nicot | . | . | . | . | 12 | 3 | . | . | . |
| *Thanatenphorus cucumeris* (Frank) Donk | . | . | . | 1 | . | . | . | . | . |
| *Torula graminis* Desm | 22 | 6 | . | 1 | 4 | . | 2 | . | . |
| *T. herbarum* Pers. ex Gray f. *herbarum* | . | . | . | . | . | . | . | . | 5 |
| *Trichoderma harzianum* Rifai | . | . | 3 | . | 2 | . | . | 8 | 12 |
| *T. piluliferum* Webster et Rifai | 10 | . | . | 5 | . | 13 | 7 | . | . |
| *T. pseudokoningii* Rifai | . | . | . | . | . | 2 | . | . | . |
| *T. viride* Pers. ex S. F. Gray | . | 37 | . | 4 | 6 | 43 | . | 13 | 10 |
| *Trichurus terrophilus* Swift et Povah | 14 | . | . | 28 | . | . | 4 | . | . |
| *Ulocladium consortiale* (Thüm.) Simmons | . | . | . | . | . | . | . | . | 5 |
| *Verticillium albo-atrum* Reinke et Berth. | . | 13 | 39 | . | 4 | 11 | . | 4 | . |
| *V. lecanii* (Zimm.) Viégas | 18 | . | . | 14 | . | . | 1 | . | . |
| *V. nigrescens* Pethybr. | 2 | . | . | 12 | . | . | 3 | . | . |
| *Volutella ciliata* (Alb. et Schw.) Fr. | . | 1 | . | . | . | . | . | . | . |
| Species non determ. | | | | | | | | | |
| *Dematiaceae* T III-5 | 16 | . | . | | | | | | |
| *Mucedinaceae* A III-38 | 4 | . | . | 4 | . | . | | | |
| Species non sporul. | | | | | | | | | |
| *Mucedinaceae* N VI-71 | | | | . | 2 | . | | | |
| Total isolates/species | 681 | 345 | 564 | 870 | 951 | 1023 | 264 | 558 | 430 |
| | 50 | 32 | 33 | 55 | 43 | 43 | 36 | 30 | 23 |

In the environment of soil covered with a mixture of alfalfa and grasses for one year, the occurrence of the "pioneer" species was distinctly manifested: *Acremoniella atra, Acrophialophora fusispora, Doratomyces* (especially *D. stemonitis*), *Gliocladium* (especially *G. catenulatum and G. virens*), *Mortierella (M. isabellina, M. parvispora* and *M. vinacea), Penicillium (P. expansum)* and *Scopulariopsis (S. brumptii* and *S. chartarum).*

In the soil cultivated with the mixture of alfalfa and grasses for 1-3 years, the fungal community was characterized by exceptionally great number of species from the genus *Penicillium.* In total, 14 species of the above genus were isolated. Most of them, however, colonized the soil during only one vegetative season. In this group, the most frequent species were: *P. claviforme, P. expansum, P. frequentans,* and *P. granulatum.*

Fungi from the genus *Fusarium* were represented by 4 species : in this number, *F. culmorum* and *F. oxysporum* regarded as pathogenic. They were isolated during the first and third year of cultivation.

In addition, the following pathogenic species of fungi were also isolated: *Phoma medicaginis* – in the first and second year of cultivation of alfalfa and grasses, *Verticillium albo-atrum* – in the second and third year of cultivation, *Cylidrocarpon destructans* in the third year of cultivation of the mixture of alfalfa with grasses. Altogether pathogens constitued 7.3 % of all the isolates.

In the soil cultivated for 2-4 years, group of dominants was composed of fungi of the genera: *Chaetomium,* mainly *C. globosum* and *C. olivaceum,* costituting 35.6 % of all the isolates, *Aspergillus* – 9.6 %, *Scopulariopsis* – 8.8 %, *Phoma* – 5.8 %, *Fusarium* – 5.4 %.

In the group sub-dominants the following species of fungi were recognized: *Penicillium,* mainly *P. expansum, P. frequentans, P. janthinellum* and *P. purpurogenum* – 4.5 %, *Acremonium murorum* – 3.7 %, *Cladosporium herbarum* – 2.7 %, *Sporormia ambigua* – 2.4 %, *Trichoderma viride* – 2.5 %, *Cylindrocarpon destructans* – 2.2 %, *Mortierella isabellina* – 1.9 %, *Doratomyces stemonitis* – 1.7 %, *Paecilomyces farinosus* – 1.4 %, *Verticillium albo-atrum* – 1.4 % and *Pseudeurotium zonatum* – 1.1 %.

In this type of soil environment, an annual increase in the number of colonies was accompanied by a decline in the number of species. An increasing tendency was particularly ahown by *Aspergillus oryzae, Chaetomium olivaceum, Phoma eupyrena* and *Trichoderma viride.*

The species of the genus *Penicillium* which formed the most numerous populations were *P. frequentans, P. janthinellum,* and *P. purpurogenum* in the third year of cultivation.

Nine species of fungi from the genus *Fusarium* (represented by a total of 11 species) were isolated in the second year of cultivation, in the third only 2, whereas in the fourth year of cultivation an increase in the number of species to 6 was observed. Only *F. poae,* was noted throughout the whole period, *F. avenacem, F. culmorum, F. solani* and *F. tabacinum* were isolated during 2 growing seasons.

Of species pathogenic to alfalfa and grasses *F. avenaceum*, *F. culmorum*, *F. oxysporum*, *F. equiseti* and *F. solani* were accompanied by *Cylindrocarpon destructans* (noted in 3 growing seasons), *Verticillium albo-atrum* (2 seasons) and occasionally, *Phoma medicaginis* and *Thanatephorus cucumeris* (1 season). The above species isolated from the soil on which a mixture of alfalfa and grasses was cultivated for 2-4 years, constituted 5.5 % of the total fungal communities.

In the environment of soil with the 3-5 year cultivation alfalfa and grasses (field IX) the prolonged monoculture affected the development of the community of soil fungi. The species composition of this community was slightly more stable than tahthtly described above, while the number of species declined in the succesive years. In the community of fungi isolated from this environment, the following species dominated: *Aspergillus*, mainly *A. oryzae* and *A. versicolor* constituting 26.4 % of the isolates, *Penicillium frequentans*, *P. granulatum* and *P. steckii* – 19.33 %, *Paecilomyces farinosus* – 6.9 %and *Mortierella parvispora* and *M. isabellina* – 5.2 %.

In the group of sub-dominants the most frequenty occuring species were: *Scopulariopsis*, mainly *S. brumptii* – 4.7 %, *Trichoderma harzianum* and *T. viride* – 4.0 %, *Cladosporium sphaerospermum* – 3.6 %, *Chaetomium globosum* – 3.4 %, *Monilia geophila* – 3.3 %, *Phoma chrysanthemicola*, *P. eupyrena* and *P. medicaginis* – 2.6 %, *Acremonium murorum* – 2.3 %, *Fusarium* sp. – 2.3 %, *Humicola fuscoatra* var. *fuscoatra* – 2.0 %, *Sclerotinia sclerotiorum* – 1.8 %, and *Rhizopus stolonifer* – 1.0 %.

After 4 years of cultivation of the mixture of alfalfa and grasses, a rapid increase in the number of colonies of *Aspergillus versicolor* and *Penicilium granulatum* was noted in field IX. These changes were accompanied by a decrease in the number of populations of *Fusarium avenaceum*, *F. nivale*, *F. oxysporum* and *Phoma medicaginis*. In the fifth year of the study, no species of the genera *Fusarium* and *Verticillum* were observed, while the number of populations of *Aspergillus oryzae* considerably increased.

Pathogenic fungi constituted 4.6% of the community of fungi isolated from the soil of the investigated field (IX).

## DISCUSSION

The results of the present study on the communities of fungi developing in three soil environments indicate that after an initial chance colonization of the dumping grounds by various species of fungi, the species composition was stabilized and a community of soil fungi specific for the cultivated plants developed.

The processes of stabilization in the communities of soil fungi effected by plants of *Papilionaceae* and *Gramineae* families were observed by many autors (Truszkowska i Legieć, 1973; Czaplińska, 1973; Truszkowska i Kalińska, 1979; Kutrzeba, 1983, 1984; Dorenda, 1986).

In the 3-year study, 40 common species of fungi were isolated from the 3 plantations (differing by age and the period of agricultural land use), this indicating that the cultivated plants had a profound effect on the development od specific fungal communities. Many autors isolated some of these species from soils with alfalfa and orchard grass cultivation (Czaplińska, 1973; Dorenda, 1982, 1986; Kutrzeba, 1983, 1984).

A comparison of the communities of soil fungi isolated from the initial soils of industrial waste dump and from arable soils (in mountainous and lowland conditions) showed that such species as *Absidia glauca, Acremonium murorum, Alternaria alternata, Aspergillus versicolor, Chaetomium globosum, Cladosporium herbarum, Cylindrocarpon destructans, Fusarium avenaceum, F. culmorum, F. oxysporum, F. solani, Gliocladium catenulatum, Paecilomyces farinosus, Penicillium chrysogenum, P. granulatum, Phoma eupyrena, P. medicaginis, Trichoderma viride, T. harzianum, Verticillium albo-atrum and V. lateritium* were fairly common in soils planted with alfalfa and orchard grass.

Though some pathogenic species of fungi such as *Phoma medicaginis* and *Verticillium albo-atrum* (Gordon et al., 1989; Hwang et al., 1989; Newcombe, Roob, 1989; Rodriguez et al., 1990) occurred in all the investigated fields with alfalfa and grasses, they did not cause infections of cultivated plants. Neither other species of fungi, such as *Fusarium avenaceum, Cylindrocarpon destructans* or *Thanatephorus cucumeris* which periodically occur in thise environments and are considered to be pathogenic to alfalfa and grasses (Czplińska, 1973; Lamprecht et al., 1986; Hwang et al., 1989) caused any diseases in plants, growing on the industrial waste dump . The constant good health condition of these plants may be associated with the presence of nonpathogenic forms of fungi particularly of the genus *Fusarium* (Schneider, 1984; Park et al., 1988), higher tolerance to fungal infections, or with the fact that the pathogens did not come in contact with the roots of the plants (Sivan, Chet, 1989). According to Truszkowska and Legieć (1973) the growth of *Verticillium albo-atrum* is inhibited above 20°C, this temperature having been noted during the studies from June to August. Moreover due to its low survival rate *V. albo-atrum* is not able to developed in soil over a longer period (Gordon et al., 1989; Newcombe, Robb , 1989).

The absence of disease symptoms in the cultivated plants in the presence of a pathogen may be associated with the fact that the roots of alfalfa plants secrete substances which make them resistant to infections. These substances may also induce the growth of antagonistic organisms (Gibert et al., 1969; Lewis, Papavizas, 1983). The stimulating action is manifested by the rapid growth of saprophytic fungi in the soil, such as: *Acremonium, Aspergillus, Chaetomium, Cladosporium, Doratomyces, Gliocladium, Scopulariopsis, Penicillium* and *Trichoderma* which prevailed over the pathogens. This finding is in agreement with the opinion of Gilbert et al. (1969) that volatile substances of distillates of alfalfa hay stimulate the growth of saprophytic fungi species, above all of the genera *Trichoderma, Aspergillus*, and some species of *Penicillium*.

In the conditions of the industrial waste dumps a rapid growth of *Rhizobium* was noted (K o w a l i k, 1989 a) while an anatgonistic effect of bacteria (K w o k et al., 1987; P a r k et al., 1988; B i n et al., 1991), including *Rhizobium*, on pathogenic fungi was also reported (F u r g a ł - W ę g r z y c k a, 1987).

Ł a c i c o w a (1989) and other autors (L e w i s, P a p a v i z a s, 1983; B e a g l e - R i s t a i n o, P a p a v i z a s, 1985; H o w e l l, 1987; B i n et al., 1991) suggest an important role of antagonistic fungi of the genera *Gliocladium* and *Trichoderma*. In the investigated area, these species, particularly *T. viride*, *T. harzianum*, *T. piluliferum*, *G. catenulatum* and *G. virens*, night have an inhabiting or reducing effect on the growth of *Verticillium albo-atrum*, *Phoma medicaginis* and *Thanatephorus cucumeris*. K w a ś n a (1987) suggests that *Mortierella isabellina*, *M. vinacea*, *Penicillium janthinellum*, *P. jensenii* and *Trichoderma viride* (often isolated from soils with alfalfa cultivation) have an antagonistic effect on *Fusarium oxysporum* and *Thanatephorus cucumeris*. C z a p l i ń s k a (1973) observes that some fungi of the genus *Fusarium*, including *F. oxysporum* isolated from dumping ground soils, inhibit the growth of *Verticillium albo-atrum*. The above autor has also reported an antagonistic effect of *Penicillium janthinellum* on *Phoma medicaginis*. According to this author the strongest resistance of the environment to *V. albo-atrum* and *P. medicaginis* (= *Ascochyta imperfecta*) is noted in the first year of cultivation. The comparison of the results of mycological analysis of soils differing in age the period of agricultural use, showed that in the case of older plantations (cultivated for 4-5 years), the number of fungi species, including pathogens, decreased. These results, however, were not confirmed by studies conducted by C z a p l i ń s k a (1973). This autor determined the highest number of pathogens of the genera *Fusarium*, *Phoma*, *Verticillium* in the soils of the oldest plantations.

The results obtained indicate that the establishment of such plantations on industrial waste grounds is justified and that the good phytosanitary condition of plantations, high yields, and very positive effects on the soil show the feasibility of (K o w a l i k, 1989 a, b) maintaining them on industrial wastes for a longer period than on arable soils.

## CONCLUSIONS

1. The mixture of alfalfa with grasses introducetd as a first culture on the raw substrate of industrial wastes favoured the development of communities of soil fungi. Since the cultivated plants were resistant to fungal infections and had a positive effect on the soil, they should be cultivated for a longer period than on well developed soils.

2. In the course of the soil development processes, communities of soil fungi isolated from the initial soil of the waste dump planted with a mixture of alfalfa and grasses, become more similar to the fungal communities colonizing well developed soils.

3. Communities of soil fungi in the initial soil of a dumping ground become more stable with the age of the plantation.

4. Since the very beginning of the development, the increasing risk of plant pathogens should be taken into cousiderations in the environement of initial soil.

## Summary

Researches were conducted during the years 1988-1990 on the "Machów" industrial waste dump. It was determined, that a mixture of alfalfa with grasses, introduced as first culture on the raw substrate of industrial wastes, was helpful for the development of soil fungi communities. The constantly good health condition of plants and their beneficial influence upon the substrate suggest the cultivation of the mixture for a longer period thatn on well developed soils.

In the course of soil development processes communities of soil fungi, isolated from the initial soil of the waste dump planted with the mixture of alfalfa and grasses, became more similar to fungal communities occupying well developed arable soils.

Along with the ageing of the plantation growing on the initial soil the communities of soil fungi became stabilized.

In the environment of initial soil, since the very beginning of its development, the increasing risk of plant pathogens outbreak should be taken into consideration.

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