韓 國 菌 學 會 誌 第 15 卷 第 3 號
Kor. J. Mycol. Vol. 15, No. 3, 187-195, 1987

Fungal flora of paddy field in Korea
IV. Filamentous fungi isolated by heat treatment

Min, Kyung Hee, Tadayoshi Ito* and Tatsuo Yokoyama*
*Department of Biology, College of Sciences, Sookmyung Women's University, Seoul 140, Korea and *Institute for Fermentation, Osaka, Osaka 532, Japan*

韓國 논土壤中의 菌類에 關한 硏究
IV. 熱處理로 分離한 絲狀菌類

閔庚喜·伊藤忠義*·橫山竜夫*
淑明女子大学校 理科大学 生物学科, *日本国 大阪市 財団法人 醱酵研究所

ABSTRACT: Two kinds of heat treatment method for the selective isolation of soil fungi to eliminate the commonest fungi and also to examine the vertical and seasonal distributions of the fungal population were applied to soil samples from two plots around Seoul area. The incubation method at 42℃ and heat treatment at 70℃ were used in this experiment. In the incubation method, the almost all the fungi isolated from two plots were mesophile, while the thermotolerant fungi was *Aspergillus fumigatus* and thermophilic fungi were *Sporotrichum thermophile* and *Malbranchea pulchella* var. *sulfrea*. The most dominant species isolated by this method was *A. fumigatus*. Nine genera and fourteen species were isolated from the two plots, and *S. thermophile, Talaromyces ucrainicus, Malbranchea pulchella* var. *sulfrea* were new to Korea. From the selection method by heat treatment at 70℃, ten genera and twenty species were isolated. Among these, the most fungi were also mesophile and thermotolerant fungus was *A. fumigatus*. The most dominant species isolated by this method was *T. stipitatus, Talaromyces helicus* var. *major, Emericella nidulans* var. *nidulans, Chaetomium subspirale* and *Neosartorya fisheri* var. *fisheri* were new to Korea. From the two isolation methods, it was found that the total number of soil fungi and frequency of species appeared including dominant ones were the highest at the soils of upper layer while the lowest at the soils of lower layer in its vertical distribution.

KEYWORDS: Heat treatment method, Mesophile, Thermotolerant fungi, Thermophilic fungi

Temperature is one of the important factors affecting the growth of all microorganisms. Cooney and Emerson(1964) described thermopilic and thermotolerant fungi consisting of twelve species and their application for industrial purposes. Eggins *et al.*(1969) and Fergus(1969a) reported the isolation of thermophilic cellulolytic fungi from the pasture and soil. Fergus(1969b) also described the amylase activity of several thermophiles and isolated a new species, *Chaetomium rectophilium*, from mushroom compost. Tansey(1971) isolated thermophiles from self-heated industrial wood chip piles and Ward *et al.*(1972) from forest soil of central South Carolina. Minoura *et al.*(1973a, 1973b) reported the thermophiles found in Japan and several interesting fungi together with many known thermophiles which were isolated from various substrates. Awao *et al.*(1973a, 1973b, 1974) identified five species of thermophilic fungi using the modified method of Warcup and Baker's heat treatment.

Recently, Ito *et al.*(1981) reported that the thermophilic and thermotolerant fungi were isolated respectively by the heat incubation method at 45℃ from soils of four paddy fields in Japan and they also investigated seasonal fluctuations in dominant species and distribution of thermophilic and thermotolerant fungi, but they also isolated the lower population of mesophilic fungi from soil samples.

In our previous papers(Min *et al.*, 1981, 1982b),

we recorded the mesophilic species isolated from the paddy field soils in Korea using two kinds of sampling methods, and showed their vertical distributions and seasonal fluctuations.

In our present work, two kinds of heat treatment methods for the selection of soil fungi were applied in order to eliminate the ubiquitous fungi from soil samples. The incubation method at 42℃ and heat treatment at 70℃ were adopted to isolate the thermophilic and thermotolerant fungi. However the major population of paddy field soils was mesophile and only a few were of thermophilic or thermotolerant. We describe hereafter the fungal population and distribution in paddy soils in Korea using the two kinds of selection methods, excluding the commonest mesophiles.

Materials and Methods

Sampling sites

Two plots each in the rice paddy fields at Yugkog-dong, Buchun in Kyunggido and Shinwon-dong in Seoul were selected. Yugkog-dong is situated at the southwest of Seoul and Shinwon-dong in the southeast of Seoul. These two sites are one of the typical rice fields in Seoul area where the rice is the most important crop and has been cultivated under typical ecological conditions.

Sampling method

Soil samples were collected once every three months from the sites from April 1980 to January 1981. Soils were taken vertically from each site of paddy fields with sterilized stainless-steel soil sampler 70 cm long and 3 cm in diameter equipped with narrow side window in order to take soil samples. Soil samples were divided into three parts representing depths of 0-10 cm(upper layer), 10-20 cm(middle layer) and 20-30 cm(lower layer), respectively, from the soil surface.

Isolation and identification

For the isolation of the microfungi excluding the most mesophilic fungi, incubation method at 42℃ and heat-treatment methods at 70℃ for 15 minutes were applied in this experiment.

The soil sample was suspended in 5 m*l* of sterilized water, and 0.2 m*l* of the suspension was spread onto MYA plate. For the incubation method at 42℃, these plates were incubated at 42℃ for two days and the isolates were transferred to malt agar(MA) slants and incubated at 37℃ for growth(Ito *et al.*, 1981).

For the treatment, one part of the suspension was treated at 70℃ for 15 min and 0.2 m*l* of the suspension was spread onto the MYA plate, and incubated at 24℃ for the isolation. The composition of the culture medium for the isolation of fungi was malt extract-yeast extract agar(MYA) containing tetracycline(50 mg/m*l*) to prevent bacterial contamination. The MYA medium was composed of malt extract 0.3%, yeast extract 0.3g, peptone 0.5g and distilled water up to 100 m*l*. The fungi appeared on the plates were transferred on the MYA slant and incubated at 24℃. Identification of the fungi was carried out by examining the cultures on Czapek agar, malt extract agar and potato dextrose agar, respectively.

Dilution method

Three soil suspensions prepared were different in their soil amounts; one gram of soil from the upper layer, two grams from the middle layer and three grams from the lower layer, respectively. Each soil sample was added to 5 m*l* of sterilized water in test tube and shaked thoroughly. The aliquots were referred to as soil suspension. A part of soil suspension, 0.2 m*l*, was spreaded on MYA plate and incubated at 42℃ for two days followed by the incubation at 37℃ to isolate the fungi. Other part of each soil suspension, 0.7 m*l* was treated at 70℃ for 15 minutes. From this aliquot, each duplicate of 0.2 m*l* soil suspension was spread on MYA plate and incubated at 24℃ for the isolation of other groups of fungi.

Results

Fungal isolation by the incubation method

This experiment included the isolation of fungal species and distribution of microfungi with the exception of the common soil fungi using the incubation method at 42℃.

Isolation of fungal species from two sites: From the soil samples collected at the paddy fields at Yugkogdong during various stages of cultivation and crop growth, eight species classified in 6 genera were isolated(Table 1), comprising 2 species of Zygomycotina, 2 of Ascomycotina, and 6 of Deuteromycotina. Twelve species classified in 7 genera were obtained from soil samples of Shinwondong

Table 1. List of microfungi from paddy field soils by the incubation method at Yugkog-dong(Fungal propagules/gram of soil)

Season / layer	April			July			Oct.			Jan.			Sub-Total			Total
Fungal species	A	B	C	A	B	C	A	B	C	A	B	C	A	B	C	
Zygomycotina																
Rhizopus arrhizus		6												6		6
Ascomycotina																
Talaromyces stipitatus		6						8						14		14
Eupenicillium sp.		6												6		6
Deuteromycotina																
Aspergillus. flavus				38	32	23							38	32	23	93
A. fumigatus	25	9	4	325	100	16	350	175	156	38	25	4	738	309	164	124
A. tereus				13									13			13
Penicillium piceum	13		4										13		4	17
Sporotrichum thermophile							13	32					13	32		45
Unidentified fungi	50	17	26	13	13	17					6	17	63	32	60	159
Total	88	44	34	38	145	57	363	215	156	38	31	21	878	435	215	1564

Table 2. List of microfungi from paddy field soil by the incubation method at Shinwon-dong(Fungal propagules/gram of soil)

Season / layer	April			July			Oct.			Jan.			Sub-Total			Total
Fungal species	A	B	C	A	B	C	A	B	C	A	B	C	A	B	C	
Ascomycotina																
Chaetomium sp.							13						13			13
Eupenicillium sp.							25						25			25
T. ucraincius	25												25			25
T. stipitatus	25						38						63	38		101
Deuteromycotina																
A. flavus				23	19	6							23	19	6	48
A. fumigatus	156	125	26	350	275	129	150	48	19	112	38	8	762	486	18	1430
A. terreus													50	19		69
P. piceum	75	25	4										75	25	4	104
P. cyclopium				63									63			63
P. funiculosum	25												25			50
Malbranchea pulchella var. *sulfrea*										25	6		25	6		31
Acromonium spp.	12												12			24
Unidentified fungi	13	25	4	25	25	38	63	25	38		6		101	81	80	262
Total	325	213	34	511	325	173	289	73	57	137	63	8	1262	674	272	2208

as shown in Table 2, consisting of 4 species of Ascomycotina, and 8 of Deuteromycotina.

As shown in Table 1, fungal species identified were *Rhizopus arrhizus, Talaromyces stipitatus, Eupenicillium* sp., *Aspergillus flavus, A. fumigatus, A. terreus, Penicillium piceum,* and *Sporotrichum thermophile* from soil samples at Yugkog-dong, and *Chaetomium* sp., *Eupenicillium* sp., *T. ucrainicus, T. stipitatus, A. flavus, A. fumigatus, A. terreus, P. piceum, P. cyclopium, P. funiculosum, Malbrachea pulchella* var. *sulfrea, Acremonium* spp. at Shinwon-dong site were identified(Table 2).

The common species in the soil samples from two sampling sites were *T. stipitatus, A. flavus, A. fumigatus, A. terreus,* and *P. piceum.* Among the fungi isolated, *Sporotrichum thermophile, T. ucrainicus,* and *Malbrnchea pulchella* var. *sulfrea* are new to Korea.

From these results, it was concluded that *A. fumigatus* was dominant species at two sites, Yugkog-dong and shinwon-dong sampling sites.

In the incubation method at 42℃, the most common fungi appeared were the mesophilic microfungi, but thermophilic fungi such as *Sporotrichum thermophile* and *Malbranchea pulchella* var. *sulfrea,* and thermotolerant fungi, *Aspergillus fumigatus,* were isolated.

Distribution of microfungi: 1. *Total fungal population;* Incubation of the plate culture at 42℃ greatly reduced the number of colonies as compared with the fungal population of original soil suspension(Min *et al.*, 1981). Tables 1 and 2 showed the number of fungal propagules per gram of soil including three soil samples with the increasing depth in two experimental sites.

Several fungal species occurred in this experiment was not common among soil fungi, and they were selected by heat incubation at 42℃.

In general, the fungal polulation per gram of soil was decreased with the increased depth from the soil surface at any seasons. This fact may suggest that these soil fungi are aerobic and also need more nutrient at the surface of soils. The similar tendency was also found in the distribution of total fungal population(Min *et al.*, 1981).

2. *Different fungal species;* As shown in Table 2, the kinds of fungal species at the upper layer of soil samples from Shinwon-dong site were more variable than those at the middle layer. Table 2 showed

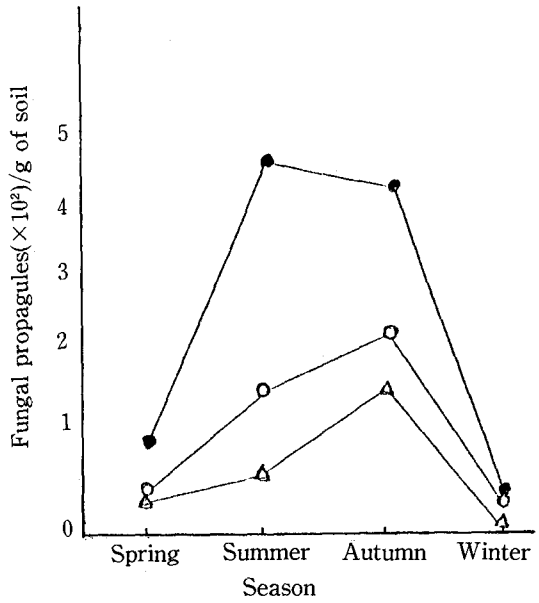

Fig.1. Seasonal fluctuation of fungal propagules at Yugkog-dong by the incubation method. Upper layer(●-●), middle layer(○-○) and lower layer(△-△).

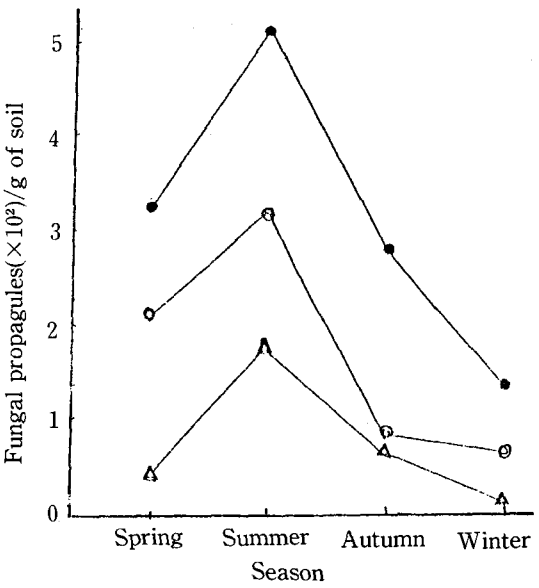

Fig.2. Seasonal fluctuation of fungal propagules by the incubation method at Shinwon-dong. Upper layer(●-●), middle layer(○-○), and lower layer (△-△).

that 6 species at the upper layer, 3 at the middle layer, and 2 at the lower layer were appeared in the spring.

As the species variation in Table 1, the similar tendency was found in the paddy soil of Yugkog-dong site(Table 2).

3. *Vertical distribution of dominant species* : The number of fungal propagules per gram of soil by the selection method of incubation at 40℃, particularly of dominant species *Aspergillus fumigatus*, was the highest at the upper layer at any seasons. Thereafter, they decreased at the middle and lower layers. These results were in good agreement with the tendency of fungal distribution at two sites. Dominant species isolated in the paddy fields was appeared during all the cultivation stages of rice plants(Figs.1 and 2).

Fungal selection with heat treatment method

Fungal species isolated at two sites: From the heat treatment at 70℃, fourteen species in ten genera were isolated from the soil samples of Yugkog-dong as shown in Table 3. They were composed of 2 species of Zygomycotina, 6 species of Ascomycotina, and 6 species of Deuteromycotina. As shown in Table 4, 12 species in 6 genera were isolated from the soil samples of Shinwon-dong, southeast of Seoul, comprising of 4 species of Ascomycotina and 8 species of Deuteromycotina.

Microfungi from paddy field soils of two sites were identified as *Rhizopus* sp., *Zygorhynchus moelleri, Talaromyces stipitatus, T. helicus* var. *major, T. flavus* var. *flavus. Emericella nidulans*

Table 3. List of microfungi from paddy field soil by heat treatment method at Yugkog-dong(Fungal propagules/gram of soil)

Season / layer	April			July			Oct.			Jan.			Sub-Total			Total
Fungal species	A	B	C	A	B	C	A	B	C	A	B	C	A	B	C	
Zygomycotina																
Rhizopus sp.								6						6		6
Zygorhynchus moelleri	25												25			25
Ascomycotina																
T. stipitatus	425	285	201	602	375	247	319	306	87				1446	966	535	2947
T. helicus var. *major*		6												6		6
Eupenicillium spp.	25												25			25
E. javanicum	13												13			13
Emericella nidulans var. *nidulans*		6												6		
Chaetomicum subspirale									4						4	4
Deuteromycotina																
A. fumigatus					13	9								13	9	22
A. versicolor										25			25			25
Cladosporium sphaerospermum	13												13			13
Cladosporium sp.				12									12			12
P.frequentans							13		4				13		4	17
P. verrucosum								6	3					6	3	9
Unidenified spp.	75	6	16	125	6	17	38	12	35							
Total													288	18	87	393
Total	576	297	217	739	394	273	370	330	133	25			1860	1021	642	3517

Table 4. List of microfungi from paddy field soil by heat treatment method at Shinwon-dong(Fungal propagules/g of soil)

Season / layer / Fungal species	April			July			Oct.			Jan.			Sub-Total			Total
	A	B	C	A	B	C	A	B	C	A	B	C	A	B	C	
Ascomycotina																
Neosartorya fischeri var. *fischeri*	25	25											25	25		50
T. flavus var. *flavus*		13												13		13
T. stipitatus	550	388	204	613	456	229	438	356	173	75	25	17	1676	1225	623	3524
Eupencillium sp.	13												13			13
Deuteromycotina																
A. fumigatus				50									50			50
A. repens			4												4	4
A. vericolor							13						13			13
C. cladosporioides								6						6		6
P. cyclopium			4			9					13			13	13	26
P. freguentans							25						25			25
P. funiculosum	25	13	4										25	13	4	42
P. verruclosum							50	63	9				50	63	9	122
Unidentified spp	13	26		25	36	13	13		4				51	62	17	
Total	626	465	216	688	492	251	539	425	186	75	38	17	1928	1420	670	4018

var. *nidulans, Eupenicillium* sp., *E. javanicum, Chaetomium subspirale, A. fumigatus, A. repens, A. versicolor, Cladosporium cladosporioides, C. sphaerospermum, C.* sp., *P. cyclopium, P. frequentans, P. funiculosum,* and *P. verruculosum.*

A comparison between the species isolated from Yugkog-dong and Shinwon-dong plots showed a good similarity(Tables 3 and 4). The common species from the soil samples in two sites were *T. stipitatus, A. fumigatus, A. versicolor, P. funiculosum* and *P. verrucosum.* Among these fungi, *T. helicus* var. *major, Emericella nidulans* var. *nidulans, Chaetomium subspirale, Neosartorya fisheri* var. *fisheri* are new to Korea. In the heat treatment method at 70℃, it was found that the most microfungi isolated were mesophilic fungi, but *T. stipitatus* was thermotolerant fungi.

B. Vertical distribution of soil fungal population:

1. *Total fungal population;* Heat treatment of soil suspension at 70℃ reduced the fungal population at two sites as shown in Tables 3 and 4, as compared with direct dilution plate method(Min *et al.*, 1981).

The results in Tables 3 and 4 showed that the fungal population isolated by the heat treatment method at 70℃ were very low, but it was dependent on the soil depth and independent on the location of the paddy fields in which the fungi were grown.

From these results, it was concluded that the distribution of the fungi at each soil depth was found to be the same tendency as those obtained by the incubation method at 42℃. The total number of microfungi from Shinwon-dong samples were higher than those from Yugkog-dong samples from the results of the incubation method at 42℃.

2. *Frequency occurring fungal species* ; The experiment included the vertical distribution of different kinds of fungal species. Table 4 showed that four kinds of fungal species was isolated from the soil of upper layer, 3 species from the middle layer, and 2 species from the lower layer. Even if some variation has appeared at other seasons and sites, the similar tendency was also obtained from this experiment.

3. *Vertical distribution of dominant species* ; It

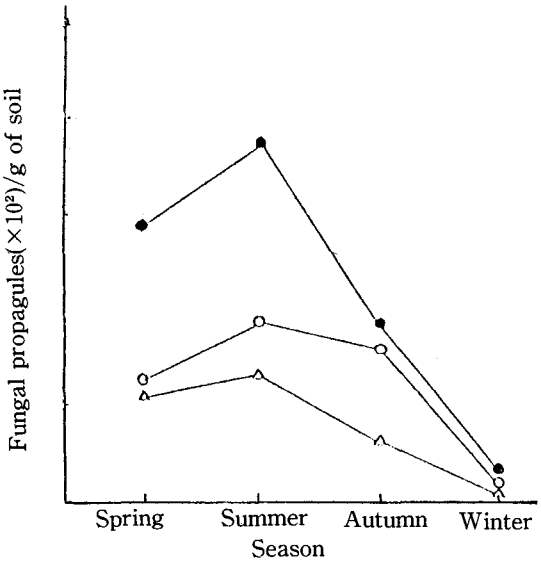

Fig.3. Seasonal fluctuation of fungal propagules by heat-treatment method at Yugkog-dong. Upper layer(● - ●), middle layer(○ - ○), and lower layer (△ - △).

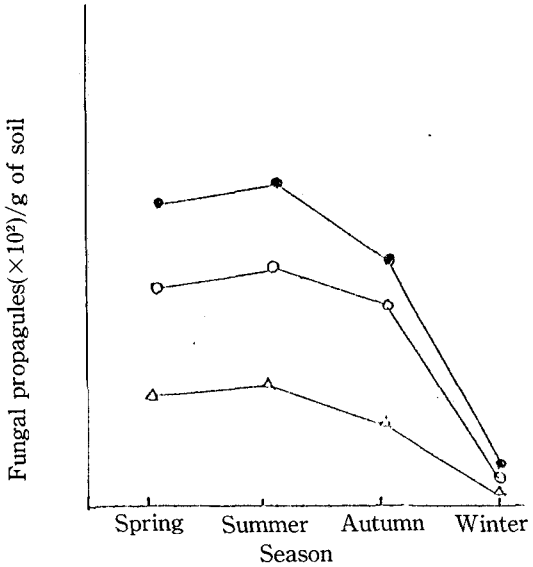

Fig.4. Seasonal fluctuation of fungal propagules by heat treatment method at Shinwon-dong. Upper layer(● - ●), middle layer(○ - ○), and lower layer (△ - △).

should be pointed out from the present work that *T. stipitatus*, a mesophile, was dominant species with heat treatment method at 70℃ among the soil samples in two sites. As shown in Tables 3 and 4, vertical distribution of dorminant species was related with soil depth. Fungal population from soil of the upper layer was 425 colonies per gram of soil, 285 from the middle layer, and 201 from the lower layer at spring season. This tendency was also obtained from the results which was investigated at four seasons and at the two sites.

Although the dominant fungi remained more or less constant throughout the year and in all fields studied, some of the fungi were found specifically at certain stages of growth of rice plants. Thus some were found to prefer water-logged condition and dry condition(Figs.1 and 2).

Discussion

The results of the surveys summarized here and in the two previous papers(Min *et al.*, 1981; Min *et al.*, 1982a) provides some information on the mycoflora of rice paddy field soils of Seoul area.

In our present investigation on the filamentous fungi of two plots, the kinds of microfungi were recognized to depend on the soil depths in which they live, as suggested by Apinis(1963) and Evans(1971). Moreover, Ito *et al.*(1981) confirmed that thermophilic and thermotolerant fungi could be constantly isolated, by the incubation method and heat treatment from the given sites throughout the investigation of three years.

We attempted to investigate the microfungal flora and distribution concerning with temperature. It was also carried out to eliminate the common fungi using two kinds of selection methods. We found that the most common fungi was mesophilic, while a few were thermophilic and thermotolerant fungi. At below 20 cm depth of soil surface, the kinds and total numbers of fungal species isolated at each site decreased remarkably. Similar results were obtained by ethanol treatment, heat treatment, and the dilution plate method(Ito *et al.*, 1981). This results also agreed well with the findings of Tansey and Jack(1976), who studied the thermophilic fungi of the grassland in Indiana in the United States at different depths of soil.

The dominant species of soil samples by incubation methods at 42℃ was *A. fumigatus*, a thermotolerant fungus, and dominant species with heat

treatment method was *T. stipitatus*, thermotolerant fungi. It was found that many species isolated from soil samples by two methods were mesophilic fungi in different two sites. Ito *et al.*(1981) described that the predominant thermophilic and thermotolerant fungi at Habikino and Ibaraki stations were *A. fumigatus*. This results were in good agreement with our experiment by incubation method at 42℃. However, it seems to be noticeable that dominant species by heat treatment method was *T. stipitatus*, a thermotolerant fungus, at different two sites.

摘 要

두가지 열처리법에 의한 균 분리방법을 사용하여, 보통균을 제외한 토양균의 분리와 그의 논토양중의 수직 및 계절별 분포를 파악하기 위하여 역곡동과 신원동의 논토양을 시료로 사용하였다. 42℃에서 2일간 배양후 37℃로 배양하는 방법과 70℃에서 15분간 열처리하는 방법을 사용하였다.

42℃ 배양법에서는 두 장소에서 분리된 토양균은 거의가 mesophile이었으며 thermotolerant fungi는 *Aspergillus fumigatus*, thermophile로는 *Sporotrichum thermophile*과 *Malbranchea pulchella* var. *sulfrea* 이었고, 이들 중 우점종은 *Aspergillus fumigatus*이었다. 두 장소에서 9속, 14종이 분리동정되었으며 *Sporotrichum thermophile, Talaromyces ucrainicus, Malbranchea pulchella* var. *sulfrea* 는 한국 미기록 종이다.

72℃ 열처리방법으로 분리한 경우 두 장소에서 10속, 20종이 분리동정되었으며, 분리된 균은 거의가 mesophile이었으며 thermotolerant fungi는 *Aspergillus fumigatus*이었고, 우점종은 *Talaromyces stipitatus* 이었다. 또한 *Talaromyces helicus* var. *major, Emericella nidulans* var. *nidulans, Chaetomium subspirale, Neosartorya fishiri* var. *fischeri*는 한국 미기록 종이다. 두가지 분리방법으로 분리된 토양균의 계절적 분포는 여름과 봄이 가을과 겨울보다 높았으며, 전체 균의 수와 우점종 균수, 그리고 나타나는 종의 빈도는 그 수직분포에 있어서 상층에서 가장 높았으나 하층으로 내려갈수록 감소하였다.

References

Apinis, A.E. (1963): Occurrence of thermophilous microfungi in certain alluvial soils near Nottingham. *Nova Hedwigia* V, **1/2:**57-78.

Awao, T. and Mitsugi, K.(1973a):Notes on thermophilic fungi in Japan(1). *Trans. mycol. Soc.* Japan **14:** 145-160.

Awao, T. and Otsuka, S.(1973b):Notes on thermophilic fungi in Japan(2). *Trans. mycol. Soc.* Japan **14:** 221-236.

Awao, T. and Otsuka, S.(1974):Notes on thermophilic fungi in Japan(3). *Trans. mycol. Soc.* Japan **15:** 7-22.

Cooney, P. G. and Emerson, R. (1964):Thermophilic fungi. An account of their biology, activity and classification. W.H. Freeman Co., San Francisco. 163p.

Eggins, H. O. W. and Malik, K. A.(1969):The occurrence of thermophilic cellulolytic fungi in a pasture land soil. *Antonie van Leeuwenhoek* **35:** 178-184.

Evans, C. L.(1971): Thermophilous fungi of coal spoil tips . I. Taxonomy. *Trans. Br. mycol. Soc.* **57:** 241-254.

Fergus, C.L. (1969a): The cellulolytic activity of thermophilic fungi and Actinomycetes. *Mycologia* **61:** 120-129.

Fergus, C.L.(1969b):The production of amylase by some thermophilic fungi. *Mycologia* **61:** 1171-1175.

Fergus, C. L. and Amelung, R. M. (1971) : The heat resistance of some thermophilic fungi on mushroom compost. *Mycologia* **63:** 675-679.

Ito, T. Ueda, M. and Yokoyama, T.(1981):Thermophilic and thermotolerant fungi in paddy field soils. IFO Res. Communications 20-32.

Min, K. H. Ito, T. and Yokoyama, T.(1981):Fungus flora of paddy fields in Korea(1). Fungal distribution of paddy fields. *Kor. J. Microbiol.* **19:** 153-162.

Min, K. H. Chun, K. S. Ito, T. and Yokoyama, T. 1982a. Fungus flora of paddy fields in Korea(II). Fungal flora of paddy fields. *Kor. J. Microbiol* **20:** 41-51.

Min, K. H. Ito, T. and Yokoyama, T.(1982) : Fungus

flora of paddy fields in Korea(III). Ascomycetes. *Kor. J. Microbiol.* **20:** 80-88.

Minoura, K. Yokoe, M. Kizima, K. and Nehira, T. (1973a) : Thermophilic filamentous fungi in Japan(1). *Trans. mycol. Soc.* Japan **14:** 352-361.

Minoura, K.Ochi, K. and Nehira, T. (1973b) : Thermophilic filamentous fungi in Japan(2). *Trans. mycol. Soc. Japan* **14:** 362-366.

Tansey, M. R. 1971: Isolation of thermophilis fungi from self-heated industrial wood chip piles. *Mycologia* **63:** 537-547.

Tansey, M. R. and Jack, M. A. (1976): Thermophilic fungi in sun-heated soils. *Mycologia* **68:** 1061-1075.

Ward, J.E. Jr. and Cowley, G. T. (1972): Thermophilic fungi of some central South Carolina forest soils. *Mycologia* **64:** 200-204.

Accepted for Publication 7 December