

New Report on Sordarialean Fungi in Korea

Seonju Lee, Jin-Chang Ryu and Seung-Joo Go*

Molecular Genetics Division, National Institute of Agricultural Science and
Technology, Rural Development Administration, Suwon 441-707, Korea

국내에 분포하는 Sordariales목 균에 관한 보고

이선주 · 류진창 · 고승주*

농업과학기술원 분자유전과

ABSTRACT: Taxonomical studies of soil-borne fungi were performed. Soil samples were collected from greenhouses located in Jinju-City, Korea. Soil dilution plating method was adopted for the pure isolation. Seven ascomycetous fungi were isolated from the soil samples, and subsequently identified. Among them, five isolates, *Chaetomium brasiliense*, *C. indicum*, *Corynascus sepedonium*, *Petriella sordida*, and *Thielavia fragilis* have never been reported in Korea. Both teleomorphic and anamorphic states were observed in the two species, *Corynascus sepedonium* and *Petriella sordida*, and only teleomorphic state was observed in *Thielavia fragilis*, *Chaetomium indicum*, and *Chaetomium brasiliense*. Illustrated descriptions are presented for the isolates examined in the present study.

KEYWORDS: Ascomycota, Sordariales, *Chaetomium brasiliense*, *Chaetomium indicum*, *Corynascus sepedonium*, *Petriella sordida*, *Thielavia fragilis*

Most sordarialean fungi known as coprophilous ascomycetes are saprobes in cellulosic substrates including dung, wood, and soil. Their taxonomic status and ordinal concept are not in complete consent among fungal taxonomists. Based on the centrum structure, members of current sordarialean fungi were placed in the separate orders, Microascales, Coronophorales, and Sphaeriales under the class Euascomycetes by Luttrell (1951). Thereafter sordarialean fungi did not locate their exclusive position as a monophyletic group until Barr (1976, 1990) established the order Sordariales under the subclass Parenchymatomyetidae based on the characteristics of centrum and ascomatal peridium. Phylogenetic studies on perithecial ascomycetes using DNA sequences revealed a number of sordarialean

species form a monophyletic groups within the clade containing Ophiostomatales, Diaporthales, and Xylariales (Alexopoulos *et al.*, 1996). Even though many taxonomic studies are left over, the order Sordariales are broadly accepted. Hawksworth *et al.* (1995) assigned eight families, 121 genera, and 676 species to the Sordariales. Alexopoulos *et al.* (1996) adopted the Sordariales to encompass six families.

Sordarialean fungi are characterized by cleistothecial ascomata or membranous-walled perithecia with short ostioles. Asci produced in basal fascicles often have apical ring which does not blue in iodine. Ascus walls may be evanescent. Ascospores are hyaline to pigmented, at least in part, with germ pores or slits, and may bear gelatinous sheaths and appendages. The spores in some species are shot out with some force and the gelatinous

*Corresponding author

appendages aid in the adherence of the spores to neighboring grasses and herbs. Conidia, when present, are small phialospores borne on short phialides, or catenulate blastospores and aleuriospores in a few species.

Most of sordariales fungi did not come into the spotlight of scientists, but several species including *Neurospora* and *Sordaria* have proved of great value as experimental organisms (Alexopoulos *et al.*, 1996).

There have been many studies on the community ecology of microfungi in soils (Lee *et al.*, 1987a, 1987b; Lee *et al.*, 1988; Lee *et al.*, 1995; Yoo *et al.*, 1983). New records of many imperfecti and several ascomycetes have been filed in Korea. Few sordariales fungi, however, have been reported. This aspect seems to be ascribed to the lack of interest in soil-borne ascomycetes. Even though Cho and Chun (1962) reported about 20 common soil-borne fungi, Min *et al.* (1980a, 1980b, 1981, 1982a, 1982b) first found ascomycetes, teleomorphic states of *Aspergillus* and *Penicillium* from soil in Korea. Some sordariales fungi, *Chaetomium globosum*, *C. funicola*, and *C. subspirale*, were also reported (Kim, 1979; Kim and Lee, 1980; and Min *et al.*, 1987).

In establishing molecular monitoring system of soil-borne fungi the distribution pattern of mycoflora was investigated by soil dilution plating method. Majority of fungi isolated were *Penicillium*, *Aspergillus*, some aleuriosporic dematiaceous imperfecti, and several ascomycetes. Among them, five sordariales were first reported in Korea and illustrated.

Materials and Methods

Soil samples were collected from muskmelon-cultivating greenhouse in Jinju-City in Korea and dried in the laboratory at room temperature. Dried samples were kept at -70°C freezer for storage and subsequent

study to prevent the fluctuation of microbial community. Initial dilution was prepared by mixing 10 g (dry weight) soil with 90 ml sterilized water containing 1% agar. After vortexing the soil dilution samples for 5 min, serial dilutions (10^3 , 10^4 , and 10^5) were made. Three plates were prepared from the initial and diluted suspensions. The dilutes of 1 ml were spread onto each of rose bengal agar (RBA) consisting of 16 g of Bacto[®] Rose Bengal Agar Base/500 ml and 100 unit of streptomycin/ml. All plates were incubated at 25°C in darkness until discrete colonies appeared. The RBA plates were examined under a dissecting microscope until no additional species were encountered. Relative abundance of each identified species was not quantified. General observations and measurements of asci and internal structures were made on fresh material mounted in water. Species of the isolates were identified according to the keys of Arx (1975) and Arx *et al.* (1986, 1988). Light micrographs were taken with a Nikon NFX-35 on Kodak Gold III film, and scanning micrographs were taken on Zeiss SEM.

Observations

Five sordariales fungi described in the present study were deposited in the KACC (Korean Agricultural Culture Collection) of NIAST and summarized in Table 1.

Corynascus sepedonium (Emmons) v. Arx (Fig. 1)

Ascomata nonostiolate, globose cleistothecia, glabrous, dark brown. Ascomatal wall brown, textura epidermoidea, $5.3\text{--}14.4 \times 7.5\text{--}11.2\text{ }\mu\text{m}$. Asci in a fascicle, obovate or nearly spherical, with a thin wall that is evanescent at maturity, 8-spored, $28.8 \times 9.6\text{--}12.8\text{ }\mu\text{m}$. Ascospores 1-celled, olivaceous to dark brown, smooth-walled, broadly fusiform, with a distinct germ pore at each end, $12\text{--}13.6 \times 6.8\text{--}8.0\text{ }\mu\text{m}$. Anamor-

Table 1. The list of ascomycete fungi presented in this study

Taxonomic status ^a	Result of Observation		Isolate No.
	Sexual stage	Asexual stage	
Ascomycota			
Sordariales			
Ceratostomataceae	<i>Corynascus sepedonium</i>	<i>Chrysosporium</i> sp.	KACC40305
Microascaceae	<i>Petriella sordida</i>	<i>Graphium</i> sp.	KACC40306
Chaetomiaceae	<i>Chaetomium indicum</i>	not observed	KACC40307
	<i>Chaetomium brasiliense</i>	not observed	KACC40309
	<i>Thielavia fragilis</i>	not observed	KACC40310

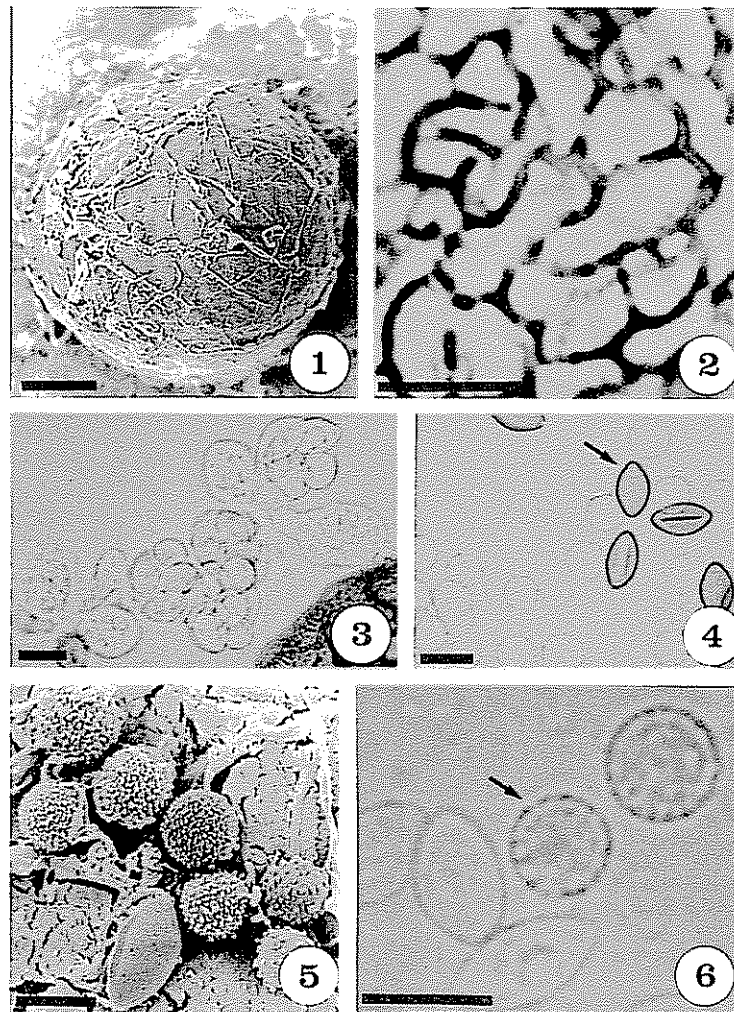
^a Alexopoulos *et al.* (1996).

Fig. 1. Morphological features of *Corynascus sepedonium*. 1, Scanning electron microscopic feature of an ascoma; 2, Ascomatal wall of textura epidermoidea; 3, Asci with ascospores; 4, Mature ascospores (arrow) with germ pores at each end; 5, Scanning electron microscopic view of an ascospore and conidia; 6, Mature conidia (arrow) and young ascospores. (bar in Fig. 1 = 50 μ m; Figs. 2~6=10 μ m)

ph observed, *Chrysosporium* sp. Conidia blastosporous, formed singly on short denticles, thick-walled, nearly hyalin and spherical when young, finely echinulate and light brown when mature, 7.2~9.6 μm in diameter.

Collected from soil of muskmelon-growing greenhouse, Jinju-City, Kyungnam province, May, 1997. Isolate deposited in KACC (#40305).

자낭과는 구형의 폐쇄 자낭각으로 진한 갈색을 띠며, 외부에 부속조직을 가지지 않고 매끈하다. 자

낭과 외벽은 갈색의 표피상 균사조직으로 구성되어 있으며, 크기는 5.3~14.4 \times 7.5~11.2 μm 이다. 자낭은 총생하고, 역난형 또는 원형이며, 8개의 포자를 가진다. 세포벽 크기는 28.8 \times 9.6~12.8 μm 이며, 초기에 소실된다. 자낭포자는 단세포로 올리브빛 갈색을 띠며, 매끈한 벽을 가진다. 양 끝에 발아공이 있는 넓은 방추형으로 크기는 12~13.6 \times 6.8~8.0 μm 이다. 분생자는 구형의 단세포로 짧은 분생자형성세포에서 단일출아를 한다. 두꺼운 세포벽을 가지며 발생 초기에는 무색으로 매끈하나 말기에는 연

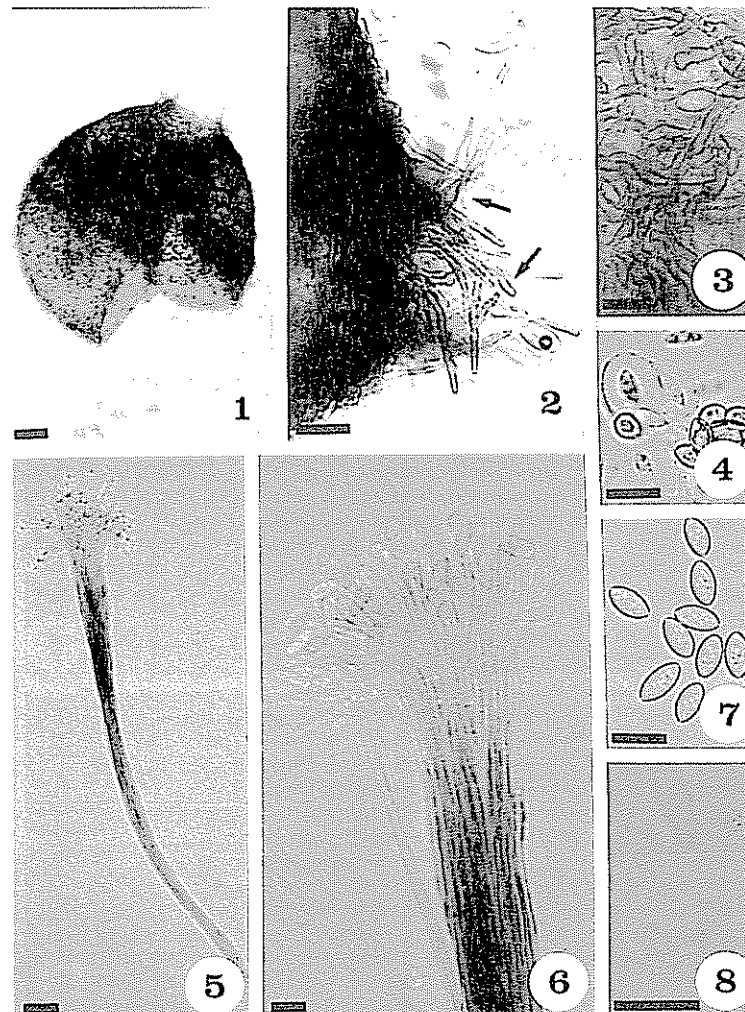


Fig. 2. Morphological features of *Petriella sordida*. 1, A mature ascoma crushed by pressure; 2, Short ostiolar neck with setae-like hyphae (arrows); 3, Ascomatal wall with interwoven hyphae; 4, Evanescent asci expanded by osmosis; 5, Synnema of anamorphic state; 6, The head part of synnema with hyaline conidia; 7, Light brown ascospores with germ pores at each end; 8, Conidia. (bar in Figs. 1, 5=50 μm ; Figs. 2, 3, 4, 6, 7=10 μm)

갈색을 띠며, 세포벽에 미세 돌기들이 붙어 있다. 크기는 직경 $7.2\sim9.6\ \mu\text{m}$ 이다.

Petriella sordida (Zukal) Barron & Gilman (Fig. 2)

Ascomata ostiolate, spherical perithecia, dark brown. Ostiolar neck short, with setae-like hyphae. Ascomatal wall dark brown, composed of interwoven hyphae. Asci fasciculate, evanescent, obovate to broadly clavate, 8-spored, $22\sim32\times12\sim20\ \mu\text{m}$. Ascospores

dextrinoid when young; pale reddish brown when mature, copper colored mass, furnished with several oil drops, $9\sim11\times4.5\sim6\ \mu\text{m}$. Anamorph observed, *Graphium* sp. synnematosus, with an apical brushes of conidiogenous cells, conidiogenous cells cylindrical or tapering, conidia blastosporous, cylindrical or slightly clavate, with a rounded apex, attenuated and truncate at base, aseptate, hyaline, $6\sim12\times3\sim5\ \mu\text{m}$.

Collected from soil of muskmelon-growing

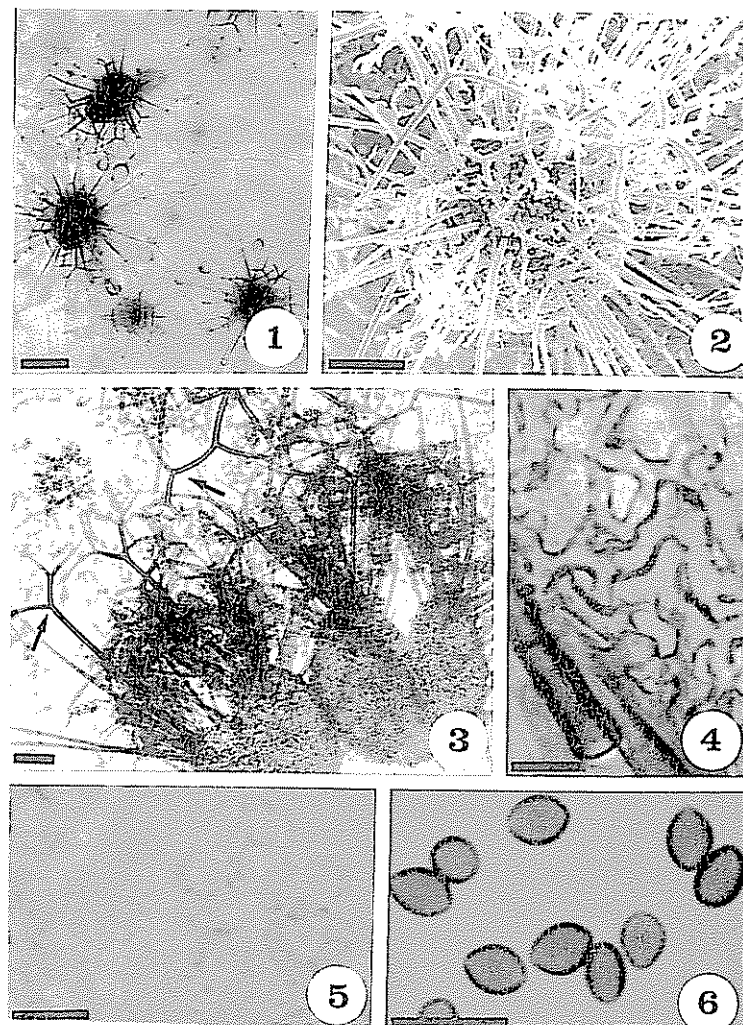


Fig. 3. Morphological features of *Chaetomium indicum*. 1, Ascomata on the agar plate; 2, Scanning electron microscope view of an ascoma with branched hairs and pushing out ascospores; 3, Ascomata with right angled branched hairs around ostiole; 4, Ascomatal wall of textura intricata; 5, A cluster of asci; 6, Dark brown ascospores. (bar in Fig. 1= $100\ \mu\text{m}$; Figs. 2, 3= $50\ \mu\text{m}$; Figs. 4-8= $10\ \mu\text{m}$)

greenhouse, Jinju-City, Kyungnam province, May, 1997. Isolate deposited in KACC (#40306).

자낭과는 진한 갈색을 띠는 구형의 유공자낭각이다. 유공경은 짧고, 강모와 같은 균사들로 구성되어 있다. 자낭과 외벽은 여러 개의 균사체가 엉겨 있으며, 암갈색을 띤다. 자낭은 총생하며 초기에 소실된다. 넓은 곤봉형 또는 역난형의 모양을 하고 8 개의 포자를 가진다. 크기는 $22\sim32\times12\sim20\ \mu\text{m}$ 이다. 자낭포자는 발생 초기에 요오드액에 보라색으로 염색이 되며, 성숙기에 붉은 빛을 띠는 갈색으로 변화한다. 뭉쳐진 포자덩어리는 구릿빛을 띠며 포자 내부

에는 6~7개의 기름액포가 있다. 양 끝에 발아공을 가지며 크기는 $9\sim11\times4.5\sim6\ \mu\text{m}$ 이다. 불완전세대가 함께 발견되었다. 분생자는 분생자경속 상부의 분생자 형성 세포에서 출아하는 단세포이다. 한쪽 끝이 가늘어지는 원통형 모양을 하며 무색을 띠고, 크기는 $6\sim12\times3\sim5\ \mu\text{m}$ 이다

Chaetomium indicum Corda (Fig. 3)

Ascomata ostiolate, globose perithecia, superficial, gregarious, dark brown to black, clothed with hairs. Later hairs dark brown, straight, simple. Terminal hairs dark oliv-

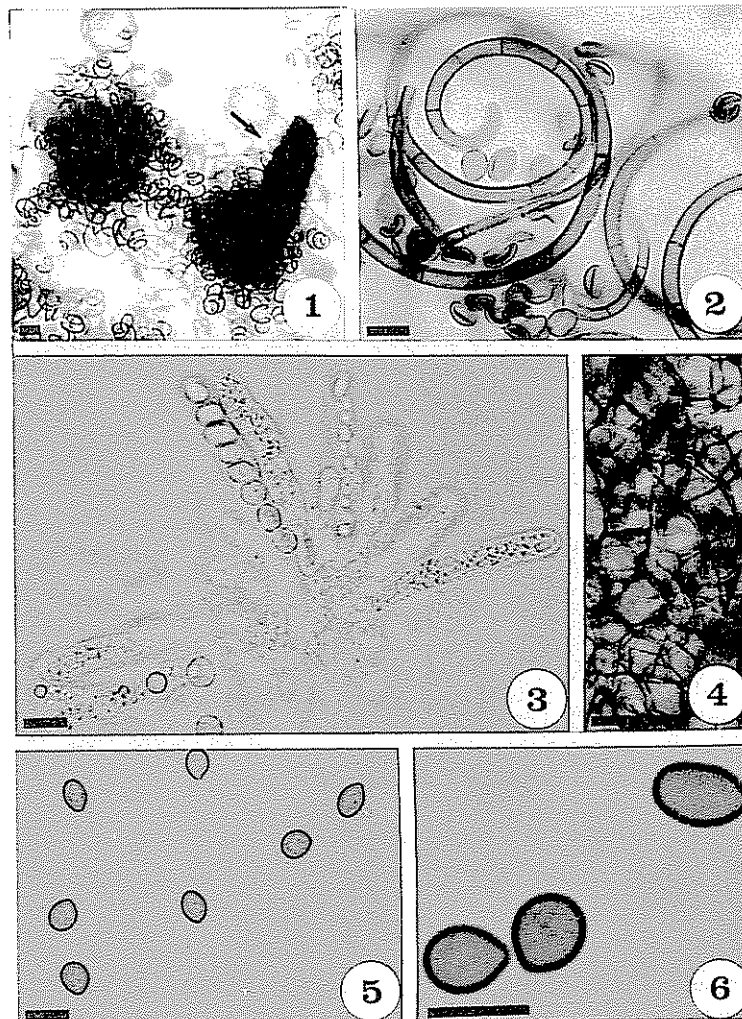


Fig. 4. Morphological features of *Chaetomium brasiliense*. 1, Ascomata with a cirrus (arrow) on V8 agar plate; 2, Curved terminal hairs; 3, Asci with ascospores; 4, Ascomatal wall of textura angularis; 5, Mature ascospores; 6, Close-up ascospores. (bar in Figs. 1, 4=50 μm ; Figs. 2, 3, 5, 6=10 μm)

aceous brown, septate, dichotomously branched in the upper half, with branches at right angle, smooth to finely roughened, thick-walled, 4-(4.3)-6 μm width at base. Ascumatal wall translucent, covered with intricating hyphae. Asci fasciculate, 8-spored, stalked, multi-seriate, clavate to broadly clavate, with evanescent walls, 22-30 \times 8-11 μm . Ascospores unicellular, initially hyaline to light brown, dark brown at maturity, flattened lemon-

shaped, with an apical germ pore, pushed out of ostiole in a cirrus, 5-6 \times 4-6 μm . Anamorph not observed.

Collected from soil of muskmelon-growing greenhouse, Jinju-City, Kyungnam province, May, 1997. Isolate deposited in KACC (#40307).

자낭과는 진한 갈색 또는 흑색을 띠며 구형의 유공자낭각이다. 자낭각 외벽은 교차균사 조직상을 하며 반투명이다. 측면 모상균사는 진한 갈색으로 끝으며 단순조각이다. 상부 모상균사는 진한 올리브

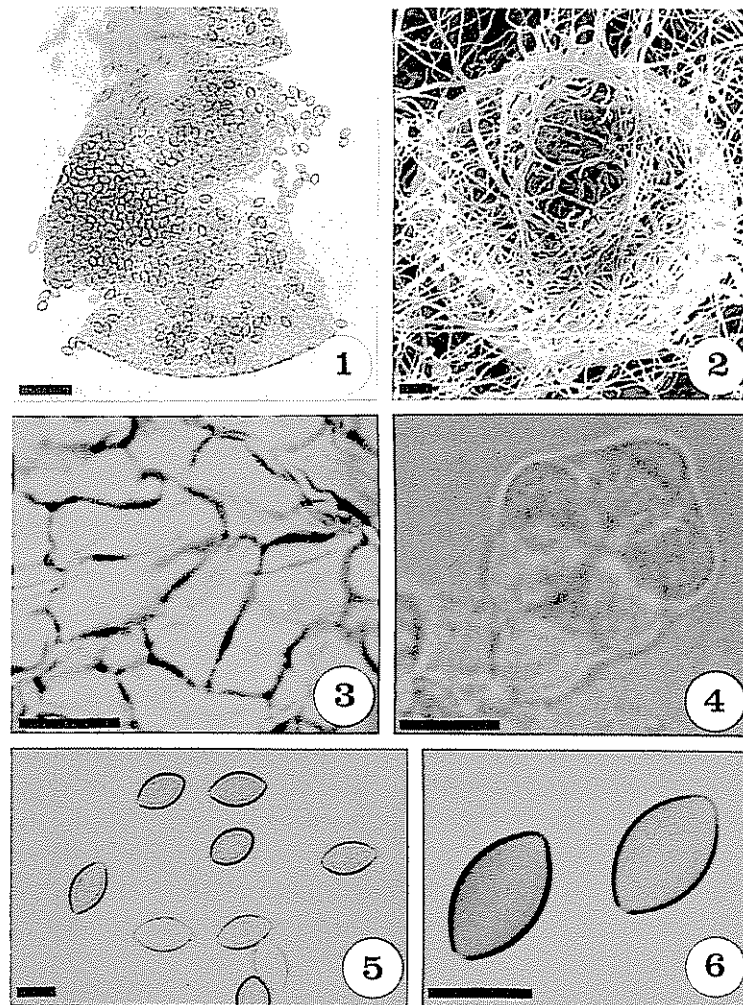


Fig. 5. Morphological features of *Thielavia fragilis*. 1, A crushed ascoma filled with ascospores; 2, Scanning electron microscopic feature of immature ascoma composed of interwoven hyphae; 3, Ascomatal wall of textura epidermoidea; 4, An ascus containing immature spores; 5, Mature ascospores of olivaceous brown color and hyaline immature ascospores; 6, Ascospores with an apical germ pore. (bar in Fig. 1=50 μm ; Figs. 2-6=10 μm)

빛의 갈색을 띠며 상부 영역에서 직각으로 분지되어 있다. 하단부 직경이 4~(4.3)~6 μm 이다. 자낭은 총생하며 곤봉형 또는 넓은 곤봉형이며, 초기에 소실되는 얇은 벽을 가진다. 크기는 22~30 \times 8~11 μm 이다. 자낭포자는 올리브빛 갈색 또는 암갈색으로 납작한 레몬 모양을 하고 있다. 크기는 5~6 \times 4~6 μm 이며 정단부에 1 개의 발아공을 가진다. 일단의 성숙된 포자들은 포자각의 형태로 유공에서 밀려나온다. 불완전세대는 관찰되지 않았다.

***Chaetomium brasiliense* Batista & Pontual (Fig. 4)**

Ascomata ostiolate, spherical perithecia, dark brown, clothed with hairs. Ascomatal wall dark brown, composed of 3~4 cells, textura angularis. Lateral hairs short, straight, simple. Terminal hairs spirally coiled in the upper part, septate, dark brown, smooth. Asci in a fascicle, cylindrical, with a stalk, 8-spored, uniseriate, rather persistent, 45~55 \times 6.5~8 μm . Ascospores ovate to nearly spherical, bilaterally flattened, dark brown when mature, with a germ pore at the attenuated end, 7~8.5 \times 5~6 μm . Anamorph not observed

Collected from soil of muskmelon-growing greenhouse, Jinju-City, Kyungnam province, May, 1997. Isolate deposited in KACC (#40309).

자낭과는 유공자낭각으로 진한 갈색을 띠며 모상균사로 덮여 있다. 자낭각 외벽은 진한 갈색으로 3~4개의 세포로 구성되어 있고, 다각균사 조직상을 하고 있다. 측면 모상균사는 짧고 곧으며, 단순구조를 하고 있다. 상부 모상균사는 상단 부위에서 코일 모양을 하며, 진한 갈색으로 격막을 가지고 매끈하다. 자낭은 총생을 하며, 원통형으로 짧은 대를 가지고 있다. 8개의 포자를 가지며, 초기에 소실되지 않고 영속한다. 크기는 45~55 \times 6.5~8 μm 이다. 자낭포자는 역난형 또는 구형이며, 약간 납작하고, 가늘어지는 한쪽 끝에 발아공을 가지고 있다. 자낭 내에서 일렬 배열을 하고 있다. 발생 말기에 진한 갈색을 띠며, 7~8.5 \times 5~6 μm 정도의 크기이다. 불완전세대는 관찰되지 않았다.

***Thielavia fragilis* (Natarajan) v. Arx (Fig. 5)**

Ascomata pale brown, nonostiolate, transparent, spherical cleistothecia, glabrous, scat-

tered, superficial. Ascomatal wall pale brown, composed of 3~4 flattened cells, textura epidermoidea. Asci in a fascicle, clavate, 8-spored, multiseriate, stipitate, with a thin evanescent wall. Ascospores 1-celled, hyaline at young, olivaceous to dark brown at maturity, clavate to obovate, rounded at each end, with an apical germ pore, 12~14 \times 7~8 μm . Anamorph not observed.

Collected from soil of muskmelon-growing greenhouse, Jinju-City, Kyungnam province, May, 1997. Isolate deposited in KACC (#40310).

자낭과는 연한 갈색의 투명한 구형의 폐쇄 자낭각으로 표면이 매끈하다. 자낭각 외벽은 얇은 갈색을 띤 3~4개의 세포로 표피상균사조직상으로 구성되어 있다. 자낭은 총생을 하며, 8개의 포자를 가지고 있다. 곤봉형 또는 역난형이며, 초기에 소실되는 얇은 세포벽을 가진다. 자낭포자는 단세포로 발생 초기에 투명하거나 연한 갈색을 띠며, 올리브빛 갈색 또는 진한 갈색으로 변화한다. 이들은 자낭 내에서 다층 배열을 하며 넓은 방추형 또는 타원형으로 양 끝에 2개의 발아공을 가지고 있다. 크기는 12~14 \times 7~8 μm 이다. 불완전세대는 관찰되지 않았다.

적 요

토양 회석방법을 이용하여 토양에 분포하는 곰팡이에 대한 분포 조사를 실시하였다. 토양 시료는 경상남도 진주시에 분포하는 머스크멜론 시설 재배지에서 채취하였으며 총 7종의 자낭균을 분리, 동정하였다. 이들 중 Sordariales목에 속하는 5종의 균을 국내에 처음 보고하며 이들의 형태적 특징을 기재하였다. 이들 균은 완전세대와 불완전세대를 동시에 가지는 *Corynascus sepedonium*과 *Petriella sordida*와 완전세대만을 가지는 *Thielavia fragilis*, *Chaetomium indicum*과 *Chaetomium brasiliense*이다. 본 논문에 기재된 균들은 농업과 학기술원 농용미생물 보존센터에 보관하였다.

References

- Alexopoulos, C. J., Mims, C. W. and Blackwell, M. 1996. Introductory Mycology. John Wiley & Sons, Inc.
- Arx, J. A. von. 1975. On *Thielavia* and some

- similar genera of ascomycetes. *Studies in Mycology*. 8: 1-33.
- Arx, J. A. von, Guarro, J. and Figueras, M. J. 1986. The ascomycete genus *Chaetomium*. *Beihefte zur Nova Hedwigia*. Heft 84.
- Barr, M. E. 1976. Perspectives in the Ascomycotina. *Mem. N. Y. Bot. Gard.* 28: 1-8.
- . 1990. Prodromus to Nonlichenized, Pyrenomycetous members of class Hymenozomycetes. *Mycotaxon* 38: 43-184.
- , ———, and ———. 1988. Sordariaceous Ascomycetes without ascospores ejaculation. *Beihefte zur Nova Hedwigia* 94: 1-104.
- Cho, D. H. and Chun, J. K. 1962. Studies on the mold flora in the several degraded paddy soils (choorak). *J. Korean Agr. Chem. Soc.* 3: 17-18.
- Hawksworth, D. L., Kirk, P. M., Sutton, B. C., and Pegler, D. N. 1995. Ainsworth & Bisby's Dictionary of The Fungi. 8th ed. International Mycological Institute. CABI.
- Huang, L. H. 1976. Developmental morphology of *Triangularia backusii* (Sordariaceae). *Canad. J. Bot.* 54: 250-267.
- Kim, K. S. 1979. The standing crops and soil-borne microfungus flora of *Phyllostachys reticulata* in Korea. *Kor. J. Mycol.* 7(2): 91-116.
- and Lee, J. Y. 1980. Soil-borne fungi of *Phyllostachys reticulata* Forests in Korea. II. *Kor. J. Mycol.* 8(1): 45-51.
- Lee, S. K., Kim, S. H. and Kim, M. S. 1987a. Studies on the antibiotic microorganisms in soil. 1. Distribution and identification of antibiotic soil microorganisms in different soil parent materials. *Res. Rept. RDA (P. M. & U.)* 29(1): 179-184.
- , Yoon, S. Y., Ryu, J. C., Suh, J. S., and Park, J. K. 1987b. Studies on the changes of soil microorganisms and their activity in continuation and rotation of vegetable crops in upland soil. *Res. Rept. RDA (H)* 29(2): 85-92.
- , Suh, J. S., Mun, J. H., and Song, C. H. 1988. Studies on identification and enumeration of soil microorganisms in mineral and volcanic ash soil of Jeju Island. *J. Korean Soc. Soil Sci. Fert.* 21(2): 135-140.
- Lee, S. B., Choi, Y. H., Lee, K. B., Yoo, C. H., and Rhee, G. S. 1995. Seasonal changes of microflora in paddy soil with long-term application of organic matter. *J. Korean Soc. Soil Sci. Fert.* 28(4): 356-362.
- Luttrell, E. S. 1951. Taxonomy of the Pyrenomycetes. *Univ. Missouri Stud. Sci. Ser.* 24(3): 1-120.
- Mai, S. H. 1976. Morphological studies in *Podospora anserina*. *Am. J. Bot.* 63: 821-825.
- Min, K. H., Hong, S. W., and Yokoyama, T. 1980a. Hyphomycetes from Korean Soil. I. The genus *Penicillium* with a teleomorphic state *Eupenicillium javanicum*. *Kor. J. Microbiol.* 18: 91-103.
- , ———, and ———. 1980b. Hyphomycetes from Korean Soil. II. The genus *Aspergillus* and some other microfungi. *Kor. J. Microbiol.* 18: 104-114.
- , Ito, T. and ———. 1981. Fungus flora of paddy fields in Korea. I. Fungal distribution of paddy fields. *Kor. J. Microbiol.* 19(4): 153-162.
- , ———, and ———. 1982a. Fungus flora of paddy fields in Korea. II. Fungal flora of paddy fields. *Kor. J. Microbiol.* 20(1): 41-51.
- , ———, and ———. 1982b. Fungus flora of paddy fields in Korea. III. Ascomycetes. *Kor. J. Microbiol.* 20(2): 80-88.
- , ———, and ———. 1987. Fungus flora of paddy fields in Korea. IV. Filamentous fungi isolated by heat treatment. *Kor. J. Mycol.* 15(3): 187-195.
- Yoo, I. D., Yun, S. Y., Lee, M. G. and Ryu, J. C. 1983. Studies on microflora of the paddy and upland soils of Korea. I. Distribution of microflora of the paddy soils. *J. Korean Soc. Soil Sci. Fert.* 16(2): 195-202.