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The Institute for Fermentation, Osaka publishes the IFO Research Communications on a biennial basis. Purchase orders of the Research Communications should be addressed to The Institute for Fermentation, Osaka, 17-85 Juso-honmachi 2-chome, Yodogawa-ku, Osaka 532, Japan.

Prices of back numbers are as follows: Nos. 1-5, ¥500 each; No. 6, ¥800; No. 7, ¥1,000; Nos.8-9, ¥800 each; No. 10, ¥1,000; and Nos. 11-12, ¥1,300, and plus postage.

REPORT OF THE DIRECTOR

Since the establishment of a cell line section in 1984, the expansion of laboratories for cell line research and the renewal of facilities have been planned, the planning being completed in November 1986. New staff has joined the institute to engage in preservation of and research into cell lines. The curator of cell lines, Dr. Masao Takeuchi, attended the ATCC Workshop on "Freezing and Quality control of cell cultures and hybridoma" held in October 1985. He acquired up-to-date techniques and procedures on animal cell lines during the workshop. After the workshop he spent several days at ATCC studying the refined techniques in preserving and controlling quality at ATCC. He also visited some of the cell line collections in the U.S.A.

His achievements have helped us to check mycoplasmic contamination in cell line in IFO and the Japanese Cancer Research Resources Bank (JCRB). IFO staff in cell line section have started to check mycoplasmic contamination and to analyse isozymes in cell lines stored in Japan, in cooperation with JCRB. They are also checking IFO cell lines, and the resultant mycoplasma-free cell lines are listed in the catalogue of cultures published in March 1986 as a supplement to the 7th edition of "IFO List of Cultures".

At the 80th annual meeting of the Board of Trustees in June 1985, Professor Teruhiko Beppu of the University of Tokyo was nominated as a new councilor. The amendment of the articles of the Act of Endowment was approved at the meeting, allowing the expansion of activities to animal cell lines. The chairman of the Board of Trustees, Mr. Shinbei Konishi, received a new fund amounting to ¥150 million from Takeda Chemical Industries Ltd. in March 1986. Addition of the fund to the foundation of IFO was approved at the 81st annual meeting of the Board of Trustees in March 1986. A similar donation was received in 1984 for the promotion of the establishment of the cell line section. The new funds will help to increase the total activities of IFO. As the result of the donation from the company, the total foundation of the institute reached ¥805,400,000.

The treasurer of the institute, Mr. Tokusaburo Fujitani retired

from the institute in December 1986 after four years of dedicated service, and Mr. Masayuki Yamada was appointed as treasurer on December 1, 1986.

Dr. Akira Yokota of the bacteriological section is staying at the Max-Planck-Institute for Immunobiology in Freiburg, West Germany, as a visiting researcher under Dr. Hubert Mayer for one year, to do research on the analysis of the chemical structure of lipids in chemolithotropic bacteria and its possible phylogenetic relationship.

The International Institute of Refrigeration (IIR), Commission C1 held a meeting on "Fundamentals and applications of Freeze-drying to biological materials, drugs and food staffs" from May 20 to 23, 1985 in Tokyo, with the cooperation of the Japanese Association of Refrigeration and the Japanese Society for Research of Freezing and Drying. Dr. T. Iijima, president of JSRFD, and a member of the local organizing committee (chairman: Professor emeritus Shozo Koga), attended the meeting and presented a paper on additives to prevent mutation. Dr. K. Imai also attended and presented a paper on the method of preservation of chemolithotropic bacteria.

The Committee for the Development of Mycology in Asian countries of International Mycological Association (IMA), met on March 13 to 15, 1986 in Kuala Lumpur, Malaysia. Dr. Tatuo Yokoyama, the secretary of the committee, attended the meeting and presented a paper introducing the activities of the fungal section of IFO. In September 1986, he was invited by Prof. Yin Yu Qi of Shihezi Agricultural College and Prof. Guo Shijian of the Ministry of Agriculture, Animal Husbadry and Fishery in Beijing, to spend one month in Shihezi Agricultural College, in Shihezi, Xinjiang, China, to lecture on the taxonomy of soil microfungi and collect soil samples in these districts. Isolation of fungi from the samples and the establishment of a new collection are now in progress with the cooperation of both institutes.

The 5th Korean-Japan Fermentation Technology Symposium on "Screening, improvement and preservation of industrial microorganisms" was held on November 4, 1986 in Seoul, Korea. Director T. Iijima was invited as a speaker and he lectured on practical management of a culture collection.

The international training course on "Bioinformatics: Data management and computer usage in culture collections" was held from November 26, 1986 in Osaka and Tokyo with 13 participants from 10

countries. The training course was supported by UNESCO, WFCC, WDC-MIRCEN, International Center of Biotech, Osaka University and JFCC. JFCC sustaining member and many Japanese companies provided financial supports for the training course. Director T. Iijima, president of JFCC, and staff of IFO, Drs. I. Banno, T. Kusaka and K. Imai, attended the training course and gave lectures and training on computers to the participants. All participants were active and enthusiastic to aquire knowledge on computer usage. The training course ended on December 5 in Tokyo as a great success.

The total number of cultures stored in the IFO culture collection reached 12,300 at the end of 1985 and 12,500 at the end of 1986. The newly accepted strains during these two years are listed in the present issue of IFO Research Communications. The total number of cultures distributed from the IFO culture collection reached to 9,700 in 1985 and 9,900 in 1986. The computerized managing of distribution of strains started in 1984 has been working effectively during the past two years.

Procedures for distribution of phytopathogenic strians in Japan and to abroad were considered and were established at the end of 1985. A number of phytopathogenic strains preserved in IFO can be distributed to customers who receive permission and certification from the phytoquarantine office as having suitable facilities and techniques for handling phytopathogenic strains.

IFO has welcomed a number of guests in the past two years. Lectures and seminars were given by the following guests.

Prof. H. Nakamura, Konan University : Sexuality of Procaryotes; Its origin and evolution.

Prof. K. Kinugawa, Faculty of Agriculture, Kinki University : Breeding of Mushroom, Flammulina.

Dr. H. J. Willetts, Universtiy of New South Wales, Australia : Taxonomic studies on Sclerotinia and Monilinia species using isoelectric focussing of extracellular cell wall degrading enzyme.

Dr. H. P. Upadhyay, Department of Mycology, Federal University of Pernambuco, Brazil : Taxonomy and ecology of nematophagous fungi.

Dr. K. Sugawara, The Institute of Physical and Chemical Research (Riken) : Data management and computer usages in culture collection.

Members of the institute have participated in mycological forrays

in Japan. Mr. T. Ito joined the foray of the Mycological Society of Japan in Mt. Daisen in September 1986.

The committee for confirmation of ISP (International Streptomyces Project) strains conducted regular confirmatory tests of ISP strains stored in IFO. As a result of the confirmation, the need for a revised manual was discussed and a new manual was prepared by the committee.

(T. Iijima)

Heartfelt condolences are extended to bereaved

Dr. Sueo Tatsuoka, who passed away on 4th July, 1985.

Professor emeritus Gyozo Terui, who passed away on 28th May, 1986 They made great contributions to the establishment and the development of the Institute for Fermentation, Osaka.

IFO Res. Comm.13,
5-12, 1987 (March)

PLASMID-POSSESSING ACTINOMYCETES IN IFO

TAIKI KUSAKA and KUNIKO SATO

Summary

Actinomycetes strains available from the IFO Culture Collection were screened for possession of plasmid DNA using the alkali micro-method and agarose gel electrophoresis. Some bands in the lane of agarose gel were detected in 14 strains (10 strains of Streptomyces; 3 strains of Rhodococcus; 1 strain of Nocardia) among the 989 strains tested. These were considered to be plasmid DNA bands both because of their reproducible detection and their estimated molecular weights of around 23 Kb. These strains can be preserved in dry or frozen states, as well as being transplantable, for several years without loss of their plasmids.

A number of biological properties of actinomycetes strains in the IFO Culture Collection have been previously reported (7,8). The recent dramatic developments in biotechnology require a variety of plasmids obtained from various kinds of microorganisms. In response to this need and in order to expand the amount of information about strains in the IFO Culture Collection, the existence of plasmid was examined using these strains.

This paper deals with the strains possessing plasmids and the possibility of their long-term preservation.

Materials and Methods

Actinomycetes strains. A total of 989 actinomycetes strains in the Culture Collection of the Institute for Fermentation, Osaka (IFO) were tested. There were in 47 genera and 708 species, as shown in Table 1. They were maintained in our laboratory by transplantation on agar slant media at intervals of 6 months. The presence of plasmids was confirmed by reviving fourteen strains as shown in Table 1, from the L-dried ampoules (5) which had been prepared 2 - 5 years before. In the preservation tests, three kinds of samples of each of the five species were used; namely, L-dried (12), frozen at -80 C (9) and agar transplanted strains.

Media. A total of thirteen different media were used and their components were reviewed in the reference (4). The designated medium for each strain was used for the growth of each strain tested.

Chemicals. Sodium dodecylsulfate, bromophenol blue and λDNA were purchased from Wako Pure Chemical Industries, Ltd. Agarose (Type II), RNAase A, ethidium bromide and lysozyme (muramidase) were purchased from Sigma Chemical Company. Hind III restriction endonuclease were obtained from Takara Shuzo Co., Ltd.

Detection of plasmid band. The method for the detection of plasmids (1,10) was slightly modified. Mycelia and spores grown on agar plate for ten days or two weeks were scratched from a 5 cm^2 area of the plate and suspended in 0.2 ml of a solution (0.1 M glucose, 10 mM EDTA, 25 mM Tris-HCl, pH 8.0) containing lysozyme (15 mg/ml). After lysing for 30 min at room temperature, 0.4 ml of a solution (10% SDS and 0.4 N NaOH) was added to the mixture to burst the cells. After 5 min, it was neutralized with 0.25 ml of acetate buffer (3 M Na acetate, pH 4.8) and centrifugated at 12,000 rpm for 5 min. DNA was precipitated with cold ethanol from the supernatant. The precipitate was dissolved in 0.2 ml of Solution I (0.1 M Na acetate, 0.05 M Tris-HCl, pH 8.0) and 0.2 ml of phenol solution (phenol 25 ml, $CHCl_3$ 24 ml, isoamylalcohol 1 ml) was added to precipitate protein. After maintenance at -20 C for 30 min, the solution was centrifugated at 12,000 rpm for 10 min. Cold ethanol was added to the supernate and its precipitant was dissolved in Solution I to which 2 times the volume of cold ethanol was again added for further purification. The precipitant was redissolved in 30 μl of 10 mM Tris-EDTA buffer (pH 8.0) and treated with 5 μl of RNAase solution (100 μg/ml) at 37 C for 30 min to eliminate RNA.

Table 2. Plasmid possessing actinomycetes strains and related migration distance of plasmids.

Name of taxon	IFO Number	Date of L-dried	Ratio of immigration distances
1) Streptomyces phaeochrmogenes	3105	70 01 05	0.68, 1.26.
2) Streptomyces olivaceus	3152	70 01 05	1.00, 1.42, 1.57.
3) Rhodococcus rhodochrous	3338	83 01 05	1.00
4) Streptomyces roseochromogenes	3442	83 01 07	0.52, 0.57, 0.68, 0.79, 1.00, 1.36, 2.10.
5) Streptomyces neyagawaensis	3784	83 01 07	0.84, 1.00, 1.58, 2.00, 2.21.
6) Nocardia orientalis	12362	83 01 13	0.68, 0.79, 1.00, 1.42.
7) Rhodococcus erythropolis	12682	77 07 14	1.05, 2.10.
8) Streptomyces arenae	13016	83 06 24	0.78, 1.39.
9) Streptomyces intermedius	13049	83 06 26	0.39, 0.61, 0.83, 1.21, 1.48.
10) Streptomyces roseochromogenes	13080	83 04 26	0.57, 0.70, 0.74, 0.87, 1.17.
11) Streptomyces luteolutescens	13489	83 01 21	0.52, 0.87, 1.04.
12) Streptomyces castaneoglobisporus	13669	84 07 04	0.48, 1.00, 1.57.
13) Streptomyces xylophagus	13845	84 08 17	0.67, 0.91, 1.26.
14) Rhodococcus marinonascens	14363	84 08 24	0.91.
λ DNA fragments by Hind III			1.00, 1.30, 1.47, 1.73, 2.52, 2.69.

Agarose gel electrophoresis. Plasmid DNA was analyzed by electrophoresis for 4.5 hr at room temperature, at 100 V, 40 - 50 mA using a Tris-Borate buffer (89 mM Tris HCl, 89 mM Boric acid, 2.5 mM EDTA 2Na, pH 8.3) and bromophenol blue as a marker with λDNA restricted by Hind III as reference. Following this, the gel was stained with 0.1% ethidium bromide for 15 min and then photographed under 260 nm UV illumination. Thus an electrophoresis profile was obtained.

Record of plasmid band. Some bands of the strain and the reference fragments of λDNA were detected on the agarose gel. The migration distance of each band was expressed relative to that of the largest standard fragment of λDNA.

Results

Some bands were reproducible and had migrated about the same distances as the standard fragments of λ DNA. They were considered to be plasmid DNA bands. The generic distribution of all 989 strains tested is

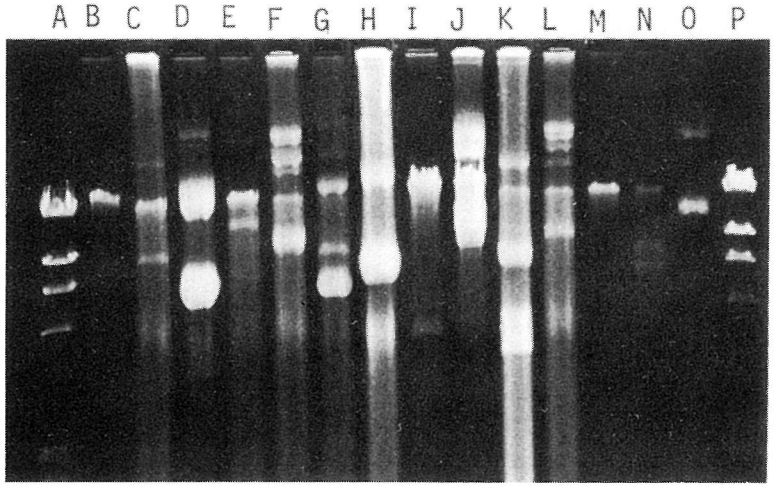

Fig. 1. Electrophoresis profiles of plasmid-possessing actinomycetes strains.

A, λDNA	B, IFO 14363	C, IFO 13845	D, IFO 13669
E, IFO 13489	F, IFO 13080	G, IFO 13049	H, IFO 13016
I, IFO 12682	J, IFO 12362	K, IFO 3784	L, IFO 3442
M, IFO 3338	N, IFO 3152	O, IFO 3105	P, λDNA

Table 1. Number of strains tested and plasmid-possessing strains.

Generic name	Species	Strains	Plasmid-possessing strains
Actinoalloteichus	1	1	0
Actinobifida	1	1	0
Actinomadura	11	13	0
Actinomyces	24	24	0
Actinoplanes	24	27	0
Actinopolyspora	1	1	0
Actinopycnidium	1	1	0
Acinosynnema	1	1	0
Agromyces	1	2	0
Amorphosporangium	3	3	0
Ampullariella	7	8	0
Catenuloplanes	1	2	0
Chainia	4	6	0
Dactylosporangium	6	6	0
Dermatophilus	1	1	0
Faenia	1	2	0
Geodermatophilus	1	4	0
Intrasporangium	1	1	0
Kineosporia	1	2	0
Kitasatoa	5	5	0
Kitasatosporia	3	3	0
Microbispora	19	13	0
Microellobosporia	3	3	0
Micromonospora	20	28	0
Micropolyspora	3	3	0
Microtetraspora	3	4	0
Nocardia	16	23	1
Nocardioides	4	6	0
Nocardiopsis	5	5	0
Oerskovia	1	1	0
Planobispora	1	2	0
Planomonospora	1	1	0
Promicromonospora	1	1	0
Pseudonocardia	2	2	0
Rhodococcus	3	7	3
Rothia	1	2	0
Saccharomonospora	1	1	0
Saccharothrix	1	1	0
Saccharopolyspora	1	1	0
Spirillospora	1	1	0
Sporichthya	1	1	0
Streptoalloteichus	1	1	0
Streptomyces	461	682	10
Streptosporangium	10	13	0
Streptoverticillium	41	62	0
Thermoactinomyces	4	7	0
Thermomonospora	4	4	0
Total	708	989	14

shown in Table 1. The number of actinomycetes strains which showed bands was 14, as is shown in Table 2. The same electrophoresis profiles (Fig. 1) were obtained from the L-dried ampoules of these 14 strains. Strains possessing plasmid(s) were found in three genera, Streptomyces, Rhodococcus and Nocardia among the 47 genera tested. Two strains, Rhodococcus rhodochrous IFO 3338, and R. marinonascens IFO 14363, showed plasmid bands at the same or a somewhat shorter distance than the 23 Kb fragment of λ DNA, and the other 12 strains showed multiple bands in locations of both shorter and longer distances than this fragment. The number of plasmid bands varied from one to seven depending on the strain, although some bands could not be clearly discriminated because of a small amount of plasmid DNA, a short migration distance or smeared DNA. Five plasmid-positive strains were chosen for further tests of the plasmid preservation methods. Figure 2 shows the profiles of samples of the five strains each preserved by the three methods. Nearly the same profile of

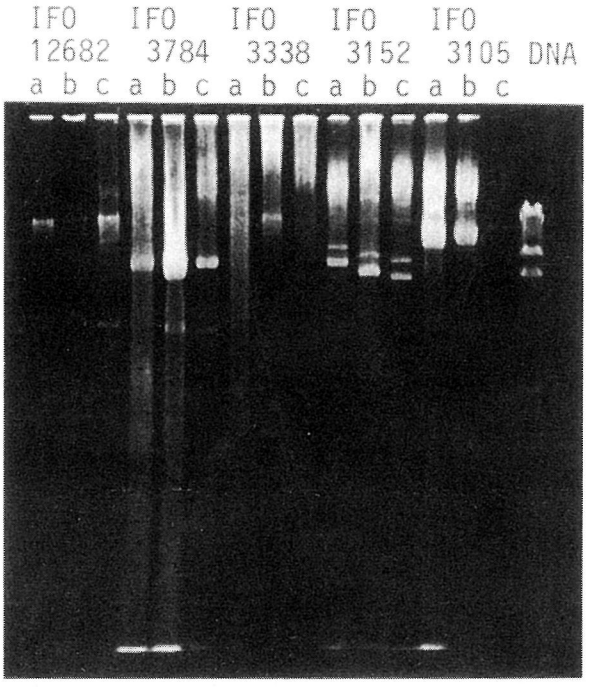

Fig. 2. Electrophoresis profiles of plasmid-possessing actinomycetes strains preserved by different methods.
a; preservation of cultured broth at -80 C.
b; transplanting agar slant cultures.
c; L-dried preservation.

each strain was detected on the agarose gel regardless of the preservation method, although a little difference in the amount of plasmids was observed.

Discussion

Reproducible plasmid DNA bands have been detected in 21 of 120 wild-type Streptomyces strains analyzed (17.5%), though in 7 of the 21 strains these might be fragments of chromosomal DNA (2). Agarose gel electrophoresis of cleared lysates of 110 Streptomyces strains revealed satelite bands indicative of plasmids in 8 strains (7.2%) (3). In these reports, 7 - 18% of Streptomyces strains had plasmids; but the percentage of strains other than Streptomyces that bear plasmids is unclear. Variations in plasmid yield may be considered due to culture age, transition of plasmid forms, activities of DNAase and extraction efficiency of plasmids, especially from strains other than Streptomyces (11). In our study, the proportion of plasmid-positive strains in the total of 989 strains tested was 1.4%. This is lower than was previously reported, even though substantially the same isolation method was adopted. The reason for this may be our strict exclusion of strains showing bands indicating fragmented chromosomal DNA or smeared DNA bands covering plasmid bands. The ratio may be increased by development of more efficient isolation methods for strains other than Streptomyces or inhibition methods to reduce the DNAase activity of the lyzed cells and eliminate smeared bands on the agarose gel. During long-term storage or transplanting maintenance in a laboratory, strains may lose their plasmids (11). However, the strong possibility of long-term preservation of plasmids was shown by this experiment. If strains are preserved under sufficiently dry or low-temperature conditions to make their metabolism static, deletion or exclusion of plasmids from their cells will not occur. In fact, four yeast strains carrying recombinant plasmid were reported to have been preserved by L-drying a long period without loss of the plasmids (6). It is concluded that among 989 actinomycetes strains, plasmid DNA band(s) are reproducible in 14 strains and that, in strains other than Streptomyces, plasmid-possessing strains are very few. In addition, those plasmids are readily preserved for several years in the

original microbe under L-dried or frozen conditions as well as by transplanting.

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13-16, 1987 (March)

BACTERIAL FLORA IN DADIH

KO IMAI, MARIKO TAKEUCHI, TAKESHI SAKANE and INDRAWATI GANDJAR*

Summary

Bacteria in dadih, a fermented product made from buffalo milk were examined. One gram of dadih contained about 7.5×10^8 bacteria, of which the major species were Lactobacillus casei subsp. casei and Lactobacillus plantarum.

Dadih or dadiah is an Indian fermented milk somewhat like yoghurt. It had been reported that yeasts and lactic acid bacteria were the microorganisms responsible for the fermentation (4). We have examined the bacterial flora in dadih obtained in Western Sumatra, Indonesia, the microorganisms in which have been reported by Gandjar, partly on our examination (3). Taxonomic reexamination of bacterial strains isolated indicated that the major species in dadih were Lactobacillus casei subsp. casei and Lactobacillus plantarum. This communication describes the results of a detailed examination of the bacteria in dadih.

Materials and Methods

Media. Peptone-yeast extract medium (PY) contained 5 g of glucose, 5 g of lactose, 10 g of peptone, 2 g of yeast extract, 2 g of NaCl, 0.5 g of Tween 80 and 1,000 ml of distilled water and was adjusted to pH 7.0

* Department of Biology, University of Indonesia.

with NaOH. Basal medium (BM) consisted 3 g of KH_2PO_4, 7 g of K_2HPO_4, 1 g of $(NH_4)_2SO_4$, 0.1 g of $MgSO_4 7H_2O$, and 2 g of NaCl and 1,000 ml of distilled water. To prevent the growth of fungi, Kabicidin (1) was added into PY agar medium (PY plus 1.5% agar) and PY soft agar medium (PY plus 1.0% agar) at a concentration of 100 μg per ml.

Determination of bacterial population. Approximately 100 mg of dadih samples were suspended in 10 ml of BM and shaken vigorously for 5 min. Suitable dilutions were made with BM, then 0.1-ml portions of diluted samples were mixed on PY agar plates containing Kabicidin with 5 ml of PY soft agar medium premelted and kept at 50 C. The plates were incuabted in an anaerobic jar containing alkaline pyrogallol at 28 C, and colonies formed were counted. After recording the colony count, 12 colonies were picked up at random and purified on PY agar plates. Taxonomic properties of these isolates were examined and identified according to "Manual for the identification of medical bacteria" written by Cowan and Steel (2) and "Identification of the lactic acid bacteria" by Shape (5).

Results and Discussion

Because a preliminary experiment had showed that fungi in dadih grew rapidly on PY agar plates and interfered with the counting of bacterial colonies, PY agar plates containing Kabicidin were used to determine bacterial counts containing Kabicidin. The results in Table 1 reveal bacterial populations of 6.8 to 8.0 x 10^8 per gram of dadih.

Table 1. Number of bacteria in dadih.

Experiment	Number of bacteria per g of dadih
1	7.7×10^8
2	8.0×10^8
3	6.8×10^8

Taxonomic characteristics of 12 strains isolated from the plates were examined, and it was found that these strains produced lactic acid from glucose homofermentatively. Taxonomic properties of the isolates are summarized in Table 2. Of 12 isolates, 10 strains were identified as Lactobacillus casei subsp. casei, and 2 strains as Lactobacillus plantarum. From these findings we concluded that the major bacterial species in dadih are Lactobacillus casei subsp. casei and Lactobacillus plantarum, which commonly occur in milk and dairy products.

Table 2. Taxonomic characteristics of 12 isolates

Characteristic	Group A	Group B
Gram staining	+	+
Cells	R	R
Motility	-	-
OF test	F	F
Gas from glucose	-	-
gluconate	+	+/-
Growth at 15 C	+	+
45 C	-	-
Lactic acid	L	DL
Esculin hydrolysis	+	+
Acid from arabinose	-	+
cellobiose	+	+
fructose	+	+
galactose	+	+
glucose	+	+
gluconate	+	+
lactose	+	+
maltose	+	+
mannitol	+	+
mannose	+	+
melezitose	+	+
melibiose	-	+

Table 2. (continued)

Characteristic	Group A	Group B
Acid from raffinose	-	+
rhamnose	-	-
ribose	+	+
salicin	+	+
sorbitol	+	+
sucrose	+	+
trehalose	+	+
xylose	-	-

Group A contained 10 isolates which were identified as Lactobacillus casei subsp. casei, and group B contained 2 isolates which were identified as Lactobacillus plantarum.

Abbreviations: R, Rods; F, fermentative; L, L(+)-lactate; and DL, DL-lactate.

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IFO Res. Comm. 13,
17-34, 1987 (March)

FILAMENTOUS FUNGI FROM LITTER SAMPLES COLLECTED IN ISRAEL

TADAYOSHI ITO and TATSUO YOKOYAMA

Summary

Sixty-eight species of filamentous fungi were recorded from litter samples collected in Israel.

Of 68 species identified, 20 belong to the Ascomycotina, 47 to the Deuteromycotina, and 1 to the Zygomycotina. The most prevalent species detected was Aspergillus versicolor followed by Acremonium strictum, Penicillium cyclopium, Eurotium rubrum, Doratomyces purpureo-fuscus, Stachybotrys chartarum, and Chaetomidium fimeti. These species have often been recorded as inhabitants of soils and litter in arid districts of the world, but differ significantly from those commonly found in Japan and South East Asia.

Some noteworthy and interesting species, their substrates and geological distributions are briefly described.

Mycological studies in Israel have been made mainly on the macrofungi of the Basidiomycotina, by Moser et al. (16) and Binyamini (5-8), the Ascomycotina by Avizohar-Hershenzon and Nemlich (2) and Nemlich and Avizohar-Hershenzon (17-19), and soil microfungi, by Baum and Artis (3) and Joffe (10,11).

Although some parasitic fungi have also been studied by Joffe and Palti (12), Rotem et al. (21), and Ben-Ze'ev and Kenneth (4), and have been recorded in CMI Distribution Maps of Plant Diseases, further

investigations and more information are needed to evaluate the flora and distribution of the litter fungi in this area.

This paper contributes to the flora and ecology of the litter fungi in Israel.

Materials and Methods

Seventy litter samples were collected at sites in central and northern Israel shown by asterisks on Fig. 1 by Mr. Fusao Kawakami of Kobe Plant Protection Station, from February to June, 1983. These were put into sterile polyethylene bags to bring back to our laboratory. Most of them were rotten leaves and stems (Table 1).

In the laboratory, each sample was placed in a petri dish (9 cm diam) with a small amount of sterilized water and incubated under moist conditions at room temperature (20-24 C) for about one month. During the incubation period, the development of fungal colonies on the samples was observed three times under a binocular dissecting microscope.

All fungal colonies appearing on the samples were mounted on slide glasses for microscopic observation, and some of them were inoculated both onto the malt extract-yeast extract agar (MYA) plates adjusted to pH 4.5 with lactic acid, and onto MYA plates containing 50 μg/ml of tetracycline hydrochloride. MYA medium contained 10 g of glucose, 5 g of peptone, 3 g of malt extract, 3 g of yeast extract, 20 g of agar in 1000 ml of distilled water.

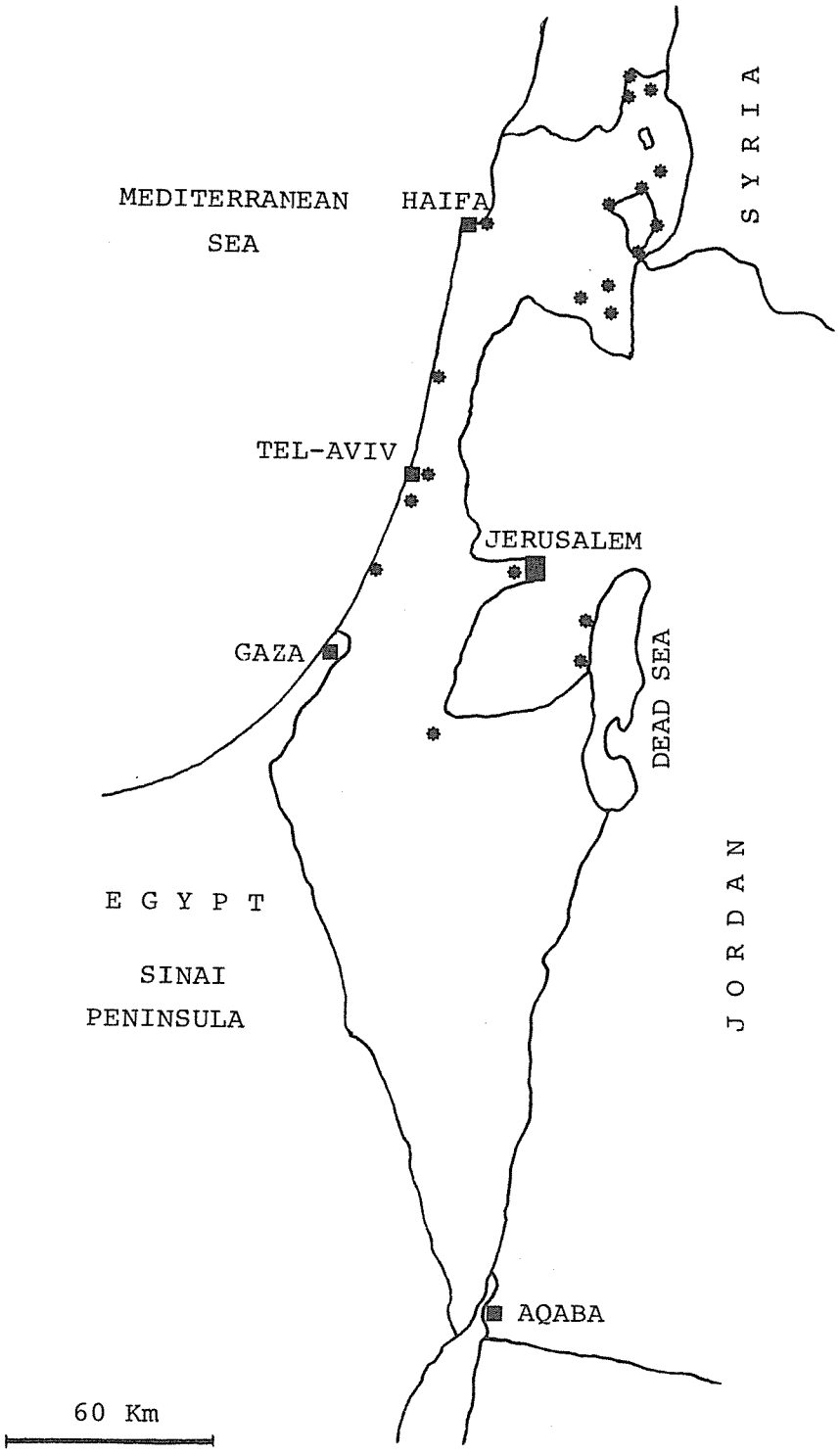
The isolates were incubated at 24 C, then transferred to suitable media for identification.

Results and Discussion

Table 2 lists the fungal species identified and the numbers of the samples from which they isolated. A total of 68 species in 40 genera were identified and classified into 20 species of the Ascomycotina, 47 of the Deuteromycotina, and 1 of the Zygomycotina.

The dominant species were Chaetomidium fimeti, Eurotium rubrum, Petriella setifera in the Ascomycotina, and Acremonium strictum,

Fig. 1. Map of Israel



(*) Asterisks show sample collection sites

Table 1. List of litter samples collected in Israel.

Sample No.	Date sampled	Sample	Locality
1	Apr., 1983	leaves, broad-leaved tree	Jerusalem
2	"	twigs	Sde Elyahu
3	------- '83	decayed leaves	-------
4	May 18, '83	leaves, broad-leaved tree	Caesarea Philippi
5	Feb. 20, '83	Rosa leaves	Ashdod
6	------- '83	Eucalyptus leaves	Capernaum
7	Mar. 12, '83	"	Masada
8	May 18, '83	"	Dabura
9	Feb. 25, '83	Hibiscus leaves	Rehovot
10	Apr., '83	Hibiscus mutabilis leaves	Ashdod
11	May 7, '83	Nerium oleander leaves	Hamat Gader
12	------- '83	twigs	-------
13	May 18, '83	decayed leaves	Caesarea Philippi
14	Mar. 6, '83	leaves, broad-leaved tree	Bet Shean
15	Mar. 12, '83	"	Ein Gedi
16	------- '83	"	-------
17	May , '83	"	Jerusalem
18	Feb., '83	"	Tel Aviv-Jaffa
19	Mar. 9, '83	Aloe leaves	Ashdod
20	Mar. 15, '83	"	"
21	------- '83	Hibiscus leaves	-------
22	------- '83	"	-------
23	Apr., '83	Chrysanthemum leaves	Ashdod
24	Feb., '83	Rosa leaves	"
25	May 3, '83	Hibiscus leaves	Netanya
26	------- '83	leaves, broad-leaved tree	-------
27	Mar., '83	"	Mt. Gilboa
28	Mar. 24, '83	"	Haifa
29	Mar., '83	"	Ashdod
30	Mar. 15, '83	Eriobotrya leaves	"
31	------- '83	Chrysanthemum leaves	-------
32	May 17, '83	Pinus leaves	Metulla
33	Apr. 3, '83	twigs	Bet Shean
34	Apr. 24, '83	Hibiscus leaves	Ashdod
35	Mar. 23, '83	leaves, broad-leaved tree	Beer Sheva
36	Mar. 3, '83	Benzoin umbellatum leaves	Tel Aviv
37	------- '83	twigs	-------
38	May 7, '83	leaves, broad-leaved tree	Hamat Gader
39	Mar., '83	twigs	Tel Aviv
40	Mar. 24, '83	Rosa leaves	"
41	Feb., '83	leaves, broad-leaved tree	"
42	Mar. 19, '83	Eucalyptus leaves	"
43	Mar. 3, '83	Eriobotrya leaves	"
44	Feb., '83	reeds	Ashdod
45	------- '83	decayed leaves	-------
46	May 7, '83	leaves, broad-leaved tree	Ein Gev
47	"	leaves, needle-leaved tree	Hamat Gader
48	"	Citrus limon leaves	Tiberias
49	May 17, '83	leaves, broad-leaved tree	Kiryat Shmona

Table 1. (continued)

Sample No.	Date sampled	Sample	Locality
50	Apr., 1983	leaves, broad-leaved tree	Tel Aviv
51	Mar. 23, '83	Eucalyptus leaves	Beer Sheva
52	------- '83	leaves, broad-leaved tree	-------
53	------- '83	leaves, needle-leaved tree	-------
54	May 2, '83	leaves, broad-leaved tree	Jerusalem
55	Mar., '83	"	Tel Aviv
56	May 12, '83	"	Ashdod
57	Feb. 25, '83	Aloe leaves	Rehovot
58	Mar. 28, '83	leaves, broad-leaved tree	Ein Gev
59	May 18, '83	Pinus leaves	Masada
60	May 17, '83	Rosa leaves	Metulla
61	May 7, '83	leaves, broad-leaved tree	Hamat Gader
62	------- '83	twigs	--------
63	Feb., '83	Aloe leaves	Tel Aviv-Jaffa
64	May 7, '83	Eucalyptus leaves	Hamat Gader
65	Mar., '83	"	Tel Aviv
66	Jun. 16, '83	leaves, broad-leaved tree	Mt. Gilboa
67	------- '83	"	-------
68	May 17, '83	Citrus limon leaves	Kiryat Shmona
69	------- '83	Nerium oleander leaves	-------
70	------- '83	leaves, broad-leaved tree	-------

Aspergillus versicolor, Doratomyces purpureo-fuscus, Penicillium cyclopium, Stachybotrys chartarum in the Deuteromycotina. According to Domsch et al. (9), these species are generally predominant on leaves, plants, crops, or in soil, and have been recorded from temperate to tropical countries. Among these dominant fungi, Eurotium rubrum and Aspergillus versicolor, which have not yet been recorded on litter samples in Japan and South East Asia, are called xerophilic fungi and can grow well at NaCl concentrations up to 30% and sucrose up to 40%. Therefore, these two species are expected to grow well under the hot, dry conditions in Israel.

Alternaria alternata, Arthrobotrys oligospora, Chaetomium globosum, Cladosporium cladosporioides, Clonostachys cylindrospora, Myrothecium leucotrichum, and Zygosporium masonii, which have been reported as dominant and ubiquitous fungi on plant substrates, were infrequently isolated in this investigation. Yokoyama and Tubaki (25), Yokoyama et al. (26), and Matsushima (14,15) showed that these species were the dominant saprophytes on leaves and twigs of Castanopsis cuspidata and

Table 2. List of fungi isolated from litter samples of Israel.

Fungus	Sample No.
ASCOMYCOTINA	
Arachniotus aurantiacus	37
Arachniotus spp.	47,70
Arachnomyces nitidus	2,20
Ascotricha sp.	1
Chaetomidium fimeti	22,31,41,53,56,62
Chaetomium elatum	39
Chaetomium globosum	2,12
Chaetomium murorum	2,25,28,29
Chaetomium olivaceum	52
Chaetomium spp.	17,23,24
Dascyphus sp.	58
Emericella variecolor	11,26
Emericella sp.	42
Eurotium rubrum	4,5,36,42,43,46,51,61
Melanospora zamiae	31
Microthecium sp.	36
Petriella setifera	5,24,30,41
DEUTEROMYCOTINA	
Acremonium strictum	5,6,13,18,20,36,46,55,61,63,64,68
Acremonium sp.	24
Alternaria alternata	49,57,65
Arthrobotrys oligospora	30,32,47,57,65
Aspergillus melleus	14
Aspergillus sclerotiorum ?	46
Aspergillus versicolor	4,5,7,8,9,11,14,18,19,20,25,27,28, 30,32,33,36,40,41,42,44,48,49,55, 58,61,63,64,65,68
Aspergillus sp.	31
Botrytis cinerea	5,60
Chaetomella sp.	12,13,27,35,38,65
Chalara sp.	38
Cladosporium cladosporioides	57,60,65
Cladosporium herbarum	49
Cladosporium sphaerospermum	32,61,63
Cladosporium spp.	11,26
Clonostachys cylindrospora	1,30
Cylindrocladium sp.	35
Dicyma olivacea	18
Doratomyces purpureo-fuscus	3,4,12,14,23,48,53,62
Doratomyces stemonitis	47
Geniculosporium sp.	6
Gliocladium sp.	13
Gonytrichella olivacea	18
Idriella lunata	36
Myrothecium leucotrichum	65
Myrothecium spp.	33,41,67
Nodulisporium sp.	54
Papulaspora sp.	40
Penicillium corylophilum	38

Table 2. (continued)

Fungus	Sample No.
Penicillium cyclopium	6,8,20,42,43,46,48,51,60,61,63,65
Penicillium rubrum	26
Penicillium spp.	29,68
Phaeoisaria clematidis	29,57
Phialophora melinii ?	50,52
Rhinocladiella sp.	15
Scolecobasidium humicola	64
Scopulariopsis brevicaulis	21,43,44
Scopulariopsis sp.	23,35
Stachybotrys chartarum	1,2,18,20,30,36,52,57
Stilbum sp.	40
Verticillium sp.	42
Zygosporium masonii	1
Zygosporium mycophilum	11
ZYGOMYCOTINA	
Rhizopus stolonifer	31

Quercus phillyraeoides or other broad-leaved trees in temperate to tropical zones. These are expected to grow as omnivorous fungi on dry plant materials in Israel.

Beltrania rhombica, *Codinaea simplex*, *Endophragmia uniseptata*, *Monacrosporium ellipsosporum*, *Mycocentrospora gracilis*, *Paecilomyces elegans*, and *Ramularia fusisaprophytica* have been recorded as dominant and typical litter fungi in temperate to tropical zones by Tubaki and Yokoyama (22-24), Yokoyama and Tubaki (25), Yokoyama *et al*. (26), and Matsushima (14,15). These fungi were, however, not found in this investigation. Their absence may be solely due to the dry climate in Israel, since these fungi are considered to be the predominant colonizers of litter in temperate to tropical zones where the temperature and humidity are significantly high. The climate in the southern part of Israel is typically of desert type and the annual rainfall is usually less than 200 mm. In the northern part, however, the climate is semiarid and the annual rainfall is generally up to 500 mm.

The fungal flora on the litter samples in Israel was characterized principally by xerophilic fungi and also by omnivorous fungi. This is in striking contrast to the fungal flora in Japan and South East Asian countries, where the typical litter-decomposing fungi are predominant.

Some noteworthy and interesting species found in this investigation, their substrates and geological distributions are briefly described.

Arachniotus aurantiacus (Kamyschko) von Arx was found on leaves and twigs of an unidentified plant. According to Orr et al. (20), this species has been recorded in soil of semiarid type in USSR and sheep dung in USA. (Figs. 2-4)

Arachnomyces nitidus Massee & Salmon was found on leaves and twigs of an unidentified tree and on Aloe leaves. Malloch and Cain (13) reported that this species was found on hay-dung compost in Canada and dead grass in England. Although a pure culture of this species from a natural substrate has not yet been obtained, we succeeded in obtaining a pure culture from leaf and twig substrates of an unidentified plant. (Figs. 5-9)

Chaetomidium fimeti (Fuckel) Saccardo was found on decayed leaves of Chrysanthemum, leaves of a broad-leaved tree, and leaves and stems of a needle-leaved tree. This species has been recorded on dung, decaying plant materials, seeds, and soil in Europe, North America, and Japan. (Figs. 10-13)

Chaetomium elatum Kunze ex Fries was isolated from stems of an unidentified tree. Although this species is known as a heterothallic species, this isolate was recognized as homothallic by the teleomorphic state formed in a single ascospore colony. This species is known from soil, dung, dead plant materials, nests and feathers of various animals in Yugoslavia, Israel, USA, Kuwait, Bulgaria, Germany, England etc., and is called "a worldwide species". (Figs. 14-17)

Emericella variecolor Berkeley & Broome was found on leaves of Nerium oleander and of an unidentified broad-leaved tree. This species is also recorded from soil in Panama, USA and India, and leaves in India, Nepal, and Pakistan. This fungus is considered to be a xerophilic, since it is recorded from date fruits and spices, and its distribution is mainly restricted to arid districts, particularly those of tropical countries. (Figs. 18-21)

Emericella sp. was isolated from leaves of an unidentified broad-leaved tree. Although the ascospores of this isolate closely resemble those of E. variecolor in shape, they differ greatly in their surface ornamentation. The surface structure of the ascospores of this isolate is rough-walled, and the anamorph is hardly found on ordinary media. The isolate is a new species and will be described elsewhere. (Figs. 22 & 23)

Melanospora zamiae Corda was isolated from decayed leaves of Chrysanthemum sp.. This species has been found on dead plant materials, seeds, fiber, banana, and some polypores in England, Burma, Uganda, Japan, Brazil, and New Guinea. (Figs. 24-27)

Aspergillus melleus Yukawa was found on leaves of an unidentified broad-leaved tree. This species is reported by Domsch et al. (9) as a fungus of worldwide distribution. It is especially prevalent in rhizosphere soils in tropical and subtropical regions, and on seeds of groundnut, soy beans, rice, corn, and dried salted food in Israel, Ghana, Egypt, Kenya, Sudan, India, Pakistan, Burma, Peru, and Argentina. (Figs. 28-30)

Dicyma olivacea (Emoto & Tubaki) von Arx was isolated from leaves of an unidentified broad-leaved tree. This species has been recorded on paper, polluted urethanform, soil, and as an air contaminant in France, Japan, and Italy. Gonytrichella olivacea Emoto & Tubaki and Puciola spinosa De Bertoldi have been accommodated in this species by von Arx (1), who also been treated them as the anamorph of Ascotricha erinacea Zambettakis. Three strains maintained in IFO as G. olivacea (IFO 9178, IFO 9841, IFO 31999) and one strain (CBS 226.76) derived from the type of P. spinosa were mixed with each other on agar medium, but no teleomorph of the fungus developed . (Figs. 31 & 32)

The authors are much indebted to Mr. Fusao Kawakami of Yokohama Plant Protection Station, Kanagawa (formerly of Kobe Plant Protection Station, Hyogo), for kindly supplying the litter samples which he collected during his stay in Israel.

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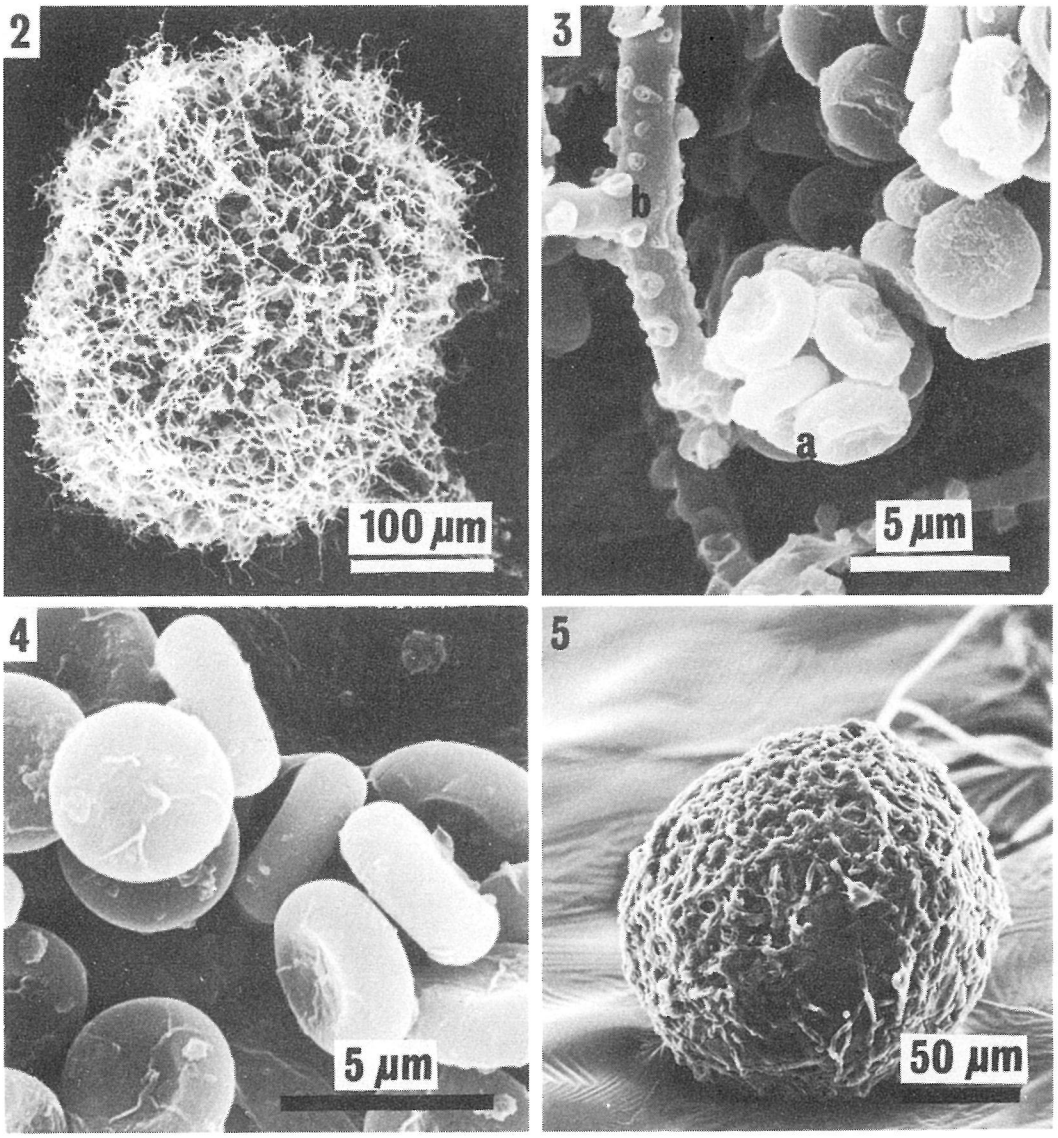
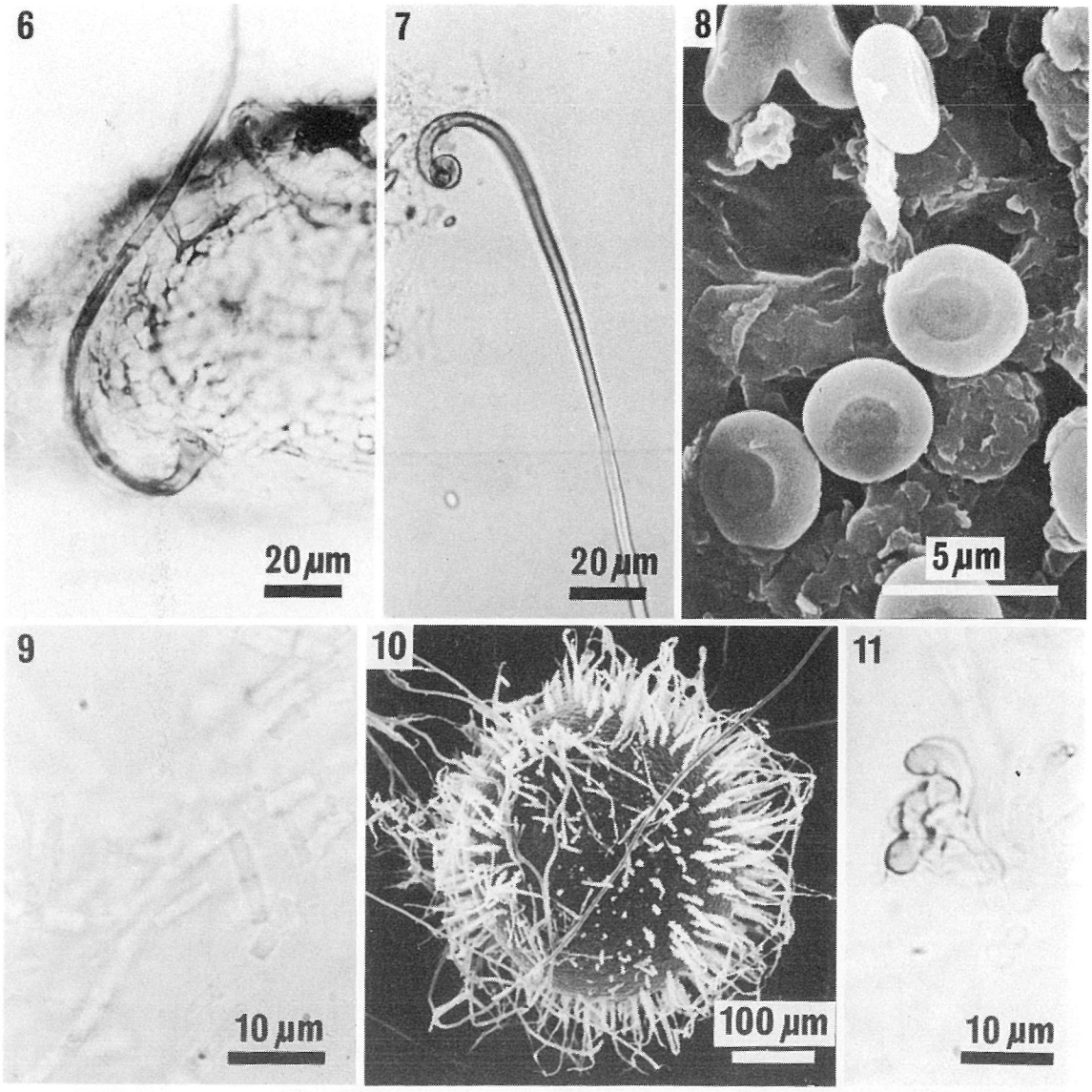
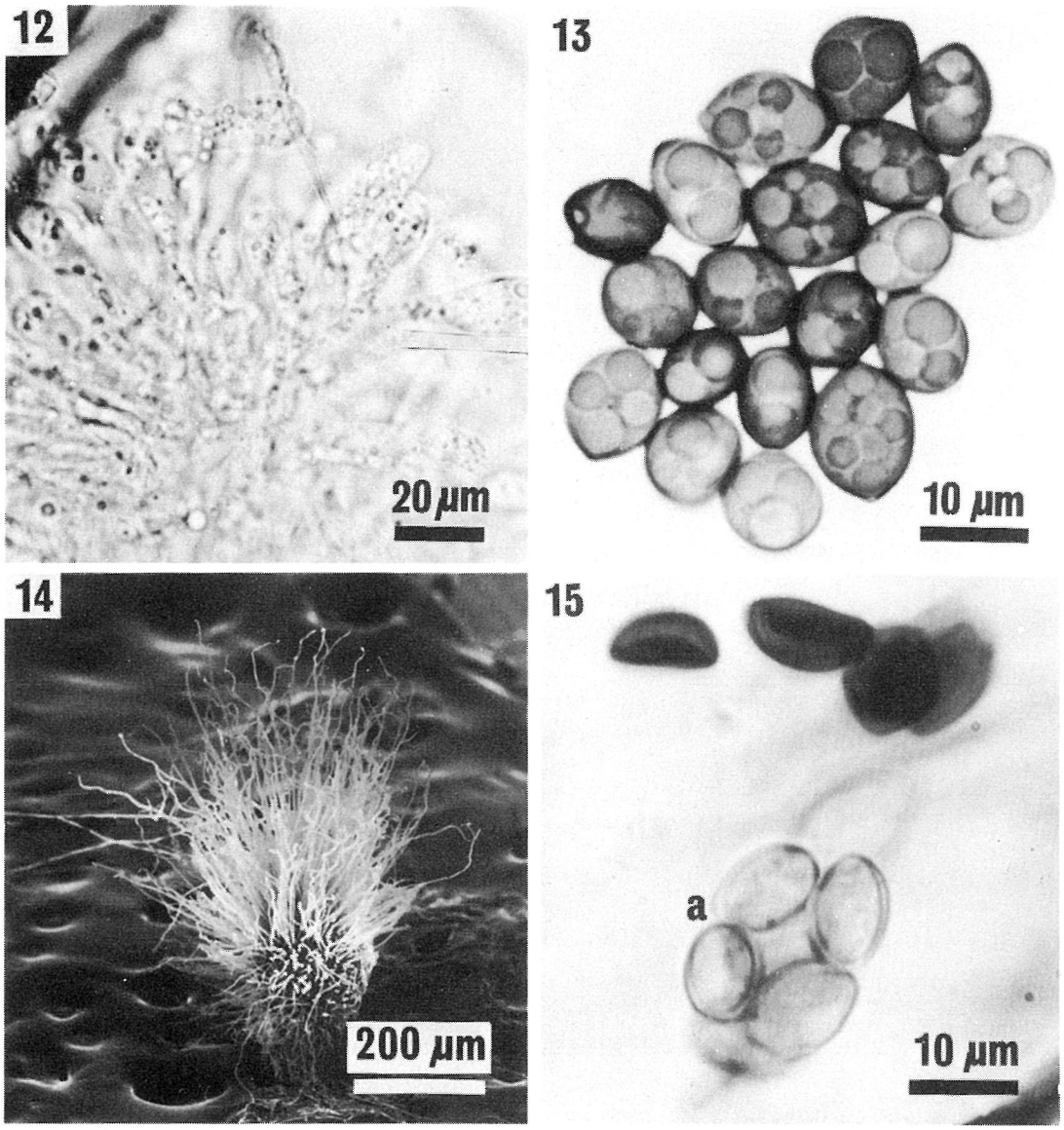


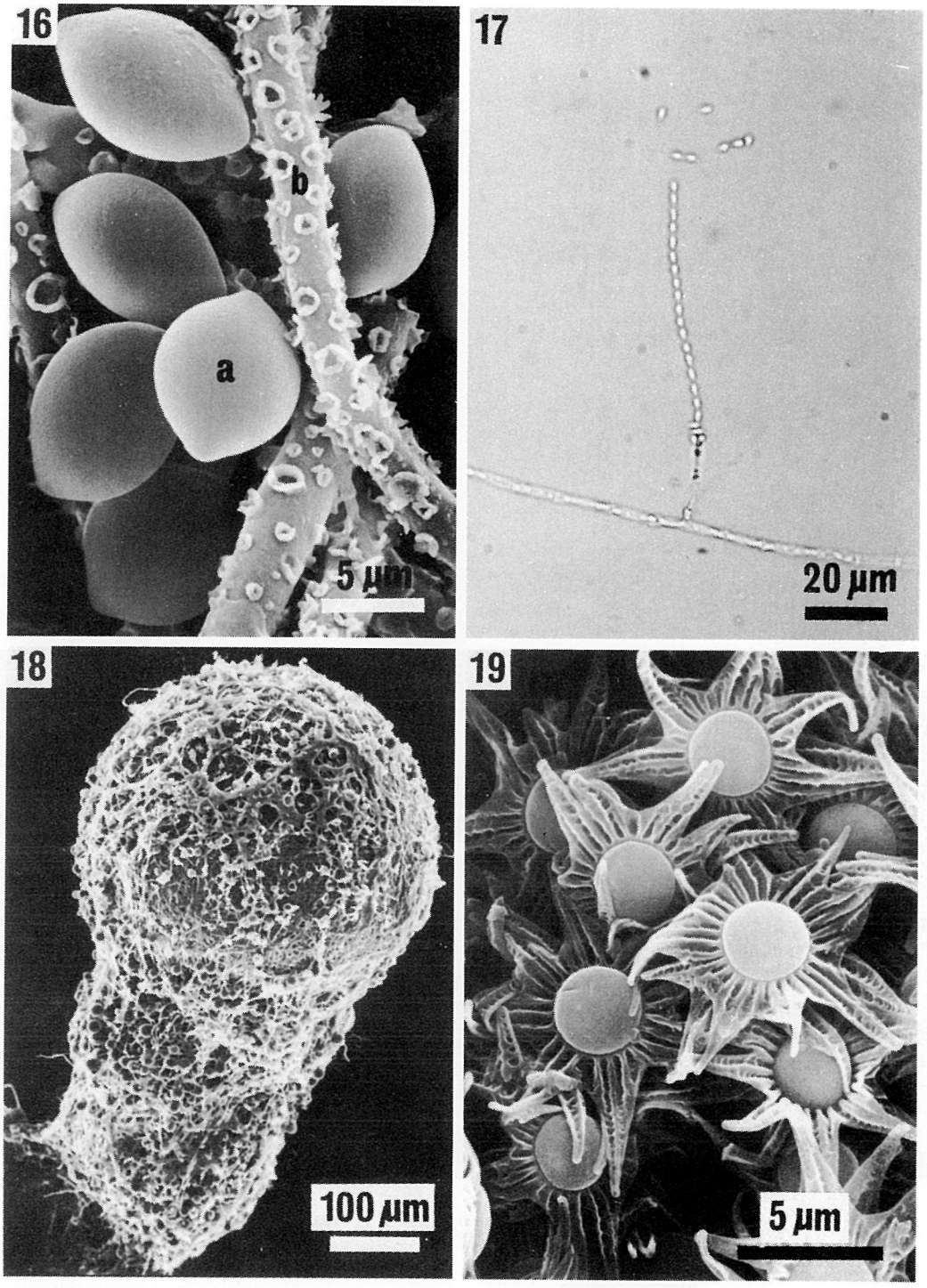
Figs. 2-4. Arachniotus aurantiacus. 2. Ascomata. 3. Ascus (a) and peridial hyphae (b). 4. Ascospores.
Fig. 5. Arachnomyces nitidus. Ascomata.



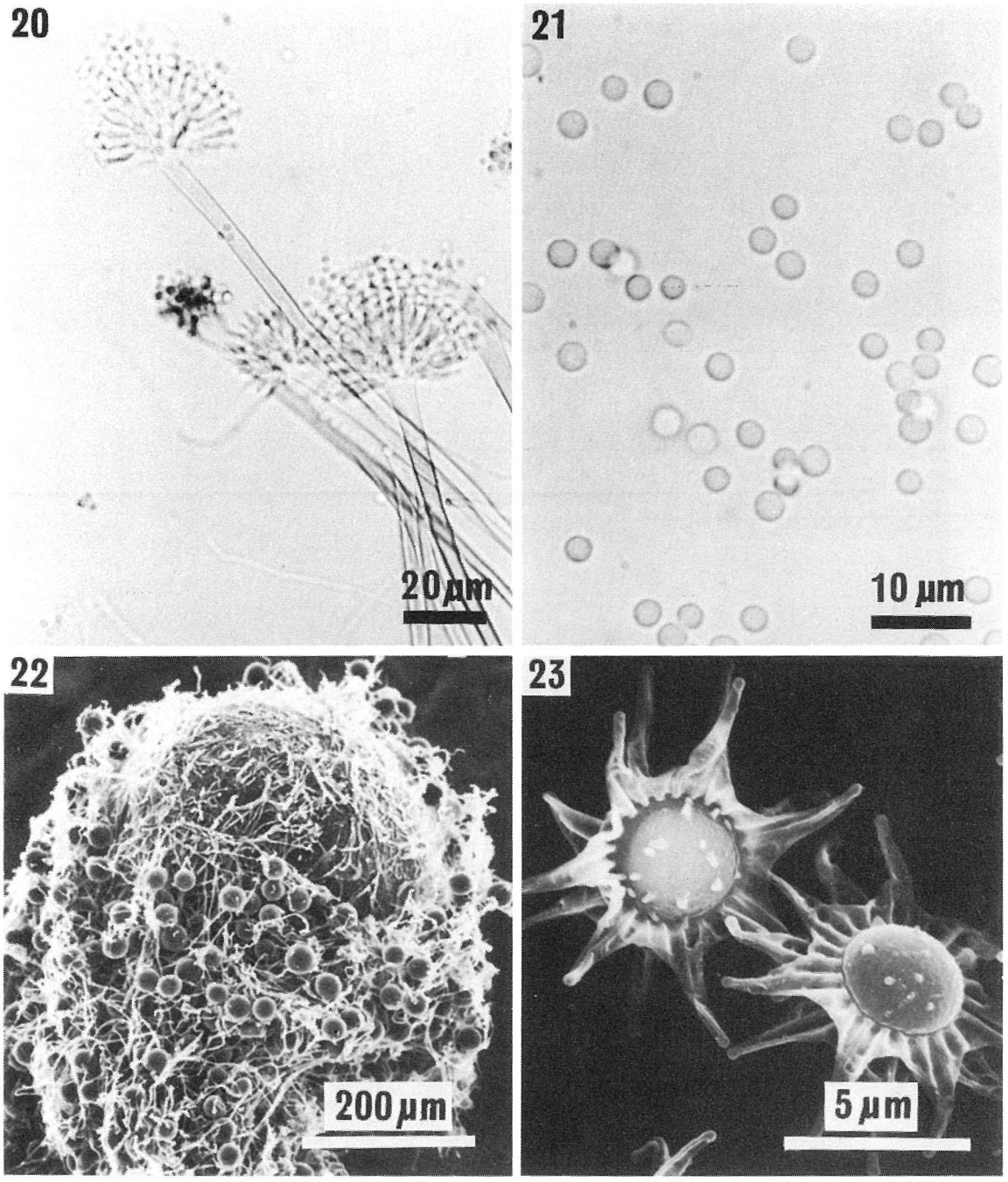
Figs. 6-9. Arachnomyces nitidus. 6 Ascomatal wall and appendage. 7. Coiled tip of appendage. 8. Ascospores. 9. Anamorph.
Figs. 10 & 11. Chaetomidium fimeti. 10. Ascomata. 11. Ascomatal initial.



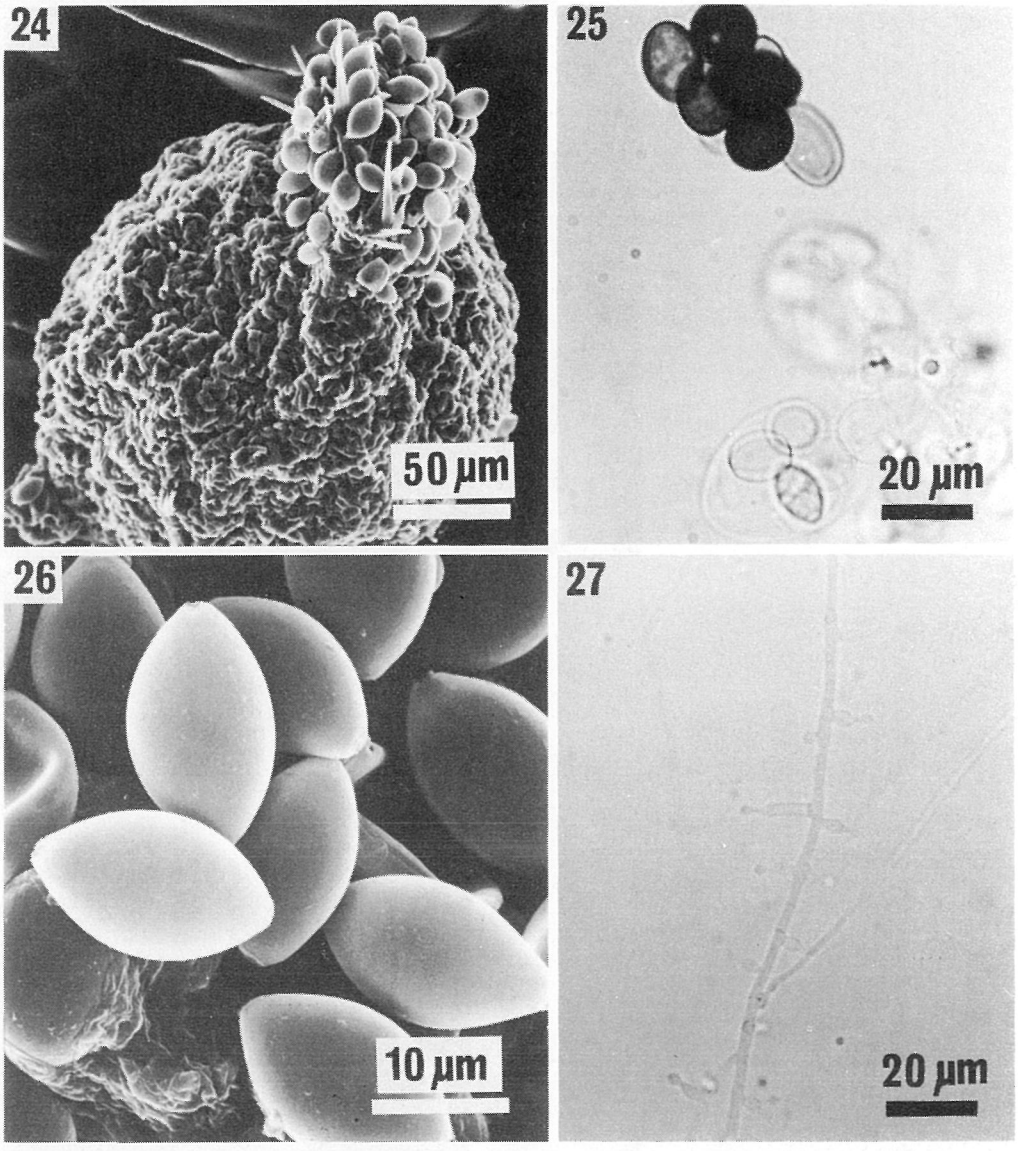
Figs. 12 & 13. Chaetomidium fimeti. 12. Immature asci. 13. Ascospores.
Figs. 14 & 15. Chaetomium elatum. 14. Ascomata. 15. Ascus (a).



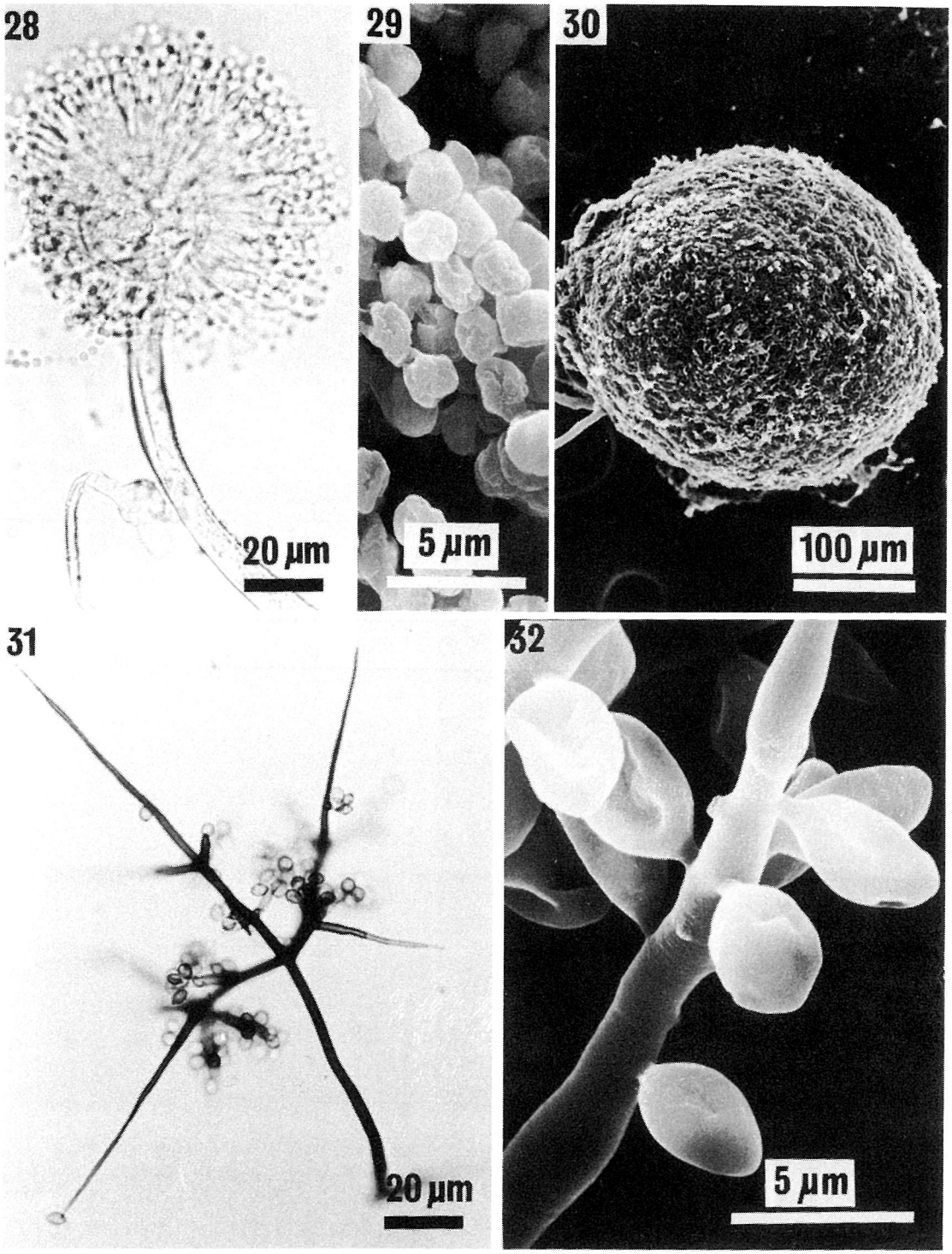
Figs. 16 & 17. Chaetomium elatum. 16. Ascospores (a) and hairs of perithecia (b). 17. Acremonium anamorph.
Figs. 18 & 19. Emericella variecolor. 18. Ascomata. 19. Ascospores.



Figs. 20 & 21. Emericella variecolor. 20. Conidial structures of Aspergillus anamorph. 21. Phialoconidia.
Figs. 22 & 23. Emericella sp. 22. Ascomata. 23. Ascospores.



Figs. 24-27. Melanospora zamiae. 24. Ascomata with ascospores on the beak. 25. Asci and ascospores. 26. Ascospores. 27. Phialophora anamorph.



Figs. 28-30. Aspergillus melleus. 28. Conidial head. 29. Phialoconidia. 30. Sclerotium.
Figs. 31 & 32. Dicyma olivacea. 31. Conidial structures. 32. Conidiogenous cell and blastconidia.

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MORPHOLOGICAL OBSERVATION OF ACTINOMYCETES STRAINS BY SCANNING ELECTRON MICROSCOPY

TAIKI KUSAKA and ISAMU ASANO

Summary

All L-dried ampoules of Actinomycetes in the IFO Culture Collection were checked morphologically by scanning electron microscopy. When revived on IFO 227 medium, Streptomyces hyalinus IFO 13850 showed straight mycelial ends and smooth spore surfaces, but when revived on IFO 228 medium, it showed the same spiral mycelial ends and spiny spore surfaces as in the original description. The smooth spore surface of Streptomyces flaveolus IFO 3715 was different from the hairy spore surface of the reference strain of this species, IFO 12768 (ISP 5061). A few hairy spores were observed when the strain was grown on IFO 227 medium but not on four other media. All cultures of Streptomyces candidus subsp. enterostaticus IFO 13821 showed compact spiral ends of mycelia and rugose spore surfaces, but this species was described originally to be straight and smooth. Streptomyces thermophilus IFO 12381 showed spiral mycelial ends and smooth spore surfaces, but the type strain showed straight mycelia and spiny spore surfaces. IFO 13821 and IFO 12381 were both deleted from the Collection.

Actinomycetes strains in the IFO Culture Collection are preserved and distributed under L-dried conditions. Once L-dried, they are considered to be preservable over a long period without serious problems of mortality or morphological, cultural and physiological variation (5, 8, 15). It is necessary to renew the L-dried specimens before they are distributed to users, and renewed specimens should be checked to see whether they retain the features of the original strains. One method of checking, morphological observation by scanning electron microscopy has been applied to revived strains which had been checked for survival and cultural features. This report deals with the observations and results.

Materials and Methods

Actinomycetes strains. Revived cultures from dry specimens or agar-transplanted cultures preserved in the IFO Culture Collection were used.
Streptomyces hyalinus IFO 13850: three strains, lot 84 08 22, lot 78 05 16 and lot MB891-A1 (prepared by depositor).
Streptomyces flaveolus IFO 3715: three strains, lot 84 10 02, lot 85 01 17 and an agar-transplanted culture.
Streptomyces candidus subsp. enterostaticus IFO 13821: four strains, lot 84 08 24 no. 10, lot 84 08 24 no. 8, lot 78 05 17 and lot 78 03 14 (prepared by depositor).
Streptomyces thermophilus IFO 12381: three strains, lot 84 09 04 no. 10, lot 84 09 04 no. 9 and an agar-transplanted culture.
Streptomyces thermophilus IFO 13370 (=ISP 5365): one strain, an agar-transplanted culture.

Media. The media used for cultivation were as follows (4, 10).

IFO 228 medium (Bennett's agar): yeast extract 1 g, beef extract 1 g, N.Z. Amine, type A (Sheffield Chem. Co.) 2 g, glucose 10 g, distilled water 1 liter, agar 20 g, pH 7.3.

IFO 231 medium: Same as IFO 228 medium, but with maltose instead of glucose as carbon source.

IFO 227 (ISP 2) medium: yeast extract 4 g, glucose 4 g, malt extract 10g, distilled water 1 liter, agar 20 g, pH 7.3.

ISP 3 medium: oat meal 20 g, agar 20 g, salts solution 1 ml, distilled water 1 liter, pH 7.2.

(Salts solution: $FeSO_4 \cdot 7H_2O$ 0.1%, $MnCl_4 \cdot 4H_2O$ 0.1%, $ZnSO_4 \cdot 7H_2O$ 0.1%)

ISP 4 medium: soluble starch 10 g, K_2HPO_4 1 g, $MgSO_4 \cdot 7H_2O$ 1 g, NaCl 1 g, $(NH_4)_2SO_4$ 2 g, $CaCO_3$ 2 g, salts solution 1 ml, agar 20 g, distilled water 1 liter, pH 7.2.

ISP 5 medium: L-asparagine 1 g, glycerol 10 g, K_2HPO_4 1 g, salts solution 1 ml, agar 20 g, distilled water 1 liter, pH 7.2.

IFO 1 medium (potato sucrose agar): potato 200 g (decoction), sucrose 20 g, distilled water 1 liter, agar 20 g, pH 5.6.

Revival of dry specimens. After sterilizing an ampoule with 75% alcohol-damped gauze, the ampoule was filed at the mid-point of the cotton wool plug and a red hot glass rod was applied to the file cut. The pointed top of the ampoule was removed and an appropriate liquid medium (0.5 ml) was added aseptically into the bottom part. The resulting suspension was transferred to an appropriate agar medium and incubated for 2 or 3 weeks at the optimum temperature for the growth of organism.

Observation by scanning electron microscopy. The morphologies of the spores and mycelia of the strains grown on the agar media were examined under a scanning electron microscope (Model JSM T-20, JEOL Ltd.). The specimens were prepared by either by simple drying or by critical drying. In the former method, an agar block bearing abundant mycelia and spores was kept at 37 C overnight to dehydrate. In the latter method, an agar block was first fixed in the vapour of osmic acid solution (2%). It was then gradually dehydrated with increasing concentrations of ethyl alcohol (3% to 100%), then placed in a solution of isoamylacetate for more than one hour. Finally it was treated in a critical point drying apparatus (Hitachi HCP-2). Dried specimens were coated with evaporated gold and examined under the scanning electron microscopy.

Results and Discussion

Streptomyces hyalinus IFO 13850

When the dried specimens of lot 84 08 22 and lot MB891-A1 were revived on IFO 227 medium, aerial mycelia showed straight or wavy ends and spore surfaces were smooth (Figs. 1 and 2). However, spiral ends of mycelia and spiny spores were observed in the strains of lot 78 05 16 and

another specimen of lot 84 08 22 that were revived on IFO 228 medium (Figs. 3 and 4). Thus the characteristic morphology of IFO 13850 varied with the medium used for revival. This species is reported to form aerial mycelia with open spiral ends and conidial spores with hairy surfaces on starch agar and calcium malate agar (7). This hairy structure, however, was said to be so vague as to be difficult to discern under a conventional electron microscope, and no pictures of the mycelial end or spore surface were presented in the literature (7). Generally, an original description lists the most characteristic morphological features of a strain on various media as its morphological properties. It is not uncommon for many spira to be observed on some media but few on others, and for compact spira or Retinaculum-Apertum to be formed on some media but loose spira or Rectus-Flexibilis (RF) on others. Thus our finding of wavy (RF) ends of mycelia instead of open spirals and smooth or spiny spore surfaces instead of hairy structures may be attributable to differences in media from those in the reference. Our dry specimens of IFO 13850 were concluded to be prepared appropriately, because they showed similar morphological features to those of original description on IFO 228 medium. The observations on IFO 227 medium were considered to be within the range of variation of cultivations on agar media.

Streptomyces flaveolus IFO 3715

Dry specimens of lot 84 10 02 and lot 85 01 17 revived on IFO 228 medium, and five subcultures of an agar-transplanted strain on IFO 227 (=ISP 2), ISP 3, ISP 4, ISP 5 and IFO 1 medium, formed aerial mycelia with spiral ends and smooth-surfaced spores (Figs. 5-11). Hairy spores were observed only in the subculture on IFO 227 medium (Fig. 5). A reference strain, IFO 12768 (ISP 5061), showed spiral-ended mycelia and hairy-surfaced spores (Figs. 12-16), although it also formed smooth-surfaced spores in ISP 5 medium (Fig. 15). According to the original (14) and the ISP (11) descriptions, which pertain to two strains derived from the same origin (4), this species has spiral mycelial ends and hairy spore surfaces. The morphological features of reference strain IFO 12768 observed by us were similar to those described, while those of IFO 3715 differed, especially in the formation of hairy structures on the spore surface. However, it is also true that IFO 3715 is still able to produce hairy spores and that IFO 12768 produces a few smooth spores under the same conditions. There was also a difference in the color tinges of the

two strains on five different media. Nevertheless, IFO 3715 is concluded to be corespond substantially to the description of the species, though it should not be used as a reference strain because of its low production of hairy spores.

Streptomyces candidus subsp. enterostaticus IFO 13821

All four strains revived on IFO 228 medium from dry specimens, namely, lot 84 08 24 (2 ampoules), lot 78 05 17 and lot 78 03 14 (prepared by depositor), formed spiral-ended aerial mycelia and rugose-surfaced spores (Figs. 17-19). According to the original description, this strain grown on Czapeck's agar medium formed straight aerial mycelia, tufts on hyphae and smooth-surfaced spores (6). Our observations were repeatedly obtainable in subcultures on various media and they are suggested to be typical morphological characters of Streptomyces hygroscopicus (1). Aerial mycelia on agar media changed color from white to black and became hygroscopic befor their vanishing. The description, however, states that aerial mycelia remain white even on prolonged cultivation (6). This discrepancy was attributed to a strain completely different from the original descripion having been accidently deposited under the accession number IFO 13821 and preserved until now. IFO 13821 was concluded to be not the strain indicated by its epithet.

Streptomyces thermophilus IFO 12381

A dry specimen of lot 84 09 04 no. 10 revived on IFO 228 medium had spiral mycelial ends and smooth-surfaced spores (Fig. 21), while another strain from lot 84 09 04 no. 8 had apparently straight mycelial ends with smooth-surfaced spores (Fig. 22). This species was reported first as Actinomyces thermophilus Gilbert (2) and was later renamed as Streptomyces thermophilus (Gilbert) Waksman and Henrici (16). Strain A_{11} was proposed as a neotype strain with straight mycelial ends and spiny spores (3). Strain A_{11} was later adopted in the International Streptomyces Project as ISP 5365 (12) and has been preserved in the IFO Collection under the accession number IFO 13370 (4). Strain IFO 12381 was deposited in the IFO Collection in 1966 through the KCC Collection (KCC S-0238) from Dr. C.L. Fergus, who had identified it. Since Fergus's original description of IFO 12381 has not yet been found (9), IFO 12381 was compared with type strain IFO 13370 of this species. IFO 13370 had morphological characters of straight, not spiral mycelia and spiny spores, just as in the ISP description (Fig. 24). However, IFO 12381 had spiral

mycelia and smooth spores (Figs. 20-23). IFO 12381 was concluded not to be the same strain as the strain description.

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Explanation of figures.

Morphological observation of Streptomyces hyalinus IFO 13850.
Fig. 1. Strain revived on IFO 227 medium from lyophilized ampoule lot MB891-AL (prepared by depositor).
Fig. 2. Strain revived on IFO 227 medium from L-dried ampoule lot 84 08 22 no.10.
Fig. 3. Strain revived on IFO 228 medium from L-dried ampoule lot 84 08 22 no. 8.
Fig. 4. Strain revived on IFO 228 medium from L-dried ampoule lot 78 05 16.

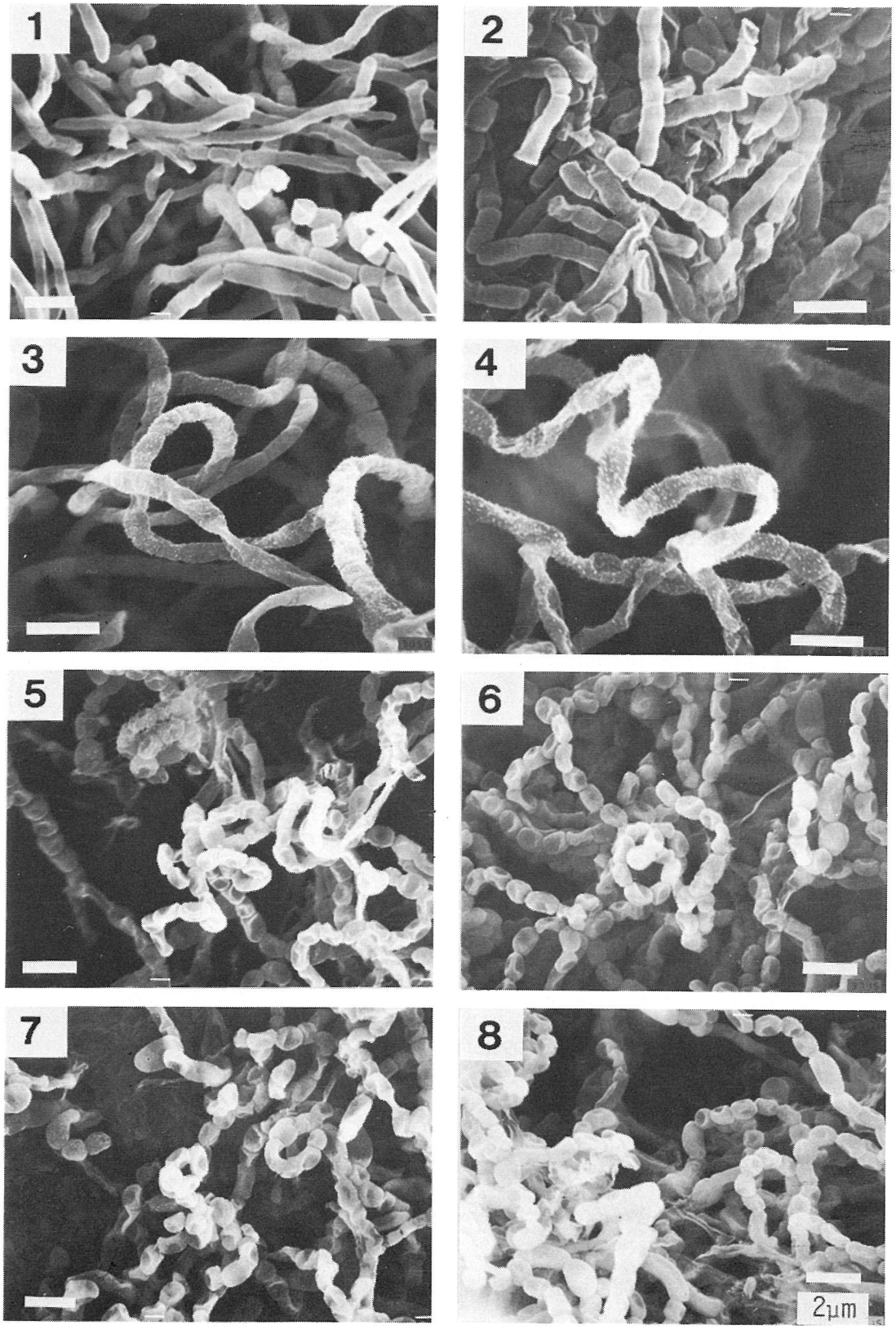
Morphological observation of Streptomyces flaveolus IFO 3715.
Fig. 5. Agar-transplanted strain grown on ISP 2 medium.
Fig. 6. Agar-transplanted strain grown on ISP 3 medium.
Fig. 7. Agar-transplanted strain grown on ISP 4 medium.
Fig. 8. Agar-transplanted strain grown on ISP 5 medium.
Fig. 9. Agar-transplanted strain grown on potato sucrose agar.
Fig.10. Strain revived on IFO 228 medium from lyophilized ampoule lot 84 10 02.
Fig.11. Strain revived on IFO 228 medium from lyophilized ampoule lot 85 01 17.

Morphological observation of Streptomyces flaveolus IFO 12768.
Fig.12. Agar-transplanted strain grown on ISP 2 medium.
Fig.13. Agar-transplanted strain grown on ISP 3 medium.
Fig.14. Agar-transplanted strain grown on ISP 4 medium.
Fig.15. Agar-transplanted strain grown on ISP 5 medium.
Fig.16. Agar-transplanted strain grown on potato sucrose agar.

Morphological observation of Streptomyces candidus subsp. enterostaticus IFO 13821.
Fig.17. Strain revived on IFO 228 medium from lyophilized ampoule lot 78 03 14 (prepared by depositor).
Fig.18. Strain revived on IFO 228 medium from L-dried ampoule lot 84 08 24.
Fig.19. Strain revived on IFO 228 medium from L-dried ampoule lot 78 05 17.

Morphological observation of Streptomyces thermophilus IFO 12381.
Fig.20. Agar-transplanted strain grown on IFO 228 medium.
Fig.21. Strain revived on IFO 228 medium from lyophilized ampoule lot 84 09 04 no 10.
Fig.22. Strain revived on IFO 228 medium from lyophilized ampoule lot 84 09 04 no 8.
Fig.23. Agar-transplanted strain grown on IFO 228 medium.

Morphological observation of Streptomyces thermophilus IFO 13370
Fig.24. Agar-transplanted strain grown on potato sucrose agar.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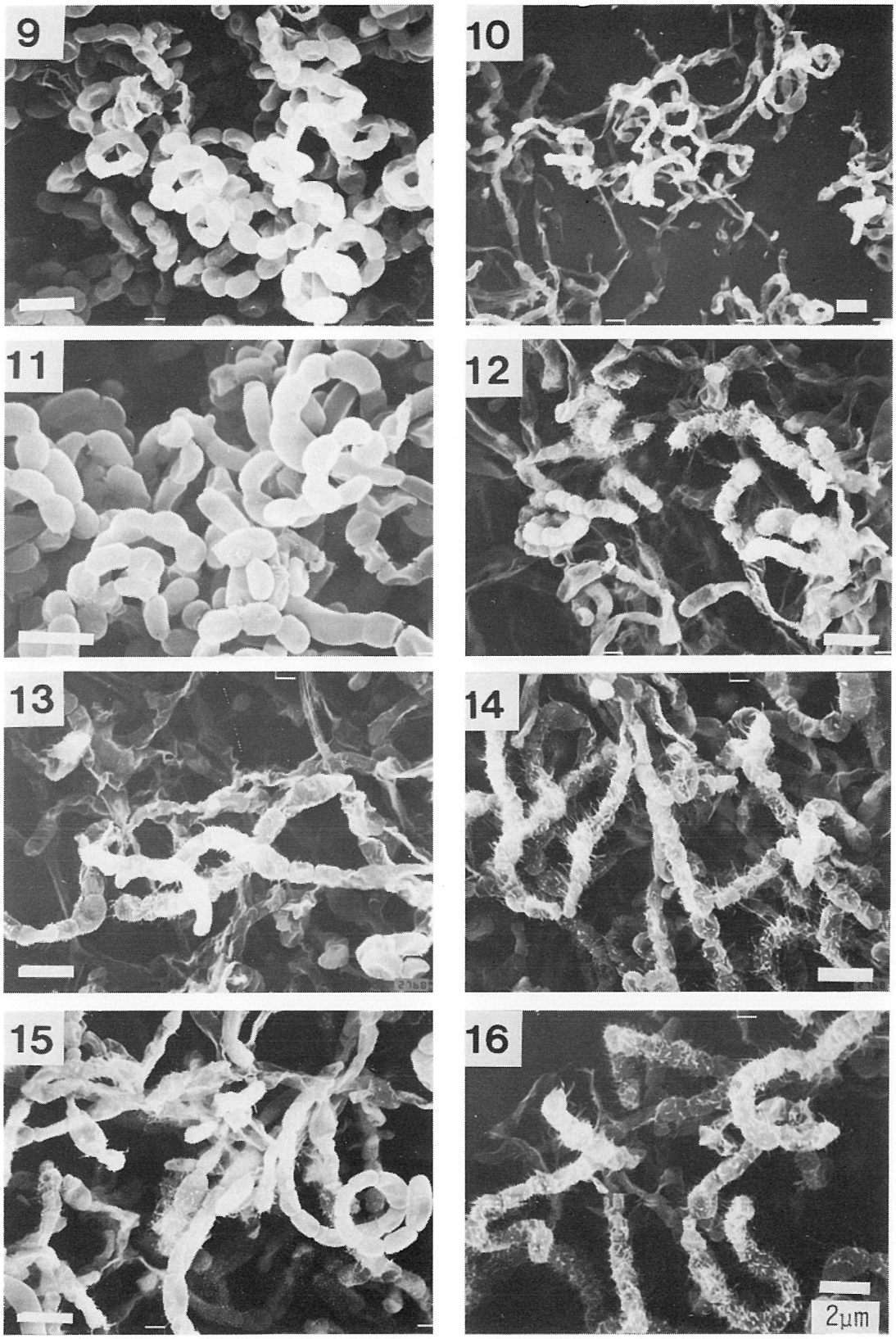
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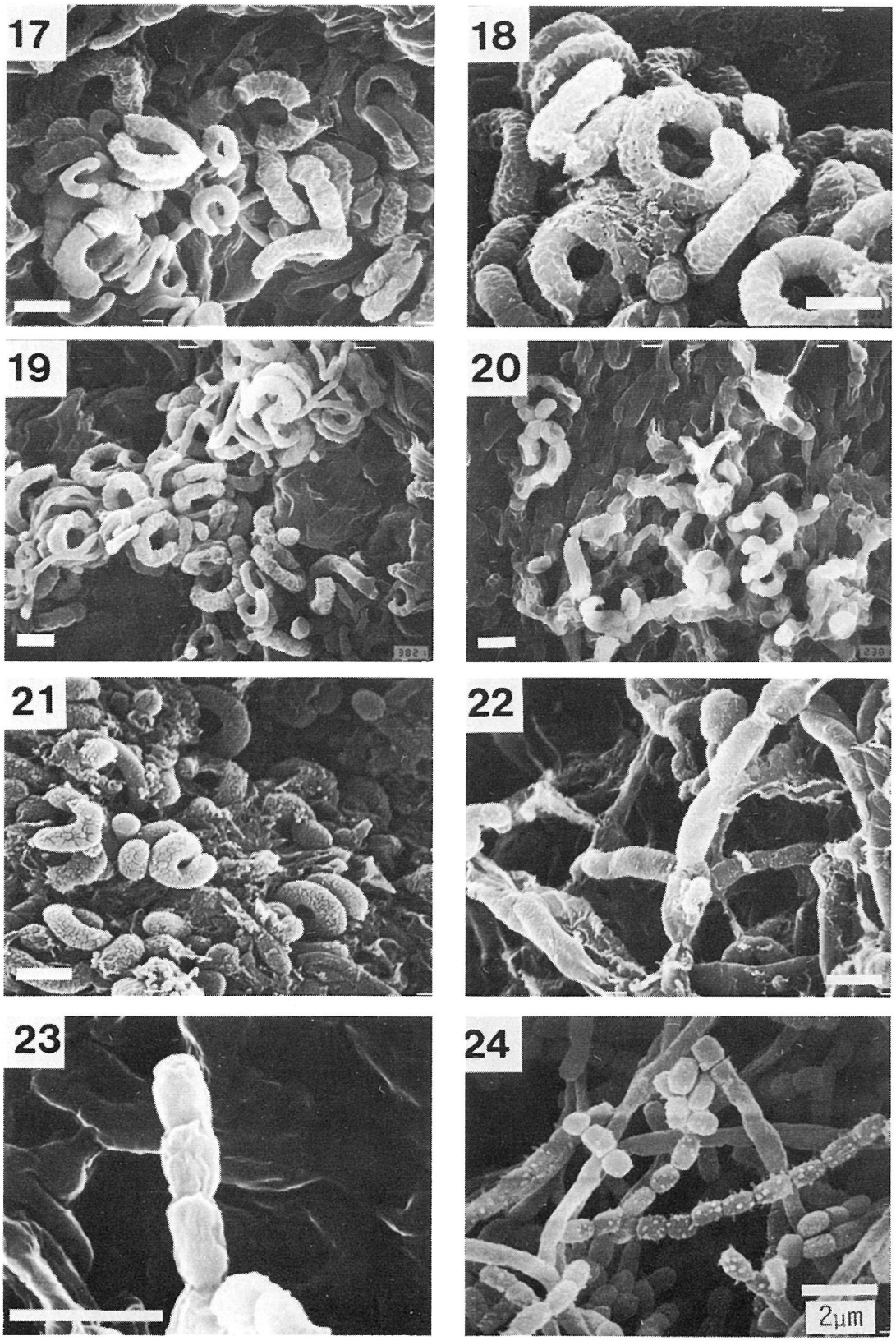
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45-51, 1987 (March)

SURFACE STRUCTURE OF ASCOSPORES OF THE GENUS *SCHIZOSACCHAROMYCES*

KOZABURO MIKATA and ISAO BANNO

Summary

The surface structure of the ascospores of the genus *Schizosaccharomyces* was examined by scanning electron microscopy. Ascospores of *S. pombe*, and *S. malidevorans* showed a warty surface, those of *S. octosporus* a smooth surface with occasional papillae, those of *S. japonicus* a smooth surface.

In Schizosaccharomycetaceae (fission yeasts), only the genus *Schizosaccharomyces* and the following 4 species have been recognized: *S. japonicus* Yukawa et Maki var. *japonicus*, *S. japonicus* Yukawa et Maki var. *versatilis* (Wicherham et Duprat) Slooff, *S. malidevorans* Rankine et Fornachon, *S. octosporus* Beijerinck, and *S. pombe* Lindner (type species). Monographs on yeast taxonomy (2,6) state that the morphology of ascospores produced by the 4 species of the genus are globose to ellipsoidal under a light microscope, and in maturation the spores of *S. japonicus* var. *versatilis* only become reniform to allantoid. Occasionally, ascospores have been found to be slightly roughend.

Osumi (4) examined ascospores of *S. pombe* using a scanning electron microscope (SEM) and presented a SEM photograph showing irregular warty protuberences on the spore-surface, but made no comment from taxonomic point of view. This note examines the fine structure of the ascospore-surface of all the 4 species and one variety of the genus.

Materials and Methods

Strains. Yeast cultures used in this work and the conditions for sporulation are summarized in Table 1.

Medium. ME medium (malt extract 10% and agar 1.5%) and YM medium (yeast extract 0.3%, malt extract 0.3%, peptone 0.5%, glucose 1.0% and agar 1.5%) were used for sporulation.

Table 1. List of Schizosaccharomyces strains.

Organism	Strain	Original name	Sporulation[a]
S. pombe	IFO 0340	S. asporus	no spore
	IFO 0342 (CBS 1043)	S. formosensis	++ YM[b]
	IFO 0343 (CBS 1044)	S. formosensis var. akoensis	++ YM
	IFO 0344 (CBS 1062)	S. formosensis var. tapaniensis	++ YM
	IFO 0345 (CBS 355)	S. pinan	++ YM
	IFO 0346^T(CBS 356)		+ YM
	IFO 0347 (CBS 2775)	S. pombe var. ogasawaraensis	++ YM
	IFO 0349 (CBS 2776)	S. pombe var. itoensis	++ YM
	IFO 0351		+w YM
	IFO 0354 (CBS 357)	S. mellacei	no spore
	IFO 0358 (CBS 1042)	S. liquefaciens	no spore
	IFO 0362		no spore
	IFO 0363		++ YM
	IFO 0364 (CBS 1063)	S. santawensis	+w YM
	IFO 0365 (CBS 1061)	S. taito	no spore
	IFO 0366 (CBS 352)	S. vordermani	+ YM
	IFO 0638	S. liquefaciens	++ YM
	IFO 1628^T(CBS 356)		++ YM
S. malidevorans	IFO 1608^T(CBS 5557)		++ YM
S. octosporus	IFO 0353		++ YM
	IFO 0360		+ YM
	IFO 0361		++ YM
S. japonicus var. versatilis	IFO 1607^T(CBS 103)		+ ME
S. japonicus var. japonicus	IFO 1609^T(CBS 354)		++ ME
	IFO 1646		no spore
	IFO 1712		+w ME
	IFO 1713		+w ME

a: sporulation medium (YM, YM agar; ME, Malt extract agar).
b: ++, +, +w indicate respectively abundant, moderate and slight production of ascospores.
T: type strain.

Preparation of ascospores. After confirmation of sufficient production of ascospores under a light microscope, a mass of sporogenous cells was suspended in 0.1 M phosphate buffer. Liberation of spores from asci, pre-fixation, dehydration, critical point drying, and Au-coating were carried out by the methods stated in previous report (1), except that

isoamylacetate was substituted for acetone before critical point drying.

Results and Discussion

Four strains of S. pombe, IFO 0354, IFO 0358, IFO 0362, IFO 0365, and one strain of S. japonicus var. japonicus, IFO 1646, failed to sporulate and were considered probably to be of haploid mating type. Spores of all other strains were examined by scanning electron microscopy.

S. pombe. (Figs. 1 - 11)

The spores were spherical to short ellipsoidal. The surface was decolated with ornamentation of irregular warts or short ridges.

S. malidevorans. (Fig. 12)

Spherical ascospores were found to be covered with large irregular warts all over the surface. The fine surface-structures of the ascospores of S. malidevorans and S. pombe are not clearly differentiable from each other or from those produced by Debaryomyces hansenii (1).

S. octosporus. (Figs. 13 - 15)

The spores were spherical to ellipsoidal. Notably, occasional papillae of irregular size were found on the fairly smooth surface. These papillae were observed in all the 3 strains tested and are considered characteristic of the species.

S. japonicus var. japonicus. (Figs. 17, 18).

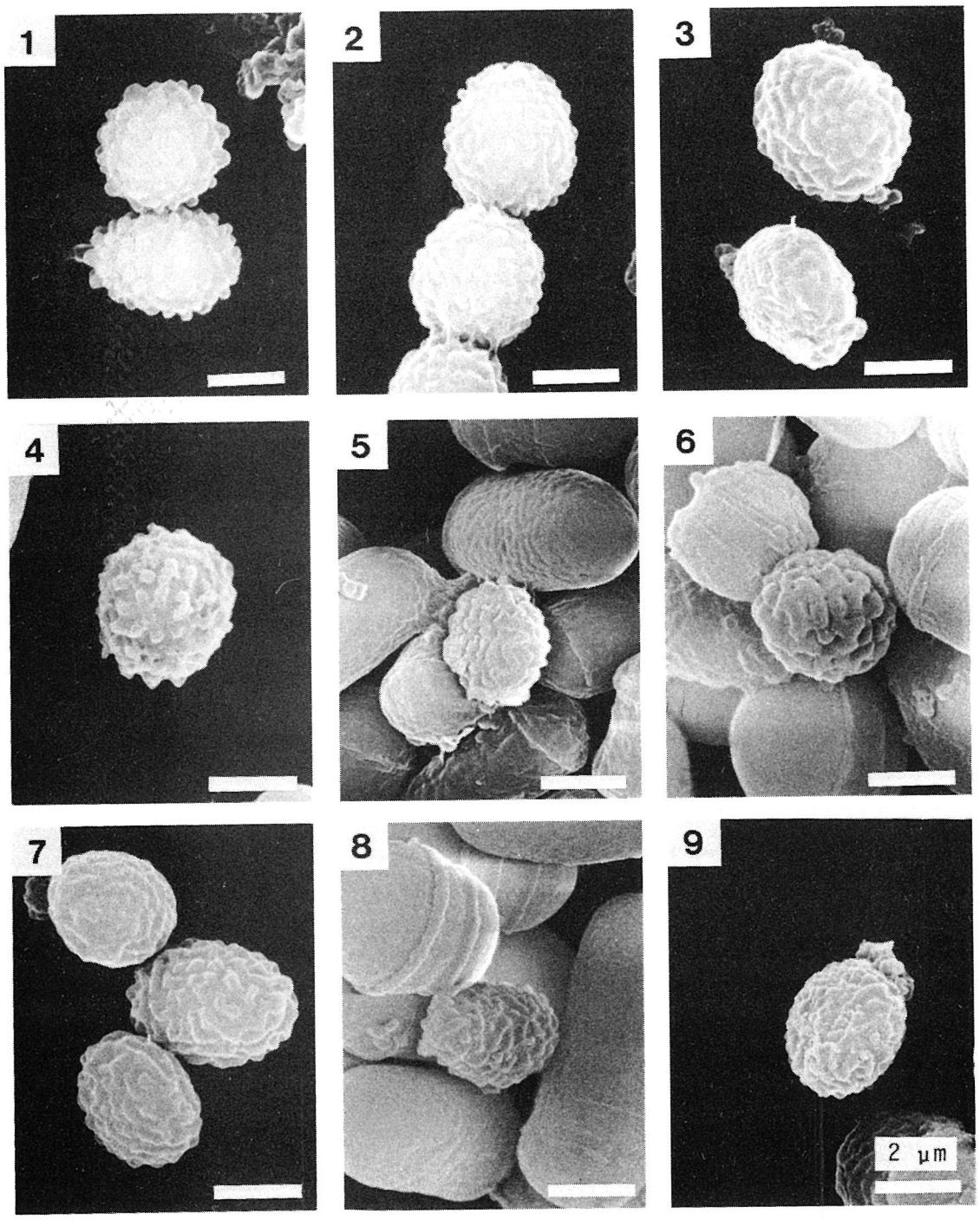
Spherical to short ellipsoidal ascospores were found. They have no ornamentation on the surface although they are not completely smooth.

S. japonicus var. versatilis. (Fig. 16)

The spores were reniform. No ornamentation was found on the surface although the surface is not entirely smooth.

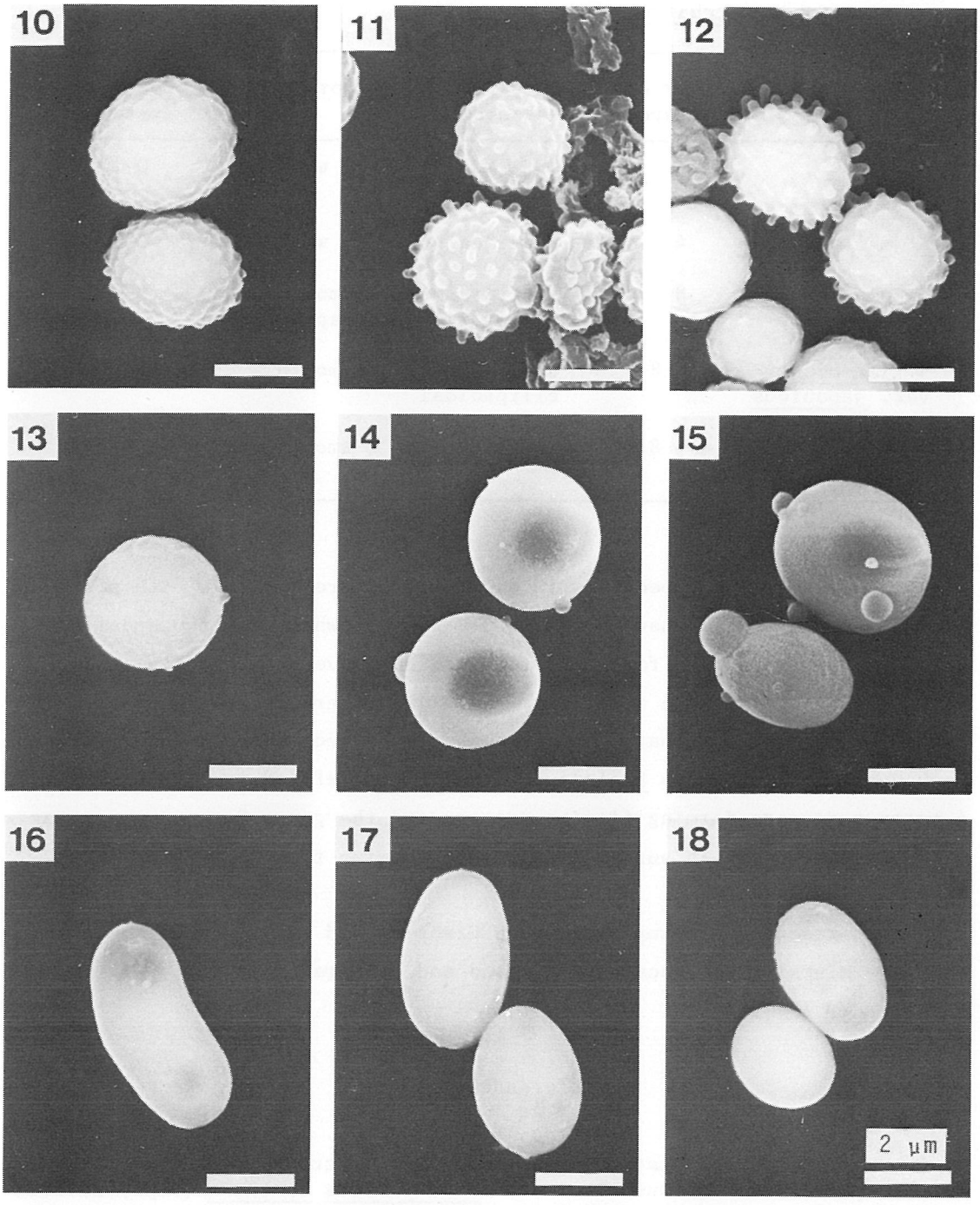
The surface-structure of ascospores of the genus is classified into 3 categories: warty, smooth with occasional papillae, and smooth.

In 1960, Kudriavzev (3) divided Schizosaccharomycetaceae into two genera: 1. Schizosaccharomyces Lindner, spores ellipsoidal, four per ascus, and 2. Octosporomyces Kudriavzev, spores bean-shaped, eight per ascus. He included S. octosporus, S. versatilis and S. japonicus in the second genus. Yamada et al. (5) have reported that the Co-Q system of S. pombe and S. malidevorans was Q-10 and that of S. octosporus, S. japonicus var. japonicus and S. japonicus var. versatilis was Q-9. It should be



Figs. 1-9. Scanning electron micrographs of ascospores of *S. pombe*.

1. IFO 0342, 2. IFO 0343, 3. IFO 0344, 4. IFO 0345, 5. IFO 0346, 6. IFO 0347, 7. IFO 0349, 8. IFO 0363, 9. IFO 0366.



Scanning electron micrographs of ascospores.

Figs. 10 and 11. *S. pombe*. 10. IFO 0638, 11. IFO 1628.
Fig. 12. *S. malidevorans* IFO 1608.
Figs. 13-15. *S. octosporus*. 13. IFO 0353, 14. IFO 0360, 15. IFO 0361.
Fig. 16. *S. japonicus* var. *versatilis* IFO 1607.
Figs. 17 and 18. *S. japonicus* var. *japonicus*. 17. IFO 1609, 18. IFO 1713.

Table 2. Possibly related characteristics of the species in the genus Schizosaccharomyces.

Species and variety	Number of ascopspores	Shape of ascospore	Spore surface	Coenzyme Q system
S. pombe	4	spherical to ellipsoidal	warty	Q-10
S. malidevorans	4	spherical	warty	Q-10
S. octosporus	8	spherical to ellipsoidal	smooth with papillae	Q-9
S. japonicus var. japonicus	6 - 8	spherical to ellipsoidal	smooth	Q-9
S. japonicus var. versatilis	6 - 8	reniform	smooth	Q-9

stressed that the two species with smooth ascospore-surfaces both produce 8 spores per ascus and have coenzyme Q-9 system, while the two species showing a warty surface form 4 spores asci and have the Q-10 system in common (Table 2). These three correlated characteristics support Kudriavzev's proposal that two genera should be recognized in Schizosaccharomycetaceae. The genus Schizosaccharomyces is defined as Q-10-equipped, 4 warty ascospore-forming fission yeasts, and the genus Octosporomyces as Q-9-equipped, 8 smooth surfaced ascospore-forming fission yeasts.

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TESTS FOR MYCOPLASMAL CONTAMINATION IN CELL LINES COLLECTED IN IFO

TOUHO YOSHIDA AND MASAO TAKEUCHI

Summary

A survey was made of mycoplasmal contamination in cell lines that were submitted to the Institute for Fermentation, Osaka (IFO). A total of 212 cell lines was tested by both the DNA staining and microbiological culture methods. Of the fifty-five cell lines (26%) that were positive by the DNA staining method, 10 were not detected by the culture method. The false-negative contaminants by the culture method were confirmed as mycoplasmas by scanning electron microscopy. Among the 45 cell lines that were detected by the culture method, 31 were detected both by aerobic and anaerobic incubations, whereas 8 were detected only by aerobic incubation and 6 only by anaerobic incubation. Furthermore, 22 of the cell lines were contaminated with fermentative mycoplasmas and 23 were contaminated with nonfermentative mycoplasmas.

Mycoplasmal contamination of cell lines is common and causes alteration of normal cell structure and function (9). In large surveys made in the United States using more than 17,000 cell cultures (1,4,9), the reported rate of contamination ranged from 5.1 to 16.5%. In Japan, Ogata et al. (12) reported a high rate of contamination (86.7%) in 60 cell lines; Koshimizu et al. (6) summarized three different surveys in which 78 out of 90 cell lines (87.0%) were contaminated with mycoplasmas; and, recently, Mizusawa (11) reported that 32 cell lines out of 164 (29.1%)

were contaminated with mycoplasmas.

A culture collection of animal cell lines was started in May, 1984. As one of the quality controls on the cell lines, tests for mycoplasmal contamination were conducted using both the DNA staining and microbiological culture methods. In this report, the detailed results of these tests on 212 cell lines are presented.

Materials and Methods

Cell cultures. Two hundred and twelve cultures to be tested for mycoplasma detection were submitted by 23 laboratories and preserved at the Institute for Fermentation, Osaka (IFO). Of these, 89 were from human beings, 66 from mice, 28 from rats, 12 from hamsters, 7 from monkeys, 4 from cows, 2 from rabbits, 2 from dogs, 1 from a potoroo, and 1 from a mosquito. Test samples containing cells and their cultured medium were taken 3 to 6 days after the last subculture. All cell cultures were cultivated in antibiotic-free media.

DNA staining method. A slightly modified version of Hay's method (5) was used. CKT-1 cells (calf kidney; IFO 50003) were used as indicator cells. This cell line was cultivated in Eagle's Minimum Essential Medium (MEM) containing 10% fetal bovine serum (FBS). CKT-1 cells were seeded on a coverslip in a well of a 24-well flat bottomed plate. After 15 hr, test samples were inoculated into each well. The plate was incubated for 5 days at 37 C and then the supernatants were discarded. CKT-1 cells on a coverslip were fixed and stained with Hoechst 33258 (Wako Pure Chemical Ind., Osaka). The coverslips containing the specimen were mounted on slide glasses and observed by fluorescence microscopy at x 400. Approximately 1,000 cells per coverslip were examined and the results were expressed as (+) if more than 0.5% of cells had perinuclear fluorescence, (-) if less than 0.5% of cells had perinuclear fluorescence.

Culture method of contaminated mycoplasmas. A slightly modified version of Hay's method (5) was used. A broth medium consisting of 60 parts basal medium (Bacto PPLO broth, Difco Laboratories, Detroit, MI), 20 parts horse serum, 10 parts of a 25% extract of fresh yeast, and 10 parts of a supplement solution containing dextrose, L-arginine, thymic DNA, choline chloride, inositol, niacinamide, D-calcium pantothenate,

pyridoxal HCl, folic acid, riboflavin, cyanocobalamin, D-biotin, thiamine HCl, penicillin, phenol red, and thallium acetate was used. The final pH was adjusted to 7.2. Noble agar (Difco Laboratories, Detroit, MI) (1.4%) was added for an agar medium. Test samples (0.1 ml) were inoculated into the broth medium and onto the agar plates and incubated at 37 C aerobically and anaerobically. After 7 and 14 days, 0.1 ml of the broth media was secondarily transferred onto the agar plates, which were then cultivated for 21 days. The broth media were examined for a shift in pH and the agar plates were examined for colony formation.

Scanning electron microscopy. Morphology was examined by scanning electron microscopy using the method of Carson et al. (3). Sample inoculations onto CKT-1 cells and cultivation were made by the same procedures as the DNA staining method. CKT-1 cells on a coverslip were fixed in 2% glutaraldehyde and postfixed with 1% osmium tetroxide. After dehydration in a graded series of ethanols, the specimens were dried in a critical-point bomb, coated with gold and then examined with a Hitachi S-570 type scanning electron microscope operated at 20 KV.

Results and Discussion

Detection of mycoplasmas by DNA staining and culture methods

Of the cell lines 55 (26%) that were found positive by the DNA staining method, 10 were not detected by the culture method (Table 1). The DNA staining method is not diagnostic for mycoplasmas because Hoechst 33258 binds to the DNA of other prokaryotic nonmycoplasma organisms. To determine whether fluorescent bodies in the Hoechst stained preparations coincide with mycoplasmas, CKT-1 cells infected with samples from the 10 false-negatives of the culture method were examined by scanning electron

Table 1. Detection of mycoplasmal contamination in 212 cell lines by DNA staining and culture methods.

Results of Detection		No. Detected
DNA staining	Culture	(%)
+	+	45 (21)
+	-	10 (5)
-	+	0 (0)
-	-	157 (74)

microscopy. The morphology of the contaminant on CKT-1 cell surfaces was generally pleomorphic: spheres, chains of spheres or elongated forms, frequently characterized by a dimple (Figure 1). From these morphological characteristics, the contaminants in the 10 cell lines were confirmed as mycoplasmas. Further characterization using an immunofluorescent staining method (2) must be performed to identify the mycoplasma species. Recently, McGarrity et al. (10) reported that 56.4% of the mycoplasmas isolated from cell cultures were Mycoplasma hyorhinis, none of which grew on mycoplasma media. In the current experiments, Mycoplasma hyorhinis BTS7 grew well on the mycoplasma media, but Mycoplasma hyorhinis DBS1050 failed to grow on the same media (data not shown). The 10 false-negatives from the culture method are considered to be caused by contamination by fastidious strains of mycoplasmas, such as Mycoplasma hyorhinis DBS1050.

The rate of contamination in these results (26%) is significantly lower than those of surveys reported by Ogata et al. (12) and Koshimizu et al. (6) and is closer to the rate of 29.1% reported by Mizusawa (11). However, the rates from the surveys made in Japan are higher than those (13.8%, 16.5%, 5.1%) from surveys made in the United States (1,4,9). The variations in the rate of contamination might be primarily due to the population of cell line being tested. In the current results, no relation between the rate of contamination of the cell lines and their origin was found (Table 2). The chances for contamination increase with the increasing number of passages. Most of cell lines tested in this survey were long-term cultures. The lower rate of contamination (5.1%) reported by McGarrity et al. probably indicates that their population survey

Table 2. Origins of cell lines tested and the rate of detection of contamination by the DNA staining method.

Origins	No. Tested	No. Detected (%)
Human	89	24 (27)
Mouse	66	17 (26)
Rat	28	6 (21)
Hamster	12	5 (42)
Monkey	7	3 (43)
Cow	4	0 (0)
Rabbit	2	0 (0)
Dog	2	0 (0)
Potoroo	1	0 (0)
Mosquito	1	0 (0)
Total	212	55 (26)

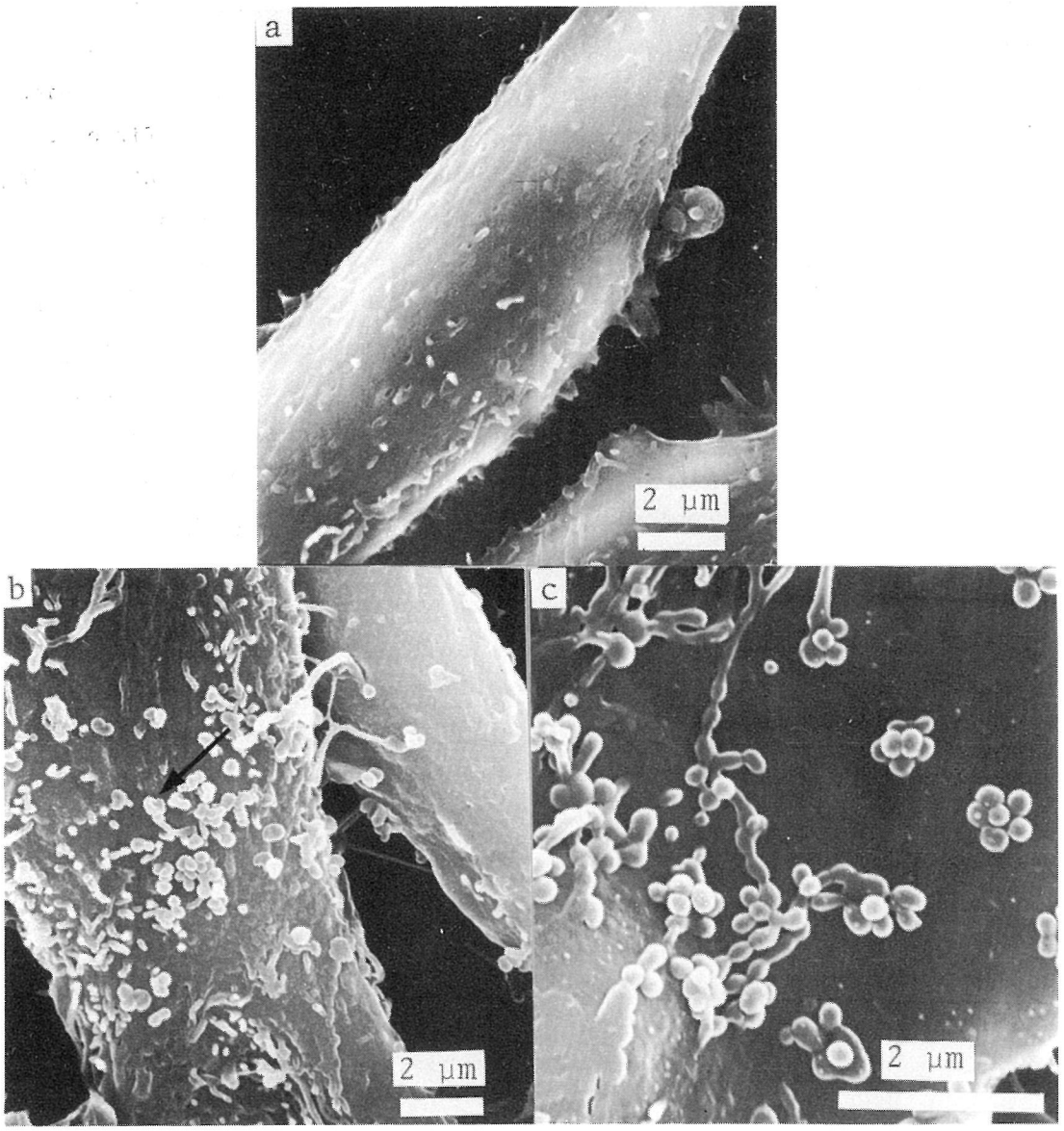

Fig. 1. Scanning electron micrographs of control cells (CKT-1 cells) and artificially contaminated cells.

(Fig. 1.a); Control cells. Most cells have smooth surfaces with very few microvilli. (Fig. 1.b); CKT-1 cells contaminated with specimens of WI-38 cells. Many particles attached to cell surfaces of CKT-1 cells are seen. A dimple in a particle is characteristic of mycoplasmas (arrow). (Fig. 1.c); CKT-1 cells contaminated with specimens of HeLa-P3 cells. Elongated forms and chains of spheres are seen.

included primary cell cultures (9). The cell lines used in this experiment were from laboratories that have shown relatively good quality control. The average rate of mycoplasmal contamination in cell lines maintained in Japan might be higher than 26%; more extensive surveys using various cell lines from many laboratories must be made to determine the current rate.

Characteristics of the contaminating mycoplasmas

To examine the characteristics of the contaminating mycoplasmas, we compared the efficiency of incubation conditions for primary isolations of mycoplasma media. As shown in Table 3, 31 out of 45 cell lines (69%) were detected by both aerobic and anaerobic incubations, whereas 8 (18%) were detected only by aerobic incubation and 6 (13%) were detected only by anaerobic incubation. In general, an anaerobic atmosphere is considered optimal for mycoplasma isolation (7). However, Acholeplasma laidlawii, a common cell culture contaminant, was occasionally isolated only aerobically (8). The reason for the high rate of detection achieved only by aerobic incubation is unclear although it may be due to the high frequency of contamination by Acholeplasma laidlawii.

The mycoplasma broth medium that contains glucose, arginine, and a dye indicator provides information on the major metabolic pathways of the contaminating mycoplasmas. Mycoplasmas are divided in two large groups: fermenters and nonfermenters. Most nonfermenters hydrolyze arginine and release ammonia, which causes an alkaline shift in the pH of the culture medium, whereas fermenters cause an acidic shift. Among 45 cell lines, 22 (49%) produced acidic shifts in the pH of the broth media and 23 (51%) produced alkaline shifts (Table 4). These results indicate that 49% of cell lines were contaminated with fermentative and 51% were contaminated with nonfermentative mycoplasmas. The species of contaminating mycoplasmas is now being characterized further by the immunofluorescent staining method.

Table 3. Incubation for detection of mycoplasmas.

Conditions	No. Detected (%)
Aerobic & Anaerobic	31 (69)
Aerobic only	8 (18)
Anaerobic only	6 (13)

Table 4. Characteristics of the contaminating mycoplasmas.

Shift in pH of Mycoplasma Broth	No. Shifted (%)
Acidic	22 (49)
Alkaline	23 (51)

The authors gratefully acknowledge the helpful discussions with Dr. Hiroshi Mizusawa, National Institute of Hygienic Sciences, throughout the course of this investigation.

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PRESERVATION OF YEAST CULTURES BY FREEZING AT -80 C: I. VIABILITY AFTER 2 YEARS STORAGE AND THE EFFECTS OF REPEATED THAWING-FREEZING

KOZABURO MIKATA and ISAO BANNO

Summary

Freezing at -80 C has been applied to the preservation of 141 yeast cultures which were very sensitive to L-drying. The cells were frozen in 10% glycerol solution in an electric deep-freezer, and their survival value was determined immediately after freezing and after preservation of 1, 6 and 24 months. All cultures except 7 strains showed high viability. After 24 months storage, the viability of Candida bogoriensis IFO 1966, C. slooffii IFO 0874, Leucosporidium nivale IFO 1852, Lipomyces starkeyi IFO 1289, Nadsonia commutata IFO 10029, Saccharomyces exiguus IFO 1169 and Torulopsis holmii IFO 0660 was less than 1%, too low for these cultures to survive over a long term.

Using strains IFO 1966, IFO 1289 and IFO 1169, the effect of 5 cryoprotectants and repeated thawing-freezing on the viability of these cultures was examined. The viability gradually decreased with the number of thawings. DMSO gave the best viability for all 3 strains.

Ultra-low freezing in liquid nitrogen (L.N.) is widely used in the preservation of microorganisms. Yeast and yeast-like fungi are able to survive after freezing in L.N., and various yeast species have been

successfully stored in L.N. by several laboratories. The viability of the cells of 19 different species was kept unchanged and their phenotipic character remained stable after 5 years of storage in L.N.(1,4).

Although preservation in L.N. has been found to be reliable, L.N. refrigeration is expensive, and the handling of L.N. is troublesome. Furthermore, L.N. is unavailable in some districts. As an alternative, commercial deep-freezers with temperature ranges of -70 to -100 C are readily obtainable.

Storage at -70 C or lower has been used mainly for a variety of bacteria (Jones et al. and Okuno et al.)(3,6). Ito and Yokoyama (2) reported that 93% of mycelial basidiomycetes survive after one year of storage at -80 C. In yeasts, Sardjono et al.(7) obtained survival values ranging from 10 to 80% immediatly after freezing up to -70 C at a rate of 10 C/min with 7 different yeasts. Viability after long-term preservation of yeasts by freezing at -60 to -100 C has not been reported. Consequently, a systematic investigation was conducted to ascertain whether or not refrigeration at -80 C is applicable to long-term preservation of various kinds of yeast cultures.

Materials and methods

Strains. The strains tested (Tables 1,2 and 3) belong to 22 genera and 66 species. They were chosen because of their high sensitivity to storage by L-drying, giving viabilities of less than 1% of their cells after accelerated tests in previous work (5).

Media. YM medium (peptone 0.5%, dehydrated yeast extract 0.3%, dehydrated malt extract 0.3%, glucose 1% and agar 1.5%) was used for growth and viability measurement.

Preparation of cultures. Each strain was grown on YM agar slant for 4 days at 24 C. The cells harvested from the agar slope were suspended in a solution containing cryoprotectant so that their density might be about 10^8/ml. Aliquots of 1 ml of the suspension were distributed in NUNC plastic vials.

Freezing. The vials were directly transferred into a cabinet of the freezer set at -80 C and were rapidly frozen without programed control.

Viable count. The frozen vial was warmed and quickly thawed in a

Table 1. Viability of ascosporogenous yeast strains frozen in 10% glycerol.

Species	IFO No.	Viable count before freezing 10^7CFU/0.1ml	Survival value (%) after preservation at -80C for 1M	6M	24M
Ambrosiozyma cicatricosa	1846	0.34	59.6	42.1	36.8
A. monospora	1965	0.31		93.5	32.3
A. monospora	4841	0.42	45.2	40.0	22.4
A. philentoma	1847	1.43		55.9	39.2
Arthroascus javanensis	1579	1.84			31.5
Debaryomyces coudertii	1381	2.86			55.9
D. coudertii	1817	4.05			54.6
D. polymorphus	1166	6.80			8.6
D. polymorphus	1357	2.75			16.6
D. tamari	0854	5.85			54.1
Dekkera bruxellensis	1590	6.00			30.2
Endomyces ovetensis	1201	0.33	87.6	69.7	19.6
Kluyveromyces marxianus	0219	1.83		54.6	39.3
K. phaffii	1883	13.15			32.4
K. phaffii	1884	5.55		16.0	12.6
K. phaffii	1885	5.20		20.6	15.7
K. polysporus	0996	8.15			21.5
Lipomyces lipofer	0673	2.60	17.3	6.9	2.6
L. lipofer	1288	0.64	15.1	6.1	2.2
L. starkeyi	0678	1.88	20.7	3.4	1.3
L. starkeyi	1289	7.80		3.8	0.8
Nadsonia commutata	10029	1.25	38.4	0.2	0.001
Nematospora coryli	0658	0.73	49.7	45.2	34.4
N. coryli	1220	1.83		42.6	31.5
Pichia angophorae	10016	5.75			47.0
P. chamberdii	1029	5.65			7.8
P. fermentans	0815	5.85			29.0
P. fluxuum	0773	8.60			37.3
P. membranaefaciens	0457	8.50			35.9
P. membranaefaciens	0460	6.70			47.2
P. membranaefaciens	0814	5.95			32.3
P. pinis	1342	3.55			35.2
P. stipitis	1687	23.00			40.0
r. stipitis	1720	8.25			19.7
P. stipitis	10006	15.30			36.4
P. stipitis	10007	13.25			31.0
Saccharomyces cerevisiae	0573	3.03	94.0		76.5
S. cerevisiae	0636	2.01	69.3		59.3
S. dairensis	10008	5.65			22.6
S. dairensis	10009	5.45			32.3
S. exiguus	0215	5.25			56.2
S. exiguus	0271	11.00			19.8
S. exiguus	0956	7.90			43.0
S. exiguus	1128	15.07			23.3
S. exiguus	1141	3.52		16.5	11.9
S. exiguus	1142	6.75		3.4	1.1
S. exiguus	1169	7.00		0.1	0.006

Table 1. (continued)

Species	IFO No.	Viable count before freezing 10^7CFU/0.1ml	Survival value (%) after preservation at -80C for 1M	6M	24M
Saccharomyces exiguus	1170	11.40			14.9
S. exiguus	1616	3.74	17.9	4.3	2.2
S. telluris	1017	0.43	76.7	10.7	2.9
S. telluris	1329	1.46	72.1	23.3	9.2
S. telluris	1330	1.64		46.3	23.6
S. telluris	1331	1.87	54.7	51.0	45.8
Schizosaccharomyces japonicus var. japonicus	1713	0.31	74.2	12.9	3.1
Schwanniomyces occidentalis	1841	10.45			22.9
Zygosaccharomyces rouxii	0320	1.61			65.1
Z. rouxii	0325	5.55			40.5
Z. rouxii	0326	2.50			61.5
Z. rouxii	0328	6.50			41.5
Z. rouxii	0331	3.18			69.2
Z. rouxii	0444	2.07			74.2
Z. rouxii	0542	6.25			57.0
Z. rouxii	0570	5.90			48.0
Z. rouxii	0595	2.18			39.2
Z. rouxii	0596	5.35			43.7
Z. rouxii	0597	3.09			49.4
Z. rouxii	0687	1.09		76.2	68.5
Z. rouxii	1055	2.61			62.5
Z. rouxii	1615	3.21			66.6
Z. rouxii	1731	3.49			53.7
Z. rouxii	1732	4.14			34.1
Z. rouxii	1733	2.47			60.9
Z. rouxii	1877	1.58		18.4	8.8
Z. rouxii	1946	4.32			28.4

water bath regulated at 35 C. An aliquot of melted suspension was drawn from the vial and diluted with saline. A ten-fold dilution series prepared from each culture was plated on to YM agar plate by the overlay plating method as previously described (4).

Cryoprotectants. Glycerol, dimethyl sulfoxide (DMSO), polyethylene glycol (PEG) 4000, PEG 20000 and sorbitol were tested as protectants.

Results and Discussion

Viabilities of 141 strains after preservation at -80 C

Cells of 141 yeasts suspended in 10% glycerol solution were frozen

Table 2. Viability of asporogenous yeast strains frozen in 10% glycerol.

Species	IFO No.	Viable count before freezing 10^7CFU/0.1ml	Survival value (%) after preservation at -80C for 1M	6M	24M
Brettanomyces bruxellensis	0628	10.20			38.6
B. bruxellensis	0629	6.25			35.4
B. bruxellensis	0677	8.00			47.3
B. custersianus	1585	2.84			58.8
B. intermedius	1587	0.88	78.0	60.2	38.6
Candida australis	1515	5.20			31.7
C. australis	1516	5.20			51.5
C. australis	1517	3.19			43.3
C. australis	1518	3.85			35.3
C. australis	1519	5.75			36.2
C. australis	1520	2.72			37.5
C. bogoriensis	1966	0.39	3.1	1.5	0.2
C. boidinii	1967	6.80			45.4
C. buinensis	1642	13.10			28.7
C. curvata	1159	2.17	61.8	36.9	17.1
C. diversa	0861	13.25			26.4
C. diversa	1085	11.25			30.3
C. diversa	1091	18.40			44.8
C. humicola	1527	2.54			33.5
C. mesenterica	0969	0.93	62.4	29.0	22.6
C. mesenterica	1123	0.94	85.1	71.3	63.4
C. mesenterica	1210	7.15			41.9
C. slooffii	0874	0.27	18.1		0
Cryptococcus albidus	1322	2.70	27.3	16.3	8.9
C. dimennae	1863	1.85	51.4	42.2	9.7
C. hungaricus	1052	0.43	7.0	4.0	2.0
C. hungaricus	1380	3.51	23.4	4.7	3.3
Rhodotorula acheniorum	10052	0.08	51.3	27.5	26.0
R. araucariae	10054	0.76	13.2	5.3	4.2
R. glutinis	0391	1.53	99.2	56.9	49.5
Sporobolomyces odorus	1110	1.78	55.1	47.8	30.1
S. odorus	1597	1.52	32.9	14.5	8.5
S. odorus	1606	9.15			28.4
Torulopsis bovina	0873	1.57	52.9	29.9	5.8
T. bovina	1018	2.09			31.8
T. bovina	1069	0.50	54.0	38.0	14.6
T. bovina	1087	1.68	48.2	16.7	2.4
T. bovina	1312	1.01	79.2	63.4	57.3
T. bovina	1313	1.64	68.3	43.3	29.8
T. bovina	1315	0.44	92.3	86.4	84.9
T. etchellsii	1229	13.80			48.8
T. etchellsii	1592	10.10			31.7
T. fructus	1581	2.00			74.3
T. halonitratophila	1595	0.41		97.6	97.6
T. halonitratophila	1906	5.25			10.0
T. holmii	0660	2.79	12.5	4.3	0.9
T. holmii	1629	6.30	15.6	8.4	2.8
T. ingenniosa	10002	0.06	40.0	21.7	13.2

Table 2. (continued)

Species	IFO No.	Viable count before freezing 10^7CFU/0.1ml	Survival value (%) after preservation at -80 C for 1M	6M	24M
Tolulopsis lactis-condensii	1324	0.48	72.9	69.2	59.4
T. lactis-condensii	1325	1.65			71.9
T. lactis-condensii	1326	0.59	83.1	78.0	58.1
T. pinus	1327	8.85			67.5
T. pintolopesii	0729	0.67	67.2	53.7	45.3
T. psychrophila	1532	3.70			55.1
T. psychrophila	1533	0.19			57.3
Trichosporon pullulans	0114	0.86	35.4	29.1	14.1

and preserved at -80 C. The frozen cells were thawed as indicated and 0.1 ml aliquot of the suspension was sampled for estimation of viability. The suspension was refrozen immediately after the sampling and stored. Viability was determined before freezing and after 1, 6, and 24 months of storage.

The results obtained from the 141 yeast cultures are presented in Tables 1, 2, and 3. All cultures except Candida slooffii IFO 0874 survived after 24 months of storage. Viabilities varied between 0.001 and 97.6% of those prior to freezing. The average of viabilities was 33% in ascomycetous yeast, 34% in asporogenous yeast and 27% in Leucosporidium (basidiomycetous yeast). A significant difference in viability was not found among the 3 groups.

C. slooffii IFO 0874 survived (18%) at the first thawing after 1 month, but was not viable after 24 months. Viabilities of Nadsonia commutata IFO 10029 and Saccharomyces exiguus IFO 1169 were so extremely low that they would rapidly die out in further storage. Four strains, Candida bogoriensis IFO 1966, Leucosporidium nivale IFO 1852, Lipomyces starkeyi IFO 1289 and Torulopsis holmii IFO 0660 gave viabilities of less than 1%, demonstrating that the cells of these cultures would not survive for long. With all the other cultures, the rate of decrease from 6 months to 24 months is slower than that from one month to 6 months. Owing to their reduced rate of viabily, it may be predicted that these cultures will be alive for at least 10 years in a frozen state at -80 C even if their refrigeration is interrupted several times by thawing.

The viability of Lipomyces starkeyi after one month of storage was much higher, but after 24 months of storage with 3 thawings was lower

Table 3. Viability of basidiosporogenous yeast strains frozen in 10% glycerol.

Species	IFO No.	Viable count before freezing 10^7CFU/0.1ml	Survival value (%) after preservation at -80C for 1M	6M	24M
Leucosporidium antarcticum	1917	0.80		2.5	2.3
L. antarcticum	1918	1.14	38.6	37.5	21.1
L. antarcticum	1919	2.13	39.0	32.2	28.3
L. frigidum	1851	0.73	72.6	70.1	64.4
L. frigidum	1920	3.15	23.5	21.6	16.6
L. gelidum	1921	1.71	18.8	12.3	11.4
L. nivale	1852	0.12	1.4	1.0	0.8
L. nivale	1922	2.49		51.0	47.1
L. scottii	1287	0.59	45.8	30.5	15.3
L. scottii	1304	9.25			49.5
L. stokesii	1926	6.95			43.7

than that found in the L.N. storage reported by Hubalek and Kockova-kratochvilova (1).

It is not clear from the present results whether the decrease in viability during storage at -80 C with repeated thawings is due to the period of preservation or the number of thawings. Paralleled suspensions of all the cultures have been stored in a frozen state without thawing for recovery after 5 years. This question will be answered by the survival values shown when these suspensions are first revived after 5 years of freezing.

Although viability after storage at -80 C does not always correlate with that after storage by L-drying for each strain, the five strains S. exiguus, C. bogoriensis, Leucosporidium nivale, Lipomyces starkeyi, and T. holmii, which all showed extremely low viabilities in L-dried cultures, similarly gave the lowest viabilities in storage at low temperature in this experiment. This might imply that one of mechanisms responsible for reduction of viability is common to L-drying and freezing.

Cryoprotectants and repetition of thawing

The effects of protective agents and of periodical thawing-freezing on the viability of cells preserved at -80 C were investigated using 3 strains which showed very low viability in the experiment mentioned above, C. bogoriensis IFO 1966, L. starkeyi IFO 1289, and S. exiguus IFO 1169, and one strain, S. cerevisiae IFO 0213, as a control. Glycerol at concentrations of 10% and 20%, DMSO at 10%, PEG(4000) at 10%, PEG(20000) at

10%, and sorbitol at 10% were tested as protectants for freezing. Cell suspensions in each protectant solution were frozen at -80 C in the freezer. The frozen suspensions were subjected 20 times to thawing and freezing at intervals of one or two days for 30 days. At the 1st, 5th, and 20th thawing, 0.1 ml aliquots of the suspension were withdrawn and viable counts were determined. Another set of suspensions were kept frozen at -80 C for 30 days before being thawed to determine viability.

The results obtained for the 4 yeasts are shown in 4 semilogarithmic graphs. As Fig. 1 shows, S. cerevisiae 0213 was very stable and gave good survival value even after 20 repetitions of thawing, although a slight loss was found without the protectant. This strain is considered to be strongly resistant to freezing and thawing.

In C. bogoriensis 1966 (Fig. 2), a great loss of viable cells occurred even when the cells were transferred to glycerol and DMSO solutions. It is probable that this strain is sensitive to osmotic shock. Viability with 4 protectants was reduced linearly in accordance with the number of thawings. The reduction rate was very low and high survival values were obtained with 10% DMSO, 20% glycerol and 10% PEG. Evidently DMSO and PEG(6000) are beneficial for this strain.

In L. starkeyi 1289 (Fig. 3), a slight loss of viable cells was found when the cells were suspended in glycerol solution. Viability at

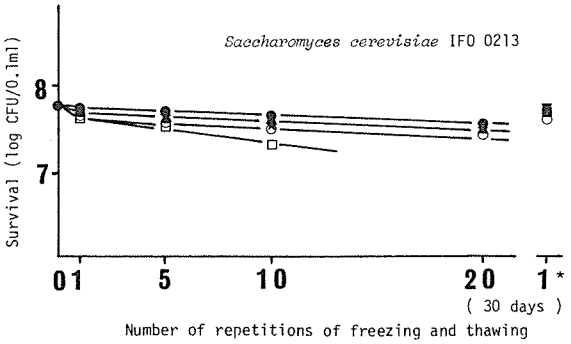

Fig.1. Effect of the frequency of freezing and thawing on viability in deep-freeze preservation.
(●) 10% polyethylene glycol 4000, (▽) 10% glycerol,
(○) 10% polyethylene glycol 20000, (▲) 20% glycerol,
(★) 10% dimethyl sulfoxide, (■) 10% sorbitol,
(□) distilled water.

*The survival value in samples which were kept in a frozen state at -80C and thawed first after 30/days.

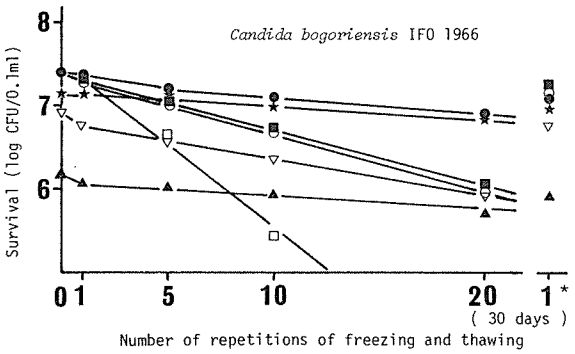

Fig.2. Effect of the frequency of freezing and thawing on viability in deep-freeze preservation.
(●) 10% polyethylene glycol 4000, (▽) 10% glycerol,
(○) 10% polyethylene glycol 20000, (▲) 20% glycerol,
(★) 10% dimethyl sulfoxide, (■) 10% sorbitol,
(□) distilled water.

*The survival value in samples which were kept in a frozen state at -80C and thawed first after 30/days.

the first thawing was 60% of that prior to freezing with each protectant. After that, approximately linear reduction of viability was found in accordance with the number of thawings. A higher survival rate was obtained in the order of 10% sorbitol, 10% DMSO, 20% glycerol, 10% glyc erol, 10% PEG(4000) and 10% PEG(20000).

In S. exiguus 1169, almost the same survival curves as those of L. starkeyi 1289 are seen in Fig. 4. A higher survival rate is found in the order of 10% sorbitol, 10% DMSO, 10%, 20% glycerol and 10% PEG(4000 and 20000). DMSO provided the highest viability in all 3 strains. It is therefore concluded that DMSO is the best protectant for freezing.

Viability when the suspension was first thawed after 30 days was almost equal to the values obtained immediately after freezing (Figs. 1-4). The results indicate that there was no loss of viability during preservation in a frozen state for 30 days. The decline of viability shown in Figs. 2 to 4 is due to the repetition of thawing and freezing, but not to the storage period. It is therefore desirable to avoid intermitant thawing of the frozen state to ensure permanent preservation.

Although a direct comparison can not be made because of differences in tested strains, the survival rates after uncontrolled freezing in the

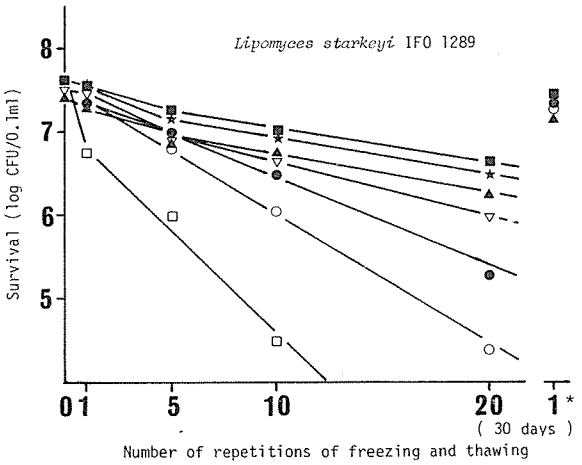

Fig.3. Effect of the frequency of freezing and thawing on viability in deep-freeze preservation.
(●) 10% polyethylene glycol 4000, (▽) 10% glycerol,
(○) 10% polyethylene glycol 20000, (▲) 20% glycerol,
(★) 10% dimethyl sulfoxide, (■) 10% sorbitol,
(□) distilled water.

*The survival value in samples which were kept in a frozen state at -80C and thawed first after 30/days.

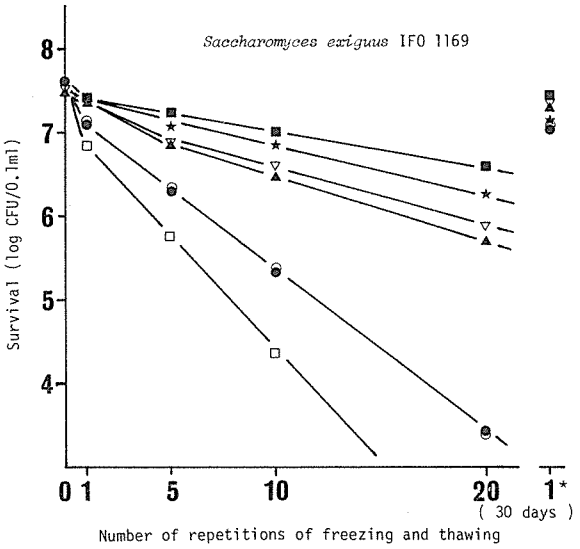

Fig.4. Effect of the frequency of freezing and thawing on viability in deep-freeze preservation.
(●) 10% polyethylene glycol 4000, (▽) 10% glycerol,
(○) 10% polyethylene glycol 20000, (▲) 20% glycerol,
(★) 10% dimethyl sulfoxide, (■) 10% sorbitol,
(□) distilled water.

*The survival value in samples which were kept in a frozen state at -80C and thawed first after 30/days.

present work were compared to those after freezing to -70 C at an appropriate rate reported by Sardjono et al. (7). The cooling rate does not seem to be such a severe factor in freezing general yeast cultures for preservation. Uncontrolled freezing in a refrigerator at -80 C or so is satisfactory for ordinary preservation.

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FURTHER INVESTIGATION ON THE PRESERVATION OF BASIDIOMYCETE CULTURES BY FREEZING

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Summary

Basidiomycete cultures maintained at the Institute for Fermentation, Osaka (IFO) have been stored in an electric refrigerator at -80 C with 10% glycerol as a cryoprotective agent. Among a total of 939 strains of the cultures tested, 865 strains survived after five years of storage. The percentage of viability obtained in this experiment was 92.1% and showed a mere 1.4% decline when compared with the results obtained after one year of storage. The results clearly show that the procedure is very effective for long-term preservation of nonsporulating cultures, such as mycelial Basidiomycetes, which have, hitherto, been considered difficult to preserve by freeze-drying and/or L-drying methods.

Various methods for long-term preservation have been developed and commonly used in order to avoid the degeneration or mutation of microorganisms. In particular, the preservation of fungi by freezing in an electric refrigerator is a simple and practical method which has already been applied by Carmichael (1,2), Hamilton and Weaver (3), and Kramer and Mix (5). This method has also been applied by Ito and Yokoyama (4) for the preservation of basidiomycete cultures, which consist mainly of

nonsporulating strains, and also by Yokoyama and Ito (6) for other fungal cultures.

A previous paper (4) showed the viability of basidiomycete cultures stored for one year at -80 C. The current paper reports the result obtained from further experiment after storage for five years at -80 C.

Materials and Methods

All 939 strains of the species of Basidiomycotina tested in the previous report (4) were tested again for their survival rate, with the exception of one strain which was discarded from the collection during the testing period.

These strains have been stored for five years at -80 C in an electric refrigerator and the second cryotubes of the same lot, each containing five mycelial plugs, were used for this experiment. The procedures for the thawing and incubating of frozen mycelial disks were exactly the same as described in our previous paper (4).

Results and Discussion

Table 1 shows the number of tested strains, the number of survived strains after one year and five years of preservation, and the percentage of survived strains per tested strains of the basidiomycete fungi which are grouped in taxonomic rank of the order.

Of a total 939 strains tested, 865 strains survived after five years of storage at -80 C, and the overall percentage of the number of survived strains / number of tested strains was 92.1%. Since the percentage of the number of survived strains / number of tested strains after one year of storage was 93.5% (4), a decline in viability was recorded at 1.4% during the storage.

Seventy-four strains, however, were not recovered, including four strains of Aphyllophorales, fifty-nine of Agaricales, four of Lycoperdales, two of Hymenogastrales, and five of Auriculariales. Five strains of Hymenogastrales among the seven strains tested survived, indicating the same result as in the previous test (4). Among these

Table 1. Viability of strains of basidiomycete fungi by freezing at -80 C.

Order	Number of tested strains	Number of survived strains after 1 year (%)	after 5 years (%)
Aphyllophorales	395*	392 (99.2)**	391 (99.0)
Agaricales	427	377 (88.3)	368 (86.2)
Lycoperdales	5	3 (60.0)	1 (20.0)
Nidulariales	2	2 (100.0)	2 (100.0)
Phallales	2	2 (100.0)	2 (100.0)
Hymenogastrales	7	5 (71.4)	5 (71.4)
Sphaerobolales	2	2 (100.0)	2 (100.0)
Ustilaginales	27	27 (100.0)	27 (100.0)
Tremellales	42	42 (100.0)	42 (100.0)
Auriculariales	11	7 (63.6)	6 (54.5)
Dacrymycetales	1	1 (100.0)	1 (100.0)
Anamorphs of Basidiomycotina	18	18 (100.0)	18 (100.0)
Total	939	878 (93.5)	865 (92.1)

*One strain of Pycnoporus coccineus which was included in the previous paper (T. Ito & T. Yokoyama, 1983) was omitted in this experiment.
**Numbers in parentheses show the percentages of the number of survived strains / number of tested strains.

strains, one of Aphyllophorales, nine of Agaricales, two of Lycoperdales, and one of Auriculariales were not recovered even after one year of storage.

On the other hand, all of the strains of Nidulariales, Phallales, Sphaerobolales, Ustilaginales, Tremellales, Dacrymycetales, and the anamorphs of Basidiomycotina were recovered as in the previous test.

It is noted that only one strain out of five of Lycoperdales (20.0%) and six strains out of eleven of Auriculariales (54.5%) were recovered after five years of storage. These fungi, particularily of Lycoperdales,

Table 2. Basidiomycete fungi preserved by freezing at -80 C.

Species	Number of tested strains	Number of survived strains after 1 year	after 5 years
Aphyllophorales			
Aleurodiscus aurantius	1	1	1
A. cerussatus	1	1	1
A. disciformis	1	1	1
A. roseus	1	1	1
Amylostereum areolatum	1	1	1
Aporpium caryae	1	1	1
Auriscalpium vulgare	1	1	1
Bjerkandera adusta	3	3	3
Clavicorona pyxidata	1	1	1
Coniophora puteana	1	1	1
Coriolus brevis	1	1	1
C. consors	7	7	7
C. elongatus	1	1	1
C. fibula	1	1	1
C. hirsutus	10	10	10
C. pargamenus	2	2	2
C. pubescens	1	1	1
C. unicolor	1	1	1
C. versicolor	13	13	13
Corticium caeruleum	1	1	1
C. catonii	2	2	2
C. fuciforme	1	1	1
C. galactinum	2	2	2
C. gramineum	1	1	1
C. lundellii	1	1	0
C. rolfsii	32	32	32
Cryptoderma pini	1	1	1
Cryptoporus volvatus	3	3	3
Cyclomyces fuscus	1	1	1
Cymatoderma elegans	1	1	1
Daedalea dickinsii	5	5	5
D. malicola	1	1	1
Daedaleopsis styracina	2	2	2
D. tricolor	3	3	3
Echinodontium japonicum	2	2	2
E. taxodii	1	1	1
Exobasidium bisporum	2	2	2
E. camelliae	1	1	1
E. gracile	5	5	5
E. japonicum	4	4	4
E. pieridis-ovalifoliae	1	1	1
E. reticulatum	2	2	2
E. shiraianum	1	1	1
E. symploci-japonicae	1	1	1
E. yoshinagai	1	1	1
Exobasidium spp.	5	5	5
Favolus arcularius	4	4	4
Fistulina hepatica	2	2	2
Fomes fomentarius	3	3	3

Table 2. (continued)

Species	Number of tested strains	Number of survived strains after 1 year	after 5 years
Fomitopsis annosa	1	1	1
F. insularis	2	2	2
F. pinicola	1	1	1
Fuscoporia obliqua	1	1	1
Ganoderma applanatum	6	6	6
G. lucidum	3	3	3
Gloeophyllum sepiarium	2	2	2
G. striatum	4	4	4
G. trabeum	3	3	3
G. ungulatum	2	2	2
Grifola frondosa	8	8	8
Gyrodontium versicolor	1	1	1
Hapalopilus croceus	2	2	2
Hericium coralloides	1	1	1
Hirschioporus abietinus	3	3	3
Hymenochaete tabacina	1	1	1
Inonotus dryadeus	1	1	1
I. mikadoi	2	2	2
Irpex lacteus	4	4	4
Laetiporus sulphureus	8	8	8
L. versisporus	4	4	4
Laurilia sulcata	1	1	1
Lenzites betulina	6	6	6
Merulius tremellosus	3	3	3
Mycoleptodonoides pergameneum	3	3	3
Onnia orientalis	1	1	1
Pellicularia filamentosa	17	17	17
P. filamentosa f. sp. microsclerotia	2	2	2
P. filamentosa f. sp. sasakii	31	31	31
P. filamentosa f. sp. solani	12	12	12
P. filamentosa f. sp. timsii	1	1	1
P. flavescens	1	1	1
P. praticola	2	2	2
Peniophora mutata	1	1	1
P. pubera	1	1	1
Phaeolus schweinitzii	3	3	3
Phellinus linteus	1	1	1
P. pomaceus	1	1	1
Pistillaria micans	1	1	1
P. setipes	1	1	1
Polyporellus brumalis	1	1	1
P. picipes	2	2	2
Polyporus sulphureus	1	1	1
Poria aurantiofibrillosus	1	1	1
P. cocos	2	2	2
Porodisculus pendulus	1	1	1

Table 2. (continued)

Species	Number of tested strains	Number of survived strains after 1 year	after 5 years
Protodaedalea hispida	2	2	2
Punctularia atropurpurascens	5	5	5
Pycnoporus cinnabarinus	4	4	4
P. coccineus	15*	15	15
Ramaria botrytis	1	0	0
R. flaccida	1	0	0
Serpula lacrymans	4	4	4
Spongiporus sinuosus	1	1	1
Stereum annosum	1	1	1
S. bicolor	1	1	1
S. frustulosum	3	3	3
S. hirsutum	1	1	1
S. roseum	2	2	2
S. spectabile	1	1	1
S. subpileatum	1	1	1
S. taxodii	1	1	1
Thanatephorus cucumeris	30	29	29
Trametes albida	5	5	5
T. cubensis	1	1	1
T. gibbosa	2	2	2
T. kusanoana	1	1	1
T. orientalis	3	3	3
T. serialis	1	1	1
Trechispora raduloides	1	1	1
Tyromyces caesius	1	1	1
T. palustris	5	5	5
T. ptychogaster	1	1	1
Veluticeps angularis	2	2	2
Agaricales			
Agaricus bisporus	5	5	5
A. campestris	3	3	3
Agrocybe cylindracea	2	2	2
A. praecox	2	2	2
Amanita aspera	1	1	1
A. citrina	3	2	1
A. muscaria	1	0	0
A. pantherina	1	0	0
A. rubescens	1	0	0
A. spissa	1	0	0
A. spissacea	1	1	1
Amanita sp.	1	1	1
Armillariella mellea	5	5	5
A. tabescens	2	2	2
Clitocybe acromelalga	1	1	1
C. clavipes	1	0	0
C. nebularis	1	0	0
Clitocybula sp.	1	1	1
Collybia butyracea	1	1	1
C. confluens	1	1	1
C. cookei	2	2	2

Table 2. (continued)

Species	Number of tested strains	Number of survived strains after 1 year	after 5 years
Collybia peronata	1	1	1
C. tuberosa	1	1	1
Collybia sp.	1	1	1
Conocybe lactea	1	1	1
C. tenera	1	0	0
Coprinus angulatus	1	1	1
C. atramentarius	1	1	0
C. bilanatus	1	1	1
C. cinereus	6	6	6
C. cinereus f. sp. microsporus	3	3	3
C. comatus	2	2	2
C. disseminatus	3	3	3
C. echinosporus	3	1	2
C. filamentifer	1	0	0
C. fissolanatus	1	1	0
C. friesii	1	1	1
C. lagopides	1	1	1
C. lagopus	4	3	3
C. macrocephalus	1	1	1
C. neolagopus	1	1	1
C. ochraceo-velatus	1	1	1
C. phlyctidosporus	3	3	3
C. pseudolagopus	1	0	1
C. radians	2	2	2
C. radiatus	1	0	0
C. rhizophorus	1	1	1
C. stercorarius	2	1	1
Coprinus spp.	3	2	2
Cortinarius cinnamomeus	1	1	1
Crinipellis stipitaria	1	1	1
Filoboletus manipularis	1	1	1
Flammulina velutipes	24	24	24
Galerina fasciculata	1	1	1
Gymnopilus aeruginosus	3	3	3
G. spectabilis	2	2	2
Hebeloma crustuliniforme f. sp. microspermum	1	1	1
H. radicosum	2	2	2
H. spoliatum	4	2	2
H. vinosophyllum	3	3	3
Hygrophoropsis aurantiaca	1	1	0
Kuehneromyces mutabilis	1	1	1
Laccaria laccata	1	1	1
L. proxima	2	1	1
Lactarius chrysorheus	1	1	1
Lampteromyces japonicus	8	8	8
Lentinus edodes	28	27	24
L. lepideus	7	7	7
Lepiota bresadolae	2	2	2

Table 2. (continued)

Species	Number of tested strains	Number of survived strains after 1 year	after 5 years
Lepista irina	1	1	1
L. luscina	1	1	1
L. nuda	7	5	5
L. personata	1	1	1
L. sordida	4	4	3
Leucoagaricus excoriatus	1	1	1
L. naucinus	3	2	1
Leucocoprinus birnbaumii	1	1	1
L. luteus	7	7	6
Lyophyllum anthracophilum	2	2	2
L. decastes	2	2	2
L. fumosum	1	1	1
L. gibberosum	2	2	2
L. leucopaxilloides	1	1	1
L. shimeji	1	0	1
L. transforme	2	1	1
L. tylicolor	4	4	4
L. ulmarium	3	3	3
Macrolepiota mastoidea	1	0	1
M. procera	1	1	1
M. rhacodes	2	2	2
Marasmius purpureostriatus	1	1	1
M. siccus	1	1	1
Mycena crocata	2	2	2
M. haematopoda	1	0	1
M. luteopallens	1	1	1
Naematoloma fasciculare	3	3	3
N. sublateritium	4	4	4
Omphalotus olearius	2	2	2
Oudemansiella mucida	1	1	1
O. radicata	2	2	2
Panaeolina rhombisperma	2	2	2
Panaeolus cambodginiensis	1	1	1
P. sphinctrinus	1	1	1
Panellus serotinus	3	3	3
P. stypticus	3	3	3
Panus conchatus	2	2	2
P. rudis	3	3	3
Phaeolepiota aurea	5	0	0
Pholiota adiposa	3	3	3
P. aurivella	3	3	3
P. carbonaria	3	3	3
P. lenta	3	3	3
P. lubrica	1	1	1
P. nameko	4	4	4
P. spumosa	1	1	1
P. terrestris	2	2	2
Pleurocybella lignatilis	1	1	1
P. porrigens	2	2	2
Pleurotus cornucopiae	2	2	2

Table 2. (continued)

Species	Number of tested strains	Number of survived strains after 1 year	after 5 years
Pleurotus cystidiosus	16	16	14
P. ostreatus	12	12	12
P. sajor-caju	2	2	0
P. salmoneo-stramineus	2	1	1
Psathyrella candolleana	3	2	2
P. velutina	3	2	2
Pseudohiatula ohshimae	1	1	1
Psilocybe argentipes	2	2	2
P. cubensis	2	2	2
P. cyanescens	1	1	1
P. fasciata	5	5	5
P. merdaria	1	1	1
P. subaeruginascens	2	2	2
P. subcaerulipes	1	1	1
Rhodophyllus hirtipes	1	0	0
Schizophyllum commune	9	9	9
Strobilurus stephanocystis	3	2	2
S. tenacellus	1	1	1
Stropharia aeruginosa	1	1	1
S. rugosoannulata	3	3	3
Suillus tomentosus	1	0	0
Tricholoma bakamatsutake	2	2	2
T. fulvocastaneum	11	9	9
T. matsutake	26	20	22
T. ponderosum	1	1	1
T. robustum	4	4	4
Tricholoma sp.	12	10	9
Volvariella speciosa var. *gloiocephala*	1	0	0
V. volvacea	2	2	1
Volvariella sp.	1	0	0
Xeromphalina cauticinalis	2	2	2
Xerula pudens	2	2	2
Lycoperdales			
Calvatia craniiformis	4	3	1
Pisolithus tinctorius	1	0	0
Nidulariales			
Cyathus stercoreus	1	1	1
C. striatus	1	1	1
Phallales			
Kobayasia nipponica	2	2	2
Hymenogastrales			
Limnoperdon incarnatum	7	5	5
Sphaerobolales			
Sphaerobolus stellatus	2	2	2
Ustilaginales			
Doassansia horiana	2	2	2
Graphiola phoenicis	3	3	3
Leucosporidium scottii	2	2	2
Neovossia danubialis	1	1	1

Table 2. (continued)

Species	Number of tested strains	Number of survived strains after 1 year	after 5 years
Tilletia caries	1	1	1
Tilletiaria anomala	1	1	1
Ustilago antherarum	1	1	1
U. cynodontis	3	3	3
U. esculenta	1	1	1
U. kusanoi	1	1	1
U. maydis	2	2	2
U. nuda	1	1	1
U. onumae	1	1	1
U. rabenhorstiana	1	1	1
U. shiraiana	2	2	2
U. violacea	4	4	4
Tremellales			
Fibulobasidium inconspicum	4	4	4
Pseudohydnum gelatinosum	1	1	1
Sporidiobolus johnsonii	2	2	2
Tremella aurantia	2	2	2
T. brasiliensis	4	4	4
T. encephala	5	5	5
T. foliacea	6	6	6
T. fuciformis	3	3	3
T. mesenterica	11	11	11
T. samoensis	2	2	2
T. subanomala	2	2	2
Auriculariales			
Auricularia auricula-judae	4	4	3
A. mesenterica	1	0	0
A. polytricha	1	1	1
Auricularia sp.	1	0	0
Helicobasidium mompa	3	1	1
Helicobasidium sp.	1	1	1
Dacrymycetales			
Femsjonia luteo-alba	1	1	1
Anamorphs of Basidiomycotina			
Ptychogaster corruscans	1	1	1
P. cubensis	1	1	1
Rhizoctonia solani	15	15	15
Sporotrichum dimorphosporum	1	1	1
Total	939	878	865

* One strain of *Pycnoporus coccineus* which was included in the previous paper (T. Ito & T. Yokoyama, 1983) was omitted in this experiment.

are considered to be very sensitive to cryopreservation at -80 C. Except for those mentioned above, most of the basidiomycete fungi are resistant and can grow vigorously even after five years of storage at -80 C.

Table 2 shows the number of tested strains and the number of survived strains after one year and five years of storage at -80 C, respectively, for each species.

Among a total of 395 strains of the species in Aphyllophorales tested, only four strains did not survived. These include two strains found in two separate species of Ramaria and one in thirty strains of Thanatephorus cucumeris (all three were not recovered after one year of storage). The fourth was one strain of Corticium lundellii, which had been recovered after one year of storage. All the strains of the other genera could be recovered after five years of storage.

The results support the fact that, with minor exceptions, these fungal taxa are considered to be resistant to the freezing process and can be preserved satisfactorily by the cryopreservation method.

In Agaricales, no strain of Clitocybe clavipes, C. nebularis, Conocybe tenera, Coprinus atramentarius, C. filamentifer, C. fissolanatus, C. radiatus, Hygrophoropsis aurantiaca, Phaeolepiota aurea, Pleurotus sajor-caju, Rhodophyllus hirtipes, Suillus tomentosus, Volvariella speciosa var. gloiocephala, and Volvariella sp. was alive. It was confirmed that none of the five strains of Phaeolepiota aurea, which had not been recovered after one year of storage, could be recovered again after five years. Some strains of the Amanita, Lepista, and Tricholoma species, which were not viable after one year of storage, did not survive again in this test. In addition to these strains, one strain each of Amanita citrina and Tricholoma sp., which had survived after one year, did not survive after five years of storage.

Contrary to their viability after one year of storage, one strain each of Coprinus atramentarius, C. fissolanatus, Hygrophoropsis aurantiaca, Lepista sordida, Leucoagaricus naucinus, Leucocoprinus luteus, and Volvariella volvacea, two strains each of Pleurotus cystidiosus and P. sajor-caju, and three strains each of Lentinus edodes, were not viable after five years of storage.

As has already been noted in the previous paper (4), it is quite uncertain why strains of these fungi did not survive. However, it is

known that the growth of these strains in the preculture stage was generally slow or very poor even under the best known culture conditions.

On the other hand, a fluctuation in the viabilities of the frozen cultures occurred occasionally in some species and strains. For instance, one strain each of Coprinus echinosporus, C. pseudolagopus, Lyophyllum shimeji, Macrolepiota mastoidea, and Mycena haematopoda, which were not viable after one year of storage, survived in this test. Of the six strains of Tricholoma matsutake, which were not viable before, two strains were found alive in this test. However, the reason for these fluctuations in viabilities is still unclarified.

In Lycoperdales, only one out of four strains of Calvatia craniiformis survived in this test, though three of them were alive in the previous test. Pisolithus tinctorius again failed to grow after five years of storage. Species in this order seems to be sensitive to cryopreservation.

Two species of Cyathus in Nidulariales and Kobayasia nipponica in Phallales were again found to survive for five years of storage.

Among the seven strains of Limnoperdon incarnatum in Hymenogastrales, five were still alive as was shown in the previous paper (4).

Two strains of Sphaerobolus stellatus in Sphaerobolales, twenty-seven strains among sixteen species in Ustilaginales, and forty-two strains among eleven species of Tremellales were all alive. It is clear that these fungi are very resistant to the freezing process.

In Auriculariales, species of Auricularia and Helicobasidium were found to be variable in their viabilities after freezing. However, it is difficult to conclude whether these fungi are basically sensitive to freezing or not, because the number of the strains tested was insufficient to evaluate their sensitivities in detail.

All the strains in Dacrymycetales and in the anamorphs of Basidiomycotina survived indicating these fungi were very resistant to the freezing process. Particularly, it is noted that all fifteen strains of Rhizoctonia solani, an anamorph of Thanatephorus cucumeris, survived well even after five years of storage at -80 C.

Thus, all of 94 strains of Nidulariales, Phallales, Sphaerobolales, Ustilaginales, Tremellales, Dacrymycetales, and the anamorphs of Basidiomycotina, particularly of Rhizoctonia solani, were found very

resistant to cryopreservation and were able to be preserved by freezing at -80 C for five years.

It is concluded that a total of 865 strains of the Basidiomycotina species among the 939 strains tested (92.1%) could be preserved for five years in an electric refrigerator at -80 C without loss of activity. It can be stressed again that cryopreservation at -80 C is considered to be the most effective and practical method for long-term preservation of nonsporulating fungal strains such as those of the Basidiomycotina.

References

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IFO Res. Comm. 13,
82, 1987 (March)

CORRECTION OF A FUNGUS NAME LISTED IN THE PREVIOUS PAPER

TADAYOSHI ITO and TATSUO YOKOYAMA

In our previous paper "Filamentous fungi collected in the Far Eastern USSR" by T. Ito and T. Yokoyama, which appeared in the March 1985 issue of IFO Research Communications, No. 12, pp. 34-62, isolate R-1529-4 was listed under the name of Mortierella longicollis (loc. cit., p. 46).

After publication of the paper, Dr. W. Gams, Centraalbureau voor Schimmelcultures, Baarn (CBS), the Netherlands, kindly suggested that the isolate seemed not to be M. longicollis, but probably M. ramanniana var. angulispora, as was our isolate R-1465-6 and others which had also been recorded in the list. Accordingly, we compared the isolate R-1529-4 with a progeny of the culture CBS 209.32 derived from the type of M. longicollis and concluded that our fungus is not M. longicollis.

After careful reexamination of the isolate R-1529-4 and comparison with the strains of M. ramanniana var. angulispora (IFO 5426, IFO 6744, IFO 8186, IFO 8187, and USSR isolates R-1465-6 and others), together with Naumov's description [Naumov, N.A. (1935). Clés des Mucorinées. Transl. by Buchet, S. & Mouraviev, I. (1939). Encycl. mycol. p. 34, Paris], we also concluded that the isolate R-1529-4 should be accommodated in M. ramanniana var. angulispora.

We thank Dr. W. Gams, CBS, the Netherlands, for his kind suggestion on the identity of our isolate and for supplying CBS isolates to make it possible for us to compare the Mortierella species concerned.

IFO Res. Comm. 13,
83-85, 1987 (March)

DESCRIPTIVE CATALOGUE OF IFO FUNGUS COLLECTION X.

In routine identification work on fungi newly isolated in Japan, and in checking the list of the fungi preserved in the IFO culture collection for published records of their occurrence in Japan, many fungi have been found to be taxa either new to Japan or obscurely or insufficiently described. In some cases, the first record of a fungus in Japan gives only the name of a taxon, without an adequate description of the species concerned. The object of this series is to provide descriptions of the fungi preserved or newly deposited in the IFO culture collection and/or in the IFO herbarium and to contribute to the knowledge of the fungal flora of Japan.

New taxa will be published in other papers. The authors of the descriptions of these fungal taxa are shown in parentheses.

84. Pseudogymnoascus roseus Raillo (Figs. 1-5) Gymnoascales Zentbl. Bakt. ParasitKde (Abt. 2) 78: 520 (1929); Samson, Acta Bot. Neerl. 21: 517 (1972); Sigler and Carmichael, Mycotaxon 4: 349 (1976); Orr, Mycotaxon 8: 165 (1979); Yokoyama et al., IFO Res. Comm. 9: 46 (1979); Tsuneda, Fungal Morphology and Ecology, p. 68 (1982); Ito and Yokoyama, IFO Res. Comm. 12: 34 (1985); Currah, Mycotaxon 24: 1 (1985).
Syn. Gymnoascus roseus (Raillo) Apinis, Mycol. Pap. 96: 8 (1964).
Gymnoascus rhousiogongylinus Wener & Cain, Can. J. Bot. 48: 325 (1970).
Status anamorphosis: Geomyces pannorum (Link) Sigler & Carmichael var. vinaceus (Dal Vesco) van Oorschot, Studies in Mycology 20: 72 (1980).

Colonies on oatmeal agar grow somewhat restrictedly at 20 C, reaching a diameter of 25-30 mm after two weeks, floccose at the center, thin and immersed at the margin, white at first, soon becoming pinkish brown; reverse yellowish red to purplish brown. Ascomata globose to subglobose, yellow brown to red brown, 60-250 μm diam. Peridial hyphae

thick-walled, 2-3.5 μm width, solid, consisting of a network of H-shaped hyphae, with hyaline to pale yellow, short, thin-walled, slightly rough-walled appendages. Asci subglobose to oval, hyaline, evanescent, with a short stalk, 8-spored, 6-10 x 5-7 μm. Ascospores ellipsoid to fusiform, one-celled, pale yellow to pink, smooth, 3-4 x 2-2.5 μm. Geomyces anamorph usually present. Conidia of aleuroconidium-type, ellipsoid to obovoid, hyaline, with a truncate base, 2.5-3 x 2-2.5 μm.

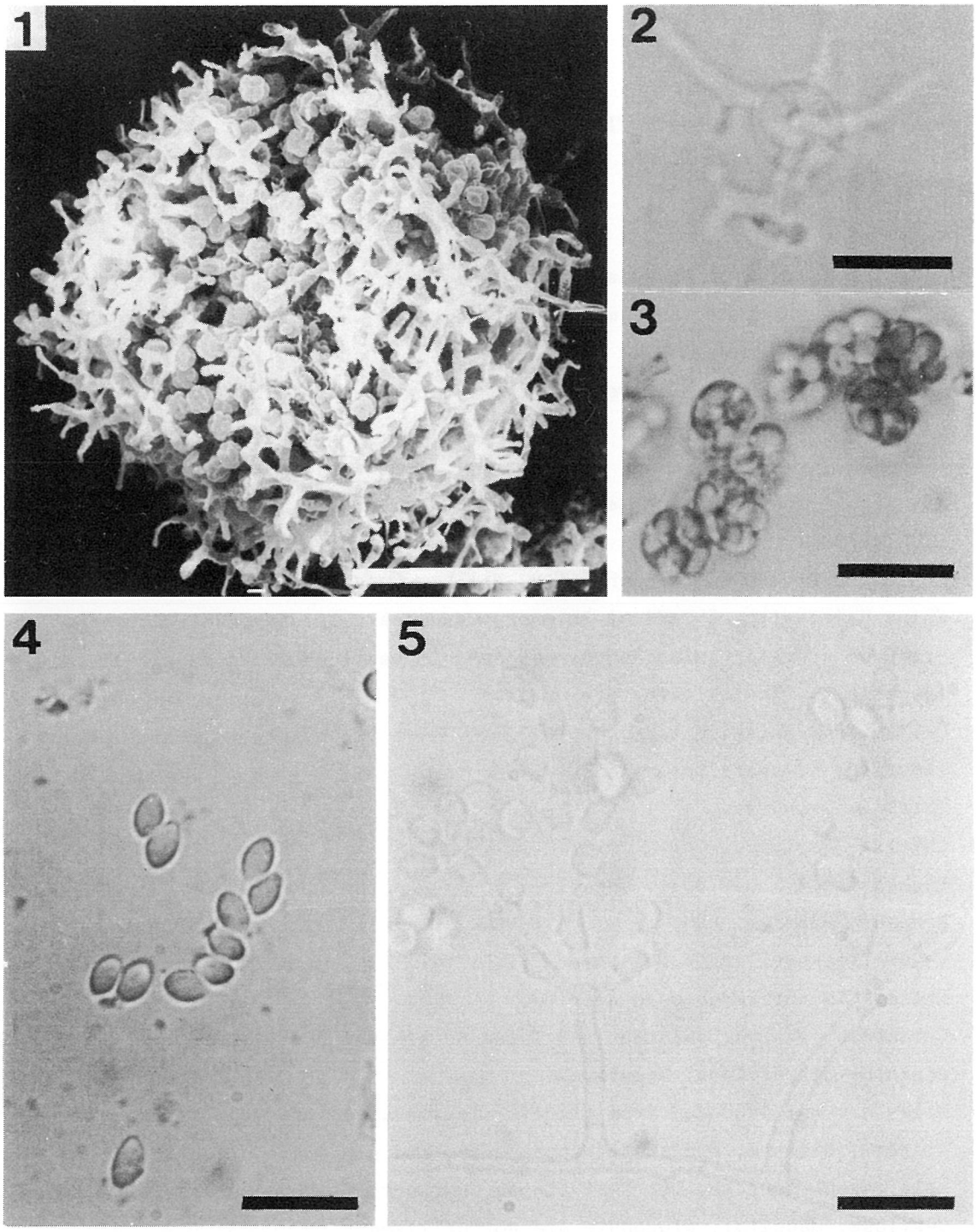
Growth is nil at 37 C.

Hab. paddy field soil, Hachioji, Ikeda, Osaka Pref., Aug. 5, 1977, T. Yokoyama YV-4-5-17; Shakudo, Habikino, Osaka Pref., Nov. 20, 1978, T. Yokoyama XX-2-5-13 (IFO 31815); Urea treated soil, Sugadaira, Chiisagata-gun, Nagano Pref., June 10, 1983, T. Ito S58-9-7.
Additional strains examined: IFO 7639 (As Gymnoascus reessii), IFO 31790 (CBS 320.62), IFO 31791 (CBS 395.65), IFO 31792 (CBS 722.69), IFO 31899.

This fungus differs from other species of the genus by its H-shaped hyphal network with a simple appendage at the apex and fusiform ascospore, and also by having Geomyces anamorph.

The present species has been found mainly in soils in cold areas, particularly as a predominant fungus in alpine or cold temperate forest soils. The geological distribution and ecology of the fungus are well known (Samson, 1972; Sigler and Carmichael, 1976; Orr, 1979; Yokoyama, et al., 1979; Ito and Yokoyama, 1985). It can grow well at 8 C and often forms ascomata at 15-20 C.

(T. Ito & T. Yokoyama)



Figs. 1-5. Pseudogymnoascus roseus (IFO 31815). 1. Ascomata. 2. Ascomatal initial. 3. Asci. 4. Ascospores. 5. Conidial stractures of Geomyces anamorph. Bars for figs. 1. 50 µm; 2-5. 10 µm.

IFO Res. Comm. 13,
86-87, 1987 (March)

DESCRIPTIVE CATALOGUE OF IFO BACTERIAL COLLECTION VIII.

The purpose of this catalogue is to describe the taxonomic properties of strains which had been misclassified or of which uncertainty remained about the proper taxonomic niche, but have since been reclassified in routine reidentification work on the IFO bacterial collection.

73. Alcaligenes faecalis Castellani and Chalmers

IFO 14479

This strain was brought by M. Nozaki, Department of Biochemistry, Shiga University of Medical Science, Otsu, as a pseudomonad which produced aromatic amine dehydrogense.* It was identified as Alcaligenes faecalis by the following properties:

Cells: Gram-negative rods, 0.5-0.7 x 0.7-1.8 μm; motile by peritrichous flagella; no spore formation.

Catalase: Positive.

Oxidase: Positive.

Hugh-Leifson's O-F test: Negative.

Urease: Negative.

Voges-Proskauel test: Negative.

Nitrate is not reduced to nitrite.

Cellulose, starch, gelatin, and Tween 80 are not hydrolyzed.

Arginine dihydrolase: Negative.

Acids are not produced from glucose, maltose, sucrose, and xylose.

Acetate, citrate, succinate, asparagine, and glutamate are utilized as sole carbon sources, but L-arabinose, D-fructose, D-galactose, D-glucose D-xylose, mannitol, gluconate are not utilized.

Vitamins and amino acids are not required for the growth.

Growth temperature: Can grow between 15 and 37 C, but not at 45 C.

(T. Sakane and K. Imai)

* Iwaki, M., T. Yagi, K. Horiike, Y. Saeki, T. Ushijima, and M. Nozaki. 1983. Arch. Biochem. Biophys. 220: 253-262.

IFO Res. Comm.13,
88-89, 1987 (March)

DESCRIPTIVE CATALOGUE OF IFO ACTINOMYCETES COLLECTION I.

The object of this catalogue is to provide descriptions of taxonomical characteristics of the actinomycetes strains which have been identified as different species.

1. Actinoplanes kinshanensis (Ruan & Zhang) Ruan & Zhang in Jiang et al.
Juan, C.-s. and Y.-m. Zhang. 1974. Acta Microbiologica Sinica 14: 31-41; Nonomura, H. et al. 1979. Hakkokogaku Kaishi 57: 79-85 (in Japanese); Jiang, C.-l. et al. 1983. Acta Microbiologica Sinica 23: 210-215.

IFO 13661

This strain was deposited under the name of Ampullariella kinshanensis Juan & Zhang. Under the scanning electron microscope, sporangia were seen to be not bottle-shaped, which is characteristic of Ampullariella, but spherical, which is characteristic of Actinoplanes. The same observation has been reported by Nonomura. The difference between these genera depends on the shape of the sporangium, and this strain should therefore be classified into Actinoplanes. The new scientific name follows that proposed by Jiang et al., 1983.

(T. Kusaka & I. Asano)

2. Nocardia capreola (Higgens) Pridham
Shirling, E.B. and D. Gottlieb. 1968. Int. J. Syst. Bacteriol. 18: 279-392; Pridham, T.G. and A.J. Lyons, Jr. 1969. Devlpmt. Indus. Microbiol. 10: 183-221; Pridham, T.G. 1970. U.S. Dept. Agr. Tech. Bull. 1424: 1-55.

IFO 12847

This strain was obtained from SAJ (Society for Actinomycetes, Japan) as one of the ISP (International Streptomyces Project) strains under the

name of Streptomyces capreolus ISP 5225. It contained meso-A_2pm in its whole cell hydrolysates. Consequently, it is not a species of Streptomyces and is best classified as a Nocardia at this time. The new scientific name follows that proposed by Pridham, 1970.

(T. Kusaka & I. Asano)

3. Nocardioides flavus Ruan & Zhang

Ruan, J.-s. and Y.-m. Zhang. 1979. Acta Microbiologica Sinica 19: 347-352.

IFO 14031, IFO 14032

These two strains were deposited by Ruan Ji-sheng, the Institute of Microbiology, Academia Sinica, Beijing, China under the strain numbers 71-N54 and 71-N82, respectively. Species of this genus contain LL-A_2pm in their whole cell hydrolysates; but both of the deposited strains contained meso-A_2pm. Therefore these strains are not as they were originally described. They were deleted from the collection, and two appropriate strains were deposited under the accession number of IFO 14396 and IFO 14397 by the same depositor, who had recognized the discrepancy.

(T. Kusaka & I. Asano)

4. Nocardioides fulvus Ruan & Zhang

Ruan, J.-s. and Y.-m. Zhang. 1979. Acta Microbiologica Sinica 19: 347-352.

IFO 14033, IFO 14034

These two strains were deposited by Ruan Ji-sheng, the Institute of Microbiology, Academia Sinica, Beijing, China under the strain numbers of 71-N86 and 65-N86, respectively. Species of this genus contain LL-A_2pm in their whole cell hydrolysates, but both of the deposited strains contanied meso-A_2pm. Therfore these strains are not as they were originally described. They were deleted from the collection, and two appropriate strains were deposited under the accession number of IFO 14398 and IFO 14399 by the same depositor, who had recognized the discrepancy.

(T. Kusaka & I. Asano)

ANNOUNCEMENTS

CATALOGUE OF NEWLY ACCEPTED STRAINS

Oct. 1984 - Oct. 1986

(NUMERICAL)

The culture involved in this catalogue can be distributed under the same condition as strains listed in IFO LIST OF CULTURES 7TH EDITION 1984.

10138 Arthroascus javanensis
HISTORY: IFO (K. Mikata; My-3n9; flower of Tricyrtia flava).
10142 Debaryomyces hansenii
HISTORY: IFO (K. Mikata and T. Sakane; F-Y-1; ham).
10144 Saccharomyces cerevisiae
HISTORY: IFO (Y. Kaneko; AH22) -- OUT (Y. Oshima; AH22).
10147 Saccharomyces cerevisiae
HISTORY: IFO (Y. Kaneko; SHY1) -- OUT (Y. Oshima; SHY1).
10148 Saccharomyces cerevisiae
HISTORY: IFO (Y. Kaneko; SHY2) -- OUT (Y. Oshima; SHY2).
10149 Saccharomyces cerevisiae
HISTORY: IFO (Y. Kaneko; SHY3) -- OUT (Y. Oshima; SHY3).
10150 Saccharomyces cerevisiae
HISTORY: IFO (Y. Kaneko; SHY4) -- OUT (Y. Oshima; SHY4).
10151 Saccharomyces cerevisiae
HISTORY: IFO (Y. Kaneko; YNN140) -- OUT (Y. Oshima; YNN140).
10153 Saccharomyces cerevisiae
HISTORY: OUT (Y. Oshima; P-28-24C, monospore culture from hybrid between H-42 & P-22-14D.
10158 Saccharomyces cerevisiae
HISTORY: IAM 12242 -- CCY 48-78 -- Inst. Biol. of Czechoslovak Academy of Science, Prague No. 00323/1 -- Canadian Breweries, Tronto.
10159 Saccharomyces cerevisiae
HISTORY: Fac. Sci., Yamaguchi Univ. (N. Sando; FE-1; flux of Fagus sp.).
10160 Saccharomyces cerevisiae
HISTORY: H. Tamaki, 5114-9A, a segregant of hybrid between S. diastaticus & genetic stock mutant.
10161 Saccharomyces cerevisiae
HISTORY: H. Tamaki, 5114-11B, a segregant of hybrid between S. diastaticus & genetic stock mutant.
10162 Saccharomyces cerevisiae
HISTORY: H. Tamaki, 5209-6A, a segregant of hybrid between S. diastaticus & genetic stock mutant.
10163 Saccharomyces cerevisiae
HISTORY: H. Tamaki, 5209-11B, a segregant of hybrid between S. diastaticus & genetic stock mutant.
10164 Saccharomyces cerevisiae
HISTORY: H. Tamaki, 5311-3C, a segregant of hybrid between S. diastaticus & genetic stock mutant.
10165 Saccharomyces cerevisiae
HISTORY: H. Tamaki, 5311-1A, a segregant of hybrid between S. diastaticus & genetic stock mutant.
10166 Bullera oryzae
HISTORY: JCM 5281 (T. Nakase; NO-65; dead stem of Oryza sativa).
10167 Candida tsuchiyae
HISTORY: JCM 1638 -- AJ 4911 (T. Nakase; moss).
10168 Sporobolomyces subbrunneus
HISTORY: JCM 5278 (T. Nakase; NO-14; dead stem of Oryza sativa).
10173 Saccharomyces cerevisiae
HISTORY: OUT (Y. Kaneko; AL101-1D).
10174 Saccharomyces cerevisiae

HISTORY: OUT (Y. Kaneko; AL101-2A).
10175 Saccharomyces cerevisiae
HISTORY: OUT (Y. Kaneko; AL211-12B).
10176 Saccharomyces cerevisiae
HISTORY: OUT (Y. Kaneko; AL211-2B).
10177 Bullera derxii
HISTORY: JCM 5280 (T. Nakase; NO-92; dead leaf of Oryza sativa).
10178 Bullera intermedia
HISTORY: JCM 5291 (T. Nakase; NO-157; dead leaf of Oryza sativa).
10179 Bullera pseudoalba
HISTORY: JCM 5290 (T. Nakase; NO-165; dead leaf of Oryza sativa).
10180 Sporobolomyces oryzicola
HISTORY: JCM 5299 (T. Nakase; NO-12; dead leaf of Oryza sativa).
10181 Saccharomyces exiguus
HISTORY: IFO (K. Mikata; Yp74L-3; partially decayed leaf).
10182 Candida antarctica
HISTORY: Dept. Agr. Chem., Shizuoka Univ. (Y. Yamada) -- CBS 6821 -- W. Henninger, aphid secretion on Solanum pseudocapsicum.
10183 Candida steatolytica
HISTORY: Dept. Agr. Chem., Shizuoka Univ. (Y. Yamada) -- CBS 4028.
10184 Candida steatolytica
HISTORY: Dept. Agr. Chem., Shizuoka Univ. (Y. Yamada) -- CBS 5839 -- J.P. van der Walt, bovine udder.
10185 Candida steatolytica
HISTORY: Dept. Agr. Chem., Shizuoka Univ. (Y. Yamada) -- CBS 6360.
10186 Candida steatolytica
HISTORY: Dept. Agr. Chem., Shizuoka Univ. (Y. Yamada) -- CBS 6726 -- A.M. van Grinsven, drink factory.
10187 Candida steatolytica
HISTORY: Dept. Agr. Chem., Shizuoka Univ. (Y. Yamada) -- CBS 6779 -- H. Scheler, frozen orange juice.
10188 Sporopachydermia cereana
HISTORY: Dept. Agr. Chem., Shizuoka Univ. (Y. Yamada) -- CBS 6645 -- UCD 73-67 (H.J. Phaff; Cereus sp.).
10189 Sporopachydermia lactativola
HISTORY: Dept. Agr. Chem., Shizuoka Univ. (Y. Yamada) -- CBS 5772 -- J.W. Fell, seawater.
10190 Sporopachydermia lactativola
HISTORY: Dept. Agr. Chem., Shizuoka Univ. (Y. Yamada) -- CBS 6058 -- D.G. Ahearn, waste lagoon of asphalt plant.
10191 Sporopachydermia lactativola
HISTORY: Dept. Agr. Chem., Shizuoka Univ. (Y. Yamada) -- CBS 6192 -- A. Kahanpaa, human mouth.
10192 Stephanoascus ciferrii
HISTORY: Dept. Agr. Chem., Shizuoka Univ. (Y. Yamada) -- CBS 4856 -- A.H. Klokke, cow neck.
10193 Stephanoascus ciferrii
HISTORY: Dept. Agr. Chem., Shizuoka Univ. (Y. Yamada) -- CBS 5165 -- A.H. Klokke, wood in cow-shed.
10194 Stephanoascus ciferrii
HISTORY: Dept. Agr. Chem., Shizuoka Univ. (Y. Yamada) -- CBS 5166 -- H. Seeliger.
10195 Stephanoascus ciferrii
HISTORY: Dept. Agr. Chem., Shizuoka Univ. (Y. Yamada) -- CBS 5646 -- Cent. Vet. Lab., Weybridge, bovine placenta.
10196 Sterigmatomyces elviae
HISTORY: Dept. Agr. Chem., Shizuoka Univ. (Y. Yamada) -- CBS 8119 -- D. Swinne, air in bakery.
10197 Sterigmatomyces elviae
HISTORY: Dept. Agr. Chem., Shizuoka Univ. (Y. Yamada) -- CBS 8121 --

D. Swinne, flour.
10198 Sterigmatomyces halophilus
HISTORY: Dept. Agr. Chem., Shizuoka Univ. (Y. Yamada) -- CBS 5449 -- C. da Silva Lacaz, keloid blastomycosis.
10199 Sterigmatomyces halophilus
HISTORY: Dept. Agr. Chem., Shizuoka Univ. (Y. Yamada) -- CBS 5632 -- J.W. Fell, seawater.
10200 Sterigmatomyces halophilus
HISTORY: Dept. Agr. Chem., Shizuoka Univ. (Y. Yamada) -- CBS 6780 (W. Gams; soil).
10201 Sterigmatomyces indicus
HISTORY: Dept. Agr. Chem., Shizuoka Univ. (Y. Yamada) -- CBS 5629 -- J.W. Fell, seawater.
10202 Sterigmatomyces indicus
HISTORY: Dept. Agr. Chem., Shizuoka Univ. (Y. Yamada) -- CBS 5631 -- J.W. Fell, seawater.
10203 Sterigmatomyces polyborus
HISTORY: Dept. Agr. Chem., Shizuoka Univ. (Y. Yamada) -- CBS 6643 -- J. Nicot, ambre gris.
10205 Sterigmatomyces tursiopsis
HISTORY: Dept. Agr. Chem., Shizuoka Univ. (Y. Yamada) -- CBS 6133 -- D.G. Ahearn.
10212 Schwanniomyces castellii
HISTORY: H. Tamaki, 1840-5.
10213 Hansenula anomala
HISTORY: CBS 5759 -- NRRL Y-366 -- F.W. Fabian.
10214 Pichia euphorbiae
HISTORY: CBS 8033 -- J.P. van der Walt, cross of CBS 7082 arg and CBS 7083 ade.
10215 Pichia membranaefaciens
HISTORY: CBS 107 -- N.H. Claussen.
10216 Pichia nakasei
HISTORY: CBS 5141 -- J. Grinbergs, apple must.
10217 Saccharomyces cerevisiae
HISTORY: CBS 1171 -- A.C. van Wijk.
10218 Torulaspora pretoriensis
HISTORY: CBS 2187 -- J.P. van der Walt, soil.
14318 Frankia sp.
HISTORY: IMRU, LLR 01321 (M.P. Lechevalier; AirI1; nodules of Alnus incana ssp. rugosa).
14319 Frankia sp.
HISTORY: IMRU, LLR 01322 (M.P. Lechevalier; AirI2; nodules of Alnus incana ssp. rugosa).
14396 Nocardioides flavus
HISTORY: IMAS (J.S. Ruan; 71-N54; soil).
14397 Nocardioides flavus
HISTORY: IMAS (J.S. Ruan; 71-N82; soil).
14398 Nocardioides fulvus
HISTORY: IMAS (J.S. Ruan; 71-N86; soil).
14399 Nocardioides fulvus
HISTORY: IMAS (J.S. Ruan; 65-N86; soil).
14400 Acholeplasma laidlawii
HISTORY: ATCC 23206 -- R. Wittler -- D.G. ff Edward -- Nat. Inst. Med. Res., London -- P. P. Laidlaw & W.J. Elford.
14401 Mycoplasma pneumoniae
HISTORY: ATCC 15531 -- L. Hayflick.
14402 Nocardia brasiliensis
HISTORY: ATCC 19296 -- IMRU 8451 (R.E. Gordon) -- J. Schneidas, Jr., 381 -- A. Batista, 631 -- Pasteur Inst., 337.
14403 Nocardia carnea

HISTORY: ATCC 6847 -- NCTC 3527 -- V. Puntoni, 30.
14404 Nocardia corynebacteroides
HISTORY: ATCC 14898 -- A.J. Crowle, air.
14405 Nocardia otitidiscarviarum
HISTORY: ATCC 14629 -- R.E. Gordon -- NCTC 1934 -- E.P. Snijders, ear of guinea pig.
14406 Nocardia petroleophila
HISTORY: ATCC 15777 -- P. Hirsch, 102, soil.
14410 Escherichia coli
HISTORY: IFO (T. Iijima; F1600) -- Otsuji -- Matsushiro, C600.
14411 Bacillus subtilis
HISTORY: IFO (K. Imai) -- BGSC 1A3.
14412 Bacillus subtilis
HISTORY: IFO (K. Imai) -- BGSC 1A4.
14413 Bacillus subtilis
HISTORY: IFO (K. Imai) -- BGSC 1A5.
14414 Bacillus subtilis
HISTORY: IFO (K. Imai) -- BGSC 1A6.
14415 Bacillus subtilis
HISTORY: IFO (K. Imai) -- BGSC 1A7.
14416 Bacillus subtilis
HISTORY: IFO (K. Imai) -- BGSC 1A8.
14417 Bacillus subtilis
HISTORY: IFO (K. Imai) -- BGSC 1A9.
14418 Bacillus subtilis
HISTORY: IFO (K. Imai) -- BGSC 1A10.
14419 Bacillus subtilis
HISTORY: IFO (K. Imai) -- BGSC 1A11.
14423 Nocardiopsis dassonvillei subsp. prasina
HISTORY: NIAES (K. Miyashita; 208; soil).
14424 Actinomadura ochracea
HISTORY: IMAS (Y.X. Deng; 71-a175; mud).
14426 Nocardia salmonicolor subsp. aurantiaca
HISTORY: IMAS (J.S. Ruan; 71-N45; soil).
14427 Nocardia violaceofusca
HISTORY: IMAS (J.S. Ruan; 78-N26; soil).
14428 Amorphosporangium castaneum
HISTORY: IMAS (Z.O. Jiang; B133; soil).
14436 Thermoanaerobacter cellulolyticus
HISTORY: Dep. Chem. Engineering, Nagoya Univ. (M. Taya; NA10; Hot spring sediments in Nozawa).
14438 Klebsiella pneumoniae
HISTORY: RTCI (T. Nishi; No.1) -- Osaka Univ., Medical School.
14439 Klebsiella pneumoniae
HISTORY: RTCI (T. Nishi; No. 9) -- Osaka Univ., Medical School.
14440 Klebsiella pneumoniae
HISTORY: RTCI (T. Nishi; No. 45) -- Osaka Univ., Medical School.
14441 Klebsiella pneumoniae
HISTORY: RTCI (T. Nishi; No. 47) -- Osaka Univ., Medical School.
14442 Klebsiella pneumoniae
HISTORY: RTCI (T. Nishi; No. 57) -- Osaka City Univ., Medical School.
14443 Klebsiella pneumoniae
HISTORY: RTCI (T. Nishi; No. 69) -- Tohoku Univ., School of Medicine.
14444 Saccharothrix australiensis
HISTORY: NRRL 11239 -- American Cyanamid Co. (D.P. Labeda; LL-BM782Ce82; soil).
14447 Nocardiopsis antarcticus
HISTORY: INMI K-4042 (S.S. Abyzov; 25-145; ice).
14455 Actinoalloteichus cyanogriseus
HISTORY: AS 4.1159 -- IMAS (Z.H. Liu; Y388; soil).

14457 Streptomyces purpeofuscus
HISTORY: ATCC 21470 -- IMC M890-C2, soil.
14458 Actinoplanes violaceus
HISTORY: IMAS -- Yunnan Inst. Microbiol. (C.L. Jiang; Y80-610; soil).
14459 Actinoplanes yunnanensis
HISTORY: IMAS -- Yunnan Inst. Microbiol. (C.L. Jiang; Y79-21; soil).
14460 Ampullariella kunmingensis
HISTORY: IMAS -- Yunnan Inst. Microbiol. (C.L. Jiang; Y79-15; soil).
14461 Micromonospora yulongensis
HISTORY: IMAS -- Yunnan Inst. Microbiol. (C.L. Jiang; Y81-917; soil).
14462 Staphylococcus aureus
HISTORY: ATCC 25923.
14463 Chainia kunmingensis
HISTORY: IMRU -- AS (J.S. Ruan; 80-3024; soil).
14471 Microbacterium laevaniformans
HISTORY: NCIB 9659 -- J.V. Bhat.
14475 Nocardiopsis mutabilis subsp. cryophilis
HISTORY: IMC (Y. Okami; TS-1980; soil).
14476 Mycoplasma arginini
HISTORY: Univ. Tokyo, Fac. Med. (K. Yamamoto; G230) -- NIH, Maryland, USA.
14477 Mycoplasma orale
HISTORY: Univ. Tokyo, Fac. Med. (K. Yamamoto; CH19299) -- NIH, Maryland, USA.
14478 Mycoplasma salivarium
HISTORY: Univ. Tokyo, Fac. Med. (K. Yamamoto; PG20) -- NIH, Maryland, USA.
14479 Alcaligenes faecalis
HISTORY: Dept. Biochem., Shiga University of Medical Science (M. Nozaki; aromatic amine dehydrogenase; soil).
14480 Excellospora viridilutea
HISTORY: VKM (G.K. Skryabin) -- INMI 187.
14485 Excellospora japonica
HISTORY: IAM (K. Furihata; 345-AT_3; soil).
14486 Excellospora japonica
HISTORY: IAM (K. Furihata; 1077-MT_1; soil).
14487 Glycomyces harbinensis
HISTORY: NRRL 15337 -- Lederle Lab. (D.P. Labeda; LL-DO5139; soil).
14488 Glycomyces rutgersensis
HISTORY: NRRL B-16106 -- IMRU (M.P. Lechevalier; LL-I-20; soil).
14491 Nocardioides luteus
HISTORY: IMET 7830 (H. Prauser; soil).
14493 Kibdelosporangium aridum
HISTORY: ATCC 39323 -- Smith Kline & French Lab. (M.C. Shearer; SK&F-AAD-216; soil).
14498 Amycolata hydrocarbonoxydans
HISTORY: NRRL B-16171 -- IMRU -- P. Hirsch, 70(N42), air.
14499 Amycolata saturnea
HISTORY: NRRL B-16172 -- IMRU 1181 -- P. Hirsch, 71, air.
14500 Amycolatopsis orientalis subsp. lurida
HISTORY: NRRL 2430 -- Abbott Labs. (M307; soil).
14506 Amycolatopsis rugosa
HISTORY: ATCC 43014 -- M.P. Lechevalier -- R.E. Gordon -- C. diMarco, rumen.
14507 Streptomyces ipomoeae
HISTORY: NIAES (T. Suzui; A698; sweet potato).
14508 Streptomyces ipomoeae
HISTORY: NIAES (T. Suzui; A699; sweet potato).
14511 Lactobacillus fermentum
HISTORY: JCM 5867 -- Inst. Phys. Chem. Res. (K. Suzuki; IIb T-46).
14512 Lactobacillus fermentum
HISTORY: JCM 5868 -- Inst. Phys. Chem. Res. (K. Suzuki; IIb 4061).
14513 Lactobacillus fermentum
HISTORY: JCM 5869 -- Inst. Phys. Chem. Res. (K. Suzuki; F-5).

14520 Nocardiopsis coeruleofusca
HISTORY: INA 1335, soil.
14521 Nocardiopsis flava
HISTORY: INA 2171 (G.F. Gauze; soil).
14522 Nocardiopsis longispora
HISTORY: INA 10222, soil.
14523 Nocardiopsis syringae
HISTORY: INA 2240 (G.F. Gauze; soil).
14524 Actinoplanes ianthinogenes subsp. octamycini
HISTORY: INA 4041 (G.F. Gauze).
14525 Actinomadura recticatena
HISTORY: INA 308 (G.F. Gauze; soil).
14526 Streptomyces coeruleoaurantiacus
HISTORY: INA 4009 (G.F. Gauze; soil).
14528 Actinosporangium coccineum
HISTORY: Yunnan Inst. Microbiol. (D.F. Jiang; JH 2622; soil).
14531 Rhodococcus globerulus
HISTORY: Univ. Newcastle (M. Goodfellow; R58) -- ATCC 25714 -- R.E. Gordon, 544 -- E.N. Azarowicz -- W.C. Haynes.
20061 Bacillus subtilis phage PBS1
HISTORY: IFO (K. Imai) -- BGSC 1P1.
20062 Bacillus pumilus phage Ø 31
HISTORY: IFO (K. Imai; 31).
20063 Escherichia coli phage Ø170 vir
HISTORY: IFO (T. Iijima).
31638 Aspergillus niger PP 56-K-1136
HISTORY: IFO (T. Yokoyama; Pak 14-5; decayed leaf of Eucalyptus sp.).
31639 Aspergillus versicolor PP 56-K-1136
HISTORY: IFO (T. Yokoyama; Pak 21-20; decayed leaf of Populus sp.).
31640 Discosia artocreas PP 56-K-1136
HISTORY: IFO (T. Yokoyama; Pak 7-2; decayed leaf).
31641 Drechslera australiensis PP 56-K-1136
HISTORY: IFO (T. Yokoyama; Pak 9-4; decayed leaf).
31642 Drechslera australiensis PP 56-K-1136
HISTORY: IFO (T. Yokoyama; Pak 24-1; decayed leaf).
31643 Lasiodiplodia theobromae PP 56-K-1136
HISTORY: IFO (T. Yokoyama; Pak 5-9; decayed leaf).
31644 Memnoniella echinata PP 56-K-1136
HISTORY: IFO (T. Yokoyama; Pak 5-6; decayed leaf).
31645 Penicillium thomii PP 56-K-1136
HISTORY: IFO (T. Yokoyama; Pak 2-6; decayed leaf).
31646 Stachybotrys chartarum PP 56-K-1136
HISTORY: IFO (T. Yokoyama; Pak 17-3; decayed leaf).
31647 Trichothecium roseum PP 56-K-1136
HISTORY: IFO (T. Yokoyama; Pak 21-1; decayed leaf of Populus sp.).
31648 Bartalinia robillardoides PP 56-K-1075
HISTORY: IFO (T. Yokoyama; SA 17-1; decayed leaf of Podocarpus sp.).
31649 Memnoniella echinata PP 56-K-1075
HISTORY: IFO (T. Yokoyama; SA 1-10; decayed leaf of Ficus sp.).
31650 Stachybotrys chartarum PP 56-K-1075
HISTORY: IFO (T. Yokoyama; SA 5-1; decayed leaf of Ficus sp.).
31651 Helicobasidium mompa
HISTORY: Nat. Inst. Agro-Env. Sci. (T. Suzui; H-19; root of Asparagus officinalis).
31652 Bondarzewia montana
HISTORY: SU (K. Yokoyama; 4211; rotten wood of Abies firma).
31653 Panus lacomtei
HISTORY: SU (K. Yokoyama; 4382; blight twig).
31654 Conocybe fragilis
HISTORY: SU (T. Horikoshi; 75; ground).

31655 Itersonilia pastinacae
HISTORY: Fac. Agr., Shizuoka Univ. -- CBS 356.64 -- Nat. Veg. Res. St. (A.G. Channon; leaf of Pastinaca sativa).
31656 Itersonilia pyriformis
HISTORY: Fac. Agr., Shizuoka Univ. -- CBS 286.50 -- G. Nyland, dead leaf of Acer macrophyllum.
31657 Fusarium decemcellulare
HISTORY: IFO (T. Yokoyama; 1; trunk of Ficus carica).
31658 Fusarium decemcellulare
HISTORY: IFO (T. Yokoyama; 2; trunk of Ficus carica).
31659 Fusarium decemcellulare
HISTORY: IFO (T. Yokoyama; A; trunk of Ficus carica).
31660 Fusarium decemcellulare
HISTORY: IFO (T. Yokoyama; B; trunk of Ficus carica).
31661 Periconia atropurpurea
HISTORY: IFO (T. Yokoyama; SA 4-3; decayed leaf of Eucalyptus sp.).
31662 Periconia atropurpurea
HISTORY: IFO (T. Yokoyama; SA 21-3; decayed leaf of Compositae plant).
31663 Myrothecium indicum
HISTORY: IFO (T. Yokoyama; SA 19'-3; decayed leaf of Myoporum sp.).
31664 Aspergillus terreus
HISTORY: IFO (T. Yokoyama; Pak 14-3; decayed leaf of Eucalyptus sp.).
31665 Fusariella concinna
HISTORY: IFO (T. Yokoyama; Pak 5-5; decayed leaf).
31666 Emericella variecolor
HISTORY: IFO (T. Yokoyama; Pak 21-9; decayed leaf of Populus sp.).
31667 Hamigera avellanea var. alba
HISTORY: HUT 4181 (T. Morinaga; soil).
31668 Tolypocladium niveum
HISTORY: CBS 824.70 (W. Gams; subalpine raw humus).
31669 Tolypocladium niveum
HISTORY: CBS 714.70 -- W.B. Cooke, AM5-11-4C, sandy soil.
31670 Tolypocladium niveum
HISTORY: CBS 715.70 -- W.B. Cooke, PR2-7-3.
31671 Tolypocladium niveum
HISTORY: CBS 716.70 -- E. Müller-Kögler, E391/64, Aradus cinnamomeus.
31672 Ustilaginoidea virens
HISTORY: Fac. Agr., Hirosaki Univ. (Y. Harada; Harada 450; grain of Oryza sativa).
31673 Ustilaginoidea virens
HISTORY: Fac. Agr., Hirosaki Univ. (Y. Harada; Harada 451; grain of Oryza sativa).
31674 Acremonium terricola
HISTORY: IFO (T. Ito; T. Yokoyama WXI-5-5-25; paddy field soil).
31675 Aspergillus terreus
HISTORY: IFO (T. Ito; T. Yokoyama XXI42-1-5-1; paddy field soil).
31676 Beauveria bassiana
HISTORY: IFO (T. Ito; T. Yokoyama WXI-1-5-21; paddy field soil).
31677 Cladosporium cladosporioides
HISTORY: IFO (T. Ito; T. Yokoyama WXI-1-5-20; paddy field soil).
31678 Cladosporium sphaerospermum
HISTORY: IFO (T. Ito; T. Yokoyama XXI70-1-15-1; paddy field soil).
31679 Dichotomomyces cejpii var. cejpii
HISTORY: IFO (T. Ito; T. Yokoyama WXI70-1-5-1; paddy field soil).
31680 Dichotomomyces cejpii var. spinosus
HISTORY: IFO (T. Ito; T. Yokoyama WXIE-1-5-3; paddy field soil)
31681 Gliocladium catenulatum
HISTORY: IFO (T. Ito; T. Yokoyama WXI-1-5-6; paddy field soil).
31682 Metarhizium anisopliae
HISTORY: IFO (T. Ito; T. Yokoyama WXI-1-5-14; paddy field soil).

31683 Neosartorya fischeri var. glabra
HISTORY: IFO (T. Ito; T. Yokoyama WXI42-3-10-3; paddy field soil).
31684 Neosartorya fischeri var. spinosa
HISTORY: IFO (T. Ito; T. Yokoyama WXI70-1-10-3; paddy field soil).
31685 Paecilomyces variotii
HISTORY: IFO (T. Ito; T. Yokoyama XXI42-4-5-3; paddy field soil).
31686 Phialocephala humicola
HISTORY: IFO (T. Ito; T. Yokoyama WXI-1-10-28; paddy field soil).
31687 Pithomyces chartarum
HISTORY: IFO (T. Ito; T. Yokoyama WXI-5-5-23; paddy field soil).
31688 Scopulariopsis brevicaulis
HISTORY: IFO (T. Ito; T. Yokoyama XXI-4-15-24; paddy field soil).
31689 Talaromyces flavus var. macrosporus
HISTORY: IFO (T. Ito; T. Yokoyama XXIE-2-10-3; paddy field soil).
31690 Talaromyces helicus var. helicus
HISTORY: IFO (T. Ito; T. Yokoyama XXIE-4-5-5; paddy field soil).
31691 Talaromyces ucrainicus
HISTORY: IFO (T. Ito; T. Yokoyama XXIE-5-15-4; paddy field soil).
31692 Talaromyces wortmannii
HISTORY: IFO (T. Ito; T. Yokoyama WXIE-3-10-3; paddy field soil).
31693 Thermoascus aurantiacus
HISTORY: IFO (T. Ito; T. Yokoyama WXI42-1-5-2; paddy field soil).
31694 Trichoderma aureoviride
HISTORY: IFO (T. Ito; T. Yokoyama WXI-5-5-3; paddy field soil).
31695 Westerdykella dispersa
HISTORY: IFO (T. Ito; T. Yokoyama WXI-2-5-7; paddy field soil).
31696 Amorphotheca resinae
HISTORY: ATCC 22065 -- D.G. Parbery, 35B, soil at base of creosoted pole.
31697 Amorphotheca resinae
HISTORY: ATCC 32945 -- J.E. Sheridan, 52/73, jet fuel.
31698 Arachniotus ruber
HISTORY: ATCC 16945 -- G.F. Orr, O-925, as Arachniotus ruber -- H. Kuehn -- H. Bohme -- K.H. Domsch, Domsch C925, wheat-field soil.
31699 Arthroderma curreyi
HISTORY: ATCC 13550 -- CBS 138.26 -- A. Nannizzi.
31700 Arthroderma curreyi
HISTORY: ATCC 26339 -- J.W. Carmichael, UAMH 3065, chicken feathers.
31701 Auxarthron pseudauxarthron
HISTORY: ATCC 22158 -- G.F. Orr, O-3083, domectic rabbit dung.
31702 Auxarthron reticulatum
HISTORY: ATCC 18426 -- G.F. Orr, O-1020, wood slat from greenhouse flat.
31703 Auxarthron thaxteri
HISTORY: ATCC 15598 -- G.F. Orr, O-532 -- H.H. Kuehn, dung.
31704 Auxarthron umbrinum
HISTORY: ATCC 15604 -- G.F. Orr, O-513 -- D.W. Emmons, E-4950 (CDC A-959).
31705 Auxarthron umbrinum
HISTORY: ATCC 15605 -- G.F. Orr, O-1023 -- H.H. Warcup, A-212/4, soil from ploughed field.
31706 Cladosporium resinae f. resinae
HISTORY: ATCC 11841 -- D.H. Marsden, as Hormodendrum resinae -- C.H. Christensen -- H. von Schrenk, creosoted wooden pole.
31707 Cladosporium resinae f. resinae
HISTORY: ATCC 11873 -- D.H. Marsden, Enola, creosoted wooden pole. As Hormodendrum resinae.
31708 Cladosporium resinae f. albidum
HISTORY: ATCC 34012 -- S. Goto, 564, plant detritus.
31709 Cladosporium resinae f. avellaneum
HISTORY: ATCC 11273 -- CBS 186.54 (G.A. de Vries, ointment).
31710 Cladosporium resinae f. avellaneum

HISTORY: ATCC 22711 -- J.J. Cooney, UD-42 -- P. Edmonds, JP-4, jet fuel.
31711 Ctenomyces serratus
HISTORY: ATCC 15502 -- G.F. Orr, SL466 -- H.S. Randhawa, soil.
31712 Ctenomyces serratus
HISTORY: ATCC 15537 -- E. Varsavsky, O-3, soil.
31713 Ctenomyces serratus
HISTORY: ATCC 15536 -- E. Varsavsky, O-2, soil.
31714 Geomyces asperulatus
HISTORY: ATCC 34521 -- J.W. Carmichael, UAMH 183 -- F. Raymond, II-1-9, soil.
31715 Gymnoascus uncinatus
HISTORY: ATCC 16006 -- G.F. Orr, Orr I-26 -- E. Varsavsky, soil.
31716 Gymnoascus dugwayensis
HISTORY: ATCC 18899 -- G.F. Orr, O-3138 (DPG-132), soil.
31717 Myxotrichum chartarum
HISTORY: ATCC 18432 -- G.F. Orr, O-3171 -- IMI 189072a -- J. Webster, corrugated cardboard.
31718 Myxotrichum deflexum
HISTORY: ATCC 15686 -- G.F. Orr, O-249, soil.
31719 Myxotrichum deflexum
HISTORY: ATCC 15690 -- G.F. Orr, O-245, soil.
31720 Myxotrichum stipitatum
HISTORY: ATCC 15684 -- G.F. Orr, O-518 -- J.H. Warcup, soil.
31721 Myxotrichum stipitatum
HISTORY: ATCC 15685 -- G.F. Orr, O-521, as Gymnoascus setosus -- CBS -- J.H. Warcup, soil.
31722 Pseudogymnoascus vinaceus
HISTORY: ATCC 28807 -- G.F. Orr, O-1119, as Pseudogymnoascus bhattii -- J. Bissett, JB 95, alpine soil.
31723 Anixiopsis fulvescens var. fulvescens
HISTORY: ATCC 36140 -- Z. Hubalek, 331 E, feathers of Anas platyrhynchos.
31724 Xylogone sphaerospora
HISTORY: ATCC 42027 -- R.A. Taber.
31726 Amorphotheca resinae
HISTORY: IMI 129862 -- D.G. Parbery, V2, alluvial soil.
31727 Arachniotus ruber
HISTORY: IMI 92796 -- J. Cox, A 157, soil.
31728 Eupenicillium alutaceum
HISTORY: IMI 136243 -- CBS 317.67 -- CSIR 1039 (De B. Scott; garden soil).
31729 Eupenicillium anatolicum
HISTORY: IMI 136242 (M. Oener; 37; soil).
31730 Eupenicillium brefeldianum
HISTORY: IMI 216895 -- FRR 71 (J.I. Pitt; soil).
31731 Eupenicillium brefeldianum
HISTORY: IMI 216896 -- FRR 710 -- NRRL 710 -- Thom 5296 -- B.O. Dodge, human alimentary tract.
31732 Eupenicillium cinnamopurpureum
HISTORY: IMI 114483 -- M.N. Gupta, unrecorded source.
31733 Eupenicillium crustaceum
HISTORY: IMI 34911 ii -- LSHB -- NRRL 939 -- Thom 4885 -- L. McCulloch, Gladiolus sp. corm.
31734 Eupenicillium erubescens
HISTORY: IMI 136204 -- CBS 318.67 -- CSIR 1040 (De B. Scott; nursery soil).
31735 Eupenicillium javanicum
HISTORY: IMI 39733 -- LSHB -- NRRL 707 -- F.H. van Beyma, Camellia sinensis roots.
31736 Eupenicillium lassenii
HISTORY: IMI 148395 -- J.W. Paden, JWP 69-26, soil of mixed conifer stand.
31737 Eupenicillium meridianum
HISTORY: IMI 136209 -- CBS 314.67 -- CSIR 1052 (De B. Scott; grassland soil).
31738 Eupenicillium molle

HISTORY: IMI 84589 -- TRTC 45714 -- S.A. Lohdi, WY 29, soil.
31739 Eupenicillium ornatum
HISTORY: IMI 137977 -- NHL 6101 (S. Udagawa; soil)
31740 Eupenicillium tularense
HISTORY: IMI 148394 -- J.W. Paden, JWP 68-31, soil under Pinus ponderosa and Quercus kelloggii.
31741 Eupenicillium zonatum
HISTORY: IMI 216907 -- FRR 1550 -- C.S. Hodges, Jr. -- J.J. Perry, coastal salt marsh soil.
31742 Hapsidospora irregularis
HISTORY: IMI 148377, -- TRTC 44852 (W.F. Williams; lawn grass compost heap).
31743 Nigrosabulum globosum
HISTORY: IMI 148372 -- TRTC 43288 (R.F. Cain; cow dung)
31744 Penicillium chermesinum
HISTORY: IMI 166620 -- FRR 3387 (J.I. Pitt) -- D.K. & R.S. Sandhu, Pe 1602, sputum of man.
31745 Penicillium chermesinum
HISTORY: IMI 191730 -- FRR 2048 -- NRRL 2048 -- W.H. Weston, deteriorating military equipment.
31746 Penicillium donkii
HISTORY: IMI 197489 -- CBS 188.72 -- L.K. Oliver, arable soil.
31747 Penicillium herquei
HISTORY: IMI 28809 -- NCTC 1721 -- Thom 4640.477 -- Da Fonseca -- G. Bainier Collection -- Herqué, Agauria pyrifolia leaf.
31748 Penicillium novae-zeelandiae
HISTORY: IMI 40584 ii -- FRR 2128 (J.I. Pitt) -- NRRL 2128 -- CBS 137.41 -- F.H. van Beyma -- J. Neill, Sclerotinia sp. apothecium.
31749 Penicillium quercetorum
HISTORY: IMI 140342 -- VKM F-1074 (L.A. Beljakova) -- V.Ch. Baghdadi, T 811, soil.
31750 Talaromyces gossypii
HISTORY: IMI 198365 -- R.B. Somani, Gossypium sp.
31751 Talaromyces helicus var. helicus
HISTORY: IMI 40593 --LSHB (K.B. Raper) -- NRRL 2106 -- H. Edy Velander, soil.
31752 Talaromyces intermedius
HISTORY: IMI 100874 -- BDUN 267 (A.E. Apinis; alluvial pasture and swamp soil.
31753 Talaromyces luteus
HISTORY: IMI 89305 -- LSHB BB.228 -- J.H. Warcup, soil.
31754 Talaromyces mimosinus
HISTORY: IMI 223991 -- FRR 1875 (J.I. Pitt) -- A.D. Hocking, soil of creek bank.
31755 Talaromyces purpureus
HISTORY: IMI 181546 -- CBS 475.71 -- E. Müller -- C. Stoll, soil.
31756 Talaromyces rotundus
HISTORY: IMI 40589 -- LSHB -- NRRL 2107 -- G.W. Martin, 6271, wood.
31757 Talaromyces trachyspermus
HISTORY: IMI 40043 -- LSHB -- NRRL 1028 -- C.W. Emmons.
31758 Talaromyces ucrainicus
HISTORY: IMI 129962 -- VKM F-907 -- Ukrainian Food Res. Inst., Kharkiv -- V.T. Panasenko.
31759 Amylocarpus encephaloides
HISTORY: CBS 128.60 -- J. Kohlmeyer, wood.
31760 Amylocarpus encephaloides
HISTORY: CBS 129.60 -- J. Kohlmeyer, wood.
31761 Anixiopsis fulvescens var. stercoraria
HISTORY: CBS 111.58 -- R.F. Cain, bear dung.
31762 Anixiopsis fulvescens var. stercoraria
HISTORY: CBS 121.64 (G.A. de Vries) -- T. Pinto Robeiro, soil.
31763 Arachniotus aureus
HISTORY: CBS 593.71 -- G.F. Orr, O-2512 -- K. Tubaki, decayed wood.

31764 Arachniotus ruber
HISTORY: CBS 194.64 -- G.F. Orr -- J. Cox, A 157, soil.
31765 Arachniotus ruber
HISTORY: CBS 112.69 -- K.H. Domsch, wheat-field soil.
31766 Arachniotus ruber
HISTORY: CBS 592.71 - J.H. van Emden, field soil.
31767 Byssochlamys fulva
HISTORY: CBS 146.48 -- K.B. Raper -- T. Rendle, bottled fruit.
31768 Byssochlamys fulva
HISTORY: CBS 604.71
31769 Byssoascus striatisporus
HISTORY: CBS 642.66 -- G.L. Barron, soil.
31770 Cladosporium resinae f. avellaneum
HISTORY: CBS 186.54 (G.A. de Vries; cosmetic face cream "Nivea").
31771 Cladosporium resinae f. avellaneum
HISTORY: CBS 176.62 (oil aircraft tank).
31772 Cladosporium resinae f. resinae
HISTORY: CBS 185.54.
31773 Cladosporium resinae f. resinae
HISTORY: CBS 187.54 (G.A. de Vries, cosmetic face cream "Nivea").
31774 Eupenicillium catenatum
HISTORY: CBS 352.67 -- CSIR 1097 (De B. Scott; desert soil).
31775 Geomyces asperulatus
HISTORY: CBS 628.79 -- G.J. Bollen, meadow soil.
31776 Geomyces pannorum var. pannorum
HISTORY: CBS 105.13 -- M. Jensen, potato field soil.
31777 Geomyces pannorum var. pannorum
HISTORY: CBS 108.14 -- A.E. Traaen, soil.
31778 Geomyces pannorum var. pannorum
HISTORY: CBS 378.76 -- ATCC 11501 -- H.A. Dade, refrigerated meat.
31779 Gymnoascus demonbreunii
HISTORY: CBS 121.67 -- L. Ajello, B646, human.
31780 Gymnoascus demonbreunii
HISTORY: CBS 122.67 -- L. Ajello, B767, soil beneath starling roost.
31781 Keratinophyton terreum
HISTORY: CBS 342.64 -- H.S. Randhava, SL 289, lawn soil.
31782 Roumegueriella rufula
HISTORY: CBS 276.59 -- S. Stribling, Axis deer dung.
31783 Nigrosabulum globosum
HISTORY: CBS 512.70 -- TRTC 43288 (R.F. Cain; cow dung).
31784 Onygena corvina
HISTORY: CBS 225.60 -- NI 2168 (K. Tubaki; man).
31785 Onygena equina
HISTORY: CBS 947.70 (cow hoof).
31786 Onygena piligena
HISTORY: CBS 298.49 -- R. Heim, wollen slipper.
31787 Petalosporus afilamentosus
HISTORY: CBS 658.71 -- G.F. Orr, O-2601 (DPG-115), clay soil.
31788 Petalosporus anodosus
HISTORY: CBS 518.68 -- G.F. Orr, O-580, rabbit dung.
31789 Petalosporus nodosus
HISTORY: CBS 577.63 -- G.R. Ghosh, G-2, guinea pig dung.
31790 Pseudogymnoascus roseus
HISTORY: CBS 320.62 -- B. Dal Vesco, T-2, soil.
31791 Pseudogymnoascus roseus
HISTORY: CBS 395.65 -- BDUN 266 (A.E. Apinis; alluvial swamp soil).
31792 Pseudogymnoascus roseus
HISTORY: CBS 722.69 -- D. Malloch -- TRTC 45536, forest soil.
31793 Shanorella spirotricha
HISTORY: CBS 304.56 -- R.K. Benjamin, RSA 64, rabbit dung.

31794 Shanorella spirotricha
HISTORY: CBS 305.56 -- R.K. Benjamin, RSA 156, feather of dead bird.
31795 Spiromastix warcupii
HISTORY: CBS 576.63 -- G.F. Orr, W84, soil.
31796 Talaromyces galapagensis
HISTORY: CBS 751.74 -- D.P. Mahoney, G1032, soil beneath Maytenus obovata.
31797 Talaromyces ohiensis
HISTORY: CBS 162.67 -- NHL 6086 (S. Udagawa; soil).
31798 Talaromyces thermophilus
HISTORY: CBS 236.58 -- NRRL 2155 -- R. Emerson, M206517, retted Parthenium argentatum (D.G. Cooney 26, as Talaromyces dupontii).
31799 Warcupia terrestris
HISTORY: CBS 891.69 -- J. Paden, VSF-66-74, Tolmie sandy loam soil.
31800 Warcupiella spinulosa
HISTORY: CBS 512.65 -- WB 4376 (K.B. Raper & D.I. Fennell) -- J.H. Warcup, A41/4, jungle soil.
31801 Westerdykella dispersa
HISTORY: CBS 297.56 -- F.M. Clum, 27, seedling of Phlox drummondii.
31802 Westerdykella multispora
HISTORY: CBS 391.51 -- K. Kominami -- K. Minoura, crop field soil.
31803 Westerdykella nigra
HISTORY: CBS 192.57 -- J.B. Routien, 17M48, soil.
31804 Westerdykella purpurea
HISTORY: CBS 297.75 (sandy soil).
31805 Alternaria alternata
HISTORY: IFO (T. Ito; T. Yokoyama XXI-1-5-24; paddy field soil).
31806 Aspergillus versicolor
HISTORY: IFO (T. Ito; T. Yokoyama ZXI-3-5-13; paddy field soil).
31807 Chaetomium thermophilum var. dissitum
HISTORY: IFO (T. Ito; T. Yokoyama YII42-1-5-5; paddy field soil).
31808 Eupenicillium parvum
HISTORY: IFO (T. Ito; T. Yokoyama XXIE-5-15-7; paddy field soil).
31809 Eurotium repens
HISTORY: IFO (T. Ito; T. Yokoyama WXE-3-5-3; paddy field soil).
31810 Geotrichum candidum
HISTORY: IFO (T. Ito; T. Yokoyama ZXI-4-10-20; paddy field soil).
31811 Humicola fuscoatra
HISTORY: IFO (T. Ito; T. Yokoyama ZXI-4-10-36; paddy field soil).
31812 Oidiodendron truncatum
HISTORY: IFO (T. Ito; T. Yokoyama ZXI-1-10-16; paddy field soil).
31813 Penicillium janthinellum
HISTORY: IFO (T. Ito; T. Yokoyama WXI-5-10-9; paddy field soil).
31814 Pseudeurotium ovalis
HISTORY: IFO (T. Ito; T. Yokoyama XXI-2-15-15; paddy field soil).
31815 Pseudogymnoascus roseus
HISTORY: IFO (T. Ito; T. Yokoyama XX-2-5-13; paddy field soil).
31816 Stachybotrys bisbyi
HISTORY: IFO (T. Ito; T. Yokoyama XXI-4-5-20; paddy field soil).
31817 Staphylotrichum coccosporum
HISTORY: IFO (T. Ito; T. Yokoyama ZXI-3-5-20; paddy field soil).
31818 Talaromyces udagawae
HISTORY: IFO (T. Ito; T. Yokoyama ZXE-3-10-5; paddy field soil).
31819 Zopfiella leucotricha
HISTORY: IFO (T. Ito; T. Yokoyama XXI-1-15-8; paddy field soil).
31820 Alatosessilispora bibrachiata
HISTORY: TKB C-1337 (K. Tubaki; rainwater from Quercus mongolica var. grosseserrata).
31821 Microstella pluvioriens
HISTORY: TKB C-1342 (K. Tubaki; rainwater from Metasequoia glyptostroboides).
31822 Curucispora ombrogena

HISTORY: TKB C-1340 (K. Tubaki; rainwater from Phyllostachys bambusoides).
31823 Dicranidion fissile
HISTORY: TKB C-1362 (K. Tubaki; rainwater from Quercus myrsinaefolia).
31824 Tricladiella pluvialis
HISTORY: TKB C-1364 (K. Tubaki; rainwater from Quercus acutissima).
31825 Tripospermum infalcatum
HISTORY: TKB C-1366 (K. Tubaki; rainwater from Pinus densiflora).
31826 Arthrobotrys ellipsospora
HISTORY: TKB C-1053 (K. Tubaki; pine sap of Pinus densiflora).
31827 Melanodothis caricis
HISTORY: Fac. Agr., Hirosaki Univ. (Y. Harada; Harada 472; Carex dispalata).
31828 Anixiella reticulata
HISTORY: IFO (T. Ito; T. Yokoyama ZX70-3-5-3; paddy field soil).
31829 Ascodesmis sphaerospora
HISTORY: IFO (T. Ito; T. Yokoyama ZIXE-1-5-4; paddy field soil).
31830 Aspergillus clavatus
HISTORY: IFO (T. Ito; T. Yokoyama ZIX70-4-15-3; paddy field soil).
31831 Botrytis cinerea
HISTORY: IFO (T. Ito; T. Yokoyama XVIII-3-15-11; paddy field soil).
31832 Byssochlamys nivea
HISTORY: IFO (T. Ito; T. Yokoyama XIE-1-15-7; paddy field soil).
31833 Chaetomium aureum
HISTORY: IFO (T. Ito; T. Yokoyama WXI-2-5-15; paddy field soil).
31834 Chaetomium bostrychodes
HISTORY: IFO (T. Ito; T. Yokoyama ZIV-2-5-20; paddy field soil).
31835 Chaetomium funicolum
HISTORY: IFO (T. Ito; T. Yokoyama YI-4-5-11; paddy field soil).
31836 Chaetomium globosum
HISTORY: IFO (T. Ito; T. Yokoyama ZVII-5-5-16; paddy field soil).
31837 Emericella nidulans var. nidulans
HISTORY: IFO (T. Ito; T. Yokoyama YIII42-3-10-1; paddy field soil).
31838 Eupenicillium ornatum
HISTORY: IFO (T. Ito; T. Yokoyama WXI-4-10-8; paddy field soil).
31839 Hamigera avellanea
HISTORY: IFO (T. Ito; T. Yokoyama WX70-5-5-2; paddy field soil).
31840 Malbranchea pulchella var. sulfurea
HISTORY: IFO (T. Ito; T. Yokoyama XVIII42-1-5-1; paddy field soil).
31841 Microascus cinereus
HISTORY: IFO (T. Ito; T. Yokoyama YV-3-5-31; paddy field soil).
31842 Monascus ruber
HISTORY: IFO (T. Ito; T. Yokoyama YIII42-4-10-2; paddy field soil).
31843 Myceliophthora thermophila
HISTORY: IFO (T. Ito; T. Yokoyama WIX42-2-5-4; paddy field soil).
31844 Neosartorya fischeri var. fischeri
HISTORY: IFO (T. Ito; T. Yokoyama ZIX42-1-10-4; paddy field soil).
31845 Neosartorya quadricincta
HISTORY: IFO (T. Ito; T. Yokoyama ZXI42-1-5-3; paddy field soil).
31846 Paecilomyces marquandii
HISTORY: IFO (T. Ito; T. Yokoyama XIII-2-5-9; paddy field soil).
31847 Penicillium lilacinum
HISTORY: IFO (T. Ito; T. Yokoyama YII-3-5-5; paddy field soil).
31848 Penicillium oxalicum
HISTORY: IFO (T. Ito; T. Yokoyama XV-2-5-11; paddy field soil).
31849 Petriellidium boydii
HISTORY: IFO (T. Ito; T. Yokoyama XVIII-2-5-22; paddy field soil).
31850 Podospora carbonaria
HISTORY: IFO (T. Ito; T. Yokoyama ZVII70-2-10-4; paddy field soil).
31851 Pseudeurotium zonatum
HISTORY: IFO (T. Ito; T. Yokoyama XIII-5-10-19; paddy field soil).
31852 Talaromyces emersonii

HISTORY: IFO (T. Ito; T. Yokoyama YV42-4-15-2; paddy field soil).
31853 Thermoascus crustaceus
HISTORY: IFO (T. Ito; T. Yokoyama YV42-1-5-2; paddy field soil).
31854 Thermomyces lanuginosus
HISTORY: IFO (T. Ito; T. Yokoyama WIV42-1-5-3; paddy field soil).
31855 Cylindrocarpon sclerotigenum
HISTORY: Hokkaido Nat. Agr. Exp. St. (N. Matsumoto; 1-4; sclerotium of Typhula incarnata).
31856 Amorphotheca resinae
HISTORY: IMI 129861 -- D.G. Parbery, WI 1, podosolic soil.
31857 Exobasidium hachijoense
HISTORY: Inst. Agr. Forest., Univ. Tsukuba (M. Kakishima; Cinnamomum japonicum).
31858 Mycena chlorophos
HISTORY: IFO (T. Yokoyama; T. Yokoyama 6007-5-1; trunk of Phoenix canariensis).
31859 Pleurotus salmoneo-stramineus
HISTORY: IFO (T. Yokoyama; T. Yokoyama 6006-23-1; decaying trunk of Quercus acutissima).
31860 Tricholoma giganteum
HISTORY: IFO (T. Yokoyama; T. Yokoyama 5909-28-1).
31861 Agaricus sp.
HISTORY: SU (K. Yokoyama; K. Yokoyama 4602; soil in green house).
31862 Flammulina velutipes
HISTORY: SU (K. Yokoyama; K. Yokoyama 4601; rotting wood).
31863 Ganoderma lucidum
HISTORY: SU (K. Yokoyama; K. Yokoyama 4620; rotting stump of Quercus gilva).
31864 Lentinus edodes
HISTORY: SU (K. Yokoyama; K. Yokoyama 2520 (=NZ43); decaying trunk of Nothofagus fusca).
31865 Lentinus edodes
HISTORY: SU (K. Yokoyama) -- Takara Shuzo Co., Ltd. (O. Nakajima; NZ 061; rotting trunk of Nothofagus fusca).
31866 Lentinus edodes
HISTORY: SU (K. Yokoyama) -- Takara Shuzo Co., Ltd. (O. Nakajima; NZ 062; rotting trunk of Nothofagus fusca).
31867 Panaeolus retirugis
HISTORY: SU (K. Yokoyama; K. Yokoyama 4359).
31868 Pholiota limonella
HISTORY: SU (K. Yokoyama; K. Yokoyama 4603) -- Takara Shuzo Co., Ltd. (Y. Kawamoto; rotting trunk of Chosenia arbutifolia).
31869 Psilocybe coprophila
HISTORY: SU (K. Yokoyama; K. Yokoyama 4623).
31870 Psilocybe coprophila
HISTORY: SU (K. Yokoyama; K. Yokoyama 4616; horse dung).
31871 Stropharia rugosoannulata
HISTORY: SU (K. Yokoyama; K. Yokoyama 4625; compost containing horse dung).
31872 Tyromyces albellus
HISTORY: SU (K. Yokoyama; K. Yokoyama 4464).
31873 Cladosporium resinae f. resinae
HISTORY: ATCC 11274 -- CBS (As Cladosporium avellaneum f. viride; ointment).
31876 Penicillium hispanicum
HISTORY: ATCC 38667 -- C. Ramirez, IJFM 3223, Citrus limomum fruit.
31877 Byssochlamys fulva
HISTORY: ATCC 10099 -- USDA, BPI (As C. Thom 5367.6a) -- E. Mrak -- LSHB (pa. 24) -- T. Rendle, bottled fruit.
31878 Byssochlamys fulva
HISTORY: ATCC 24474 -- NRRL 3493 (A.D. King) -- NRRL A-13,158 (C.W. Hesseltine; commercially canned grape juice).
31879 Talaromyces flavus var. flavus

HISTORY: ATCC 9776 -- P.B. Marsh, 1336.1.
31880 Talaromyces luteus
HISTORY: ATCC 10465 -- NRRL 2102 (As Penicillium luteum) -- CBS -- C. Thom 5357.208 -- G.R. Bisby, soil.
31881 Cylindrocarpon destructans f. sp. panacis
HISTORY: SUF 811 (T. Matuo) -- Nagano Veg. & Ornam. Crops Exp. St., Japan (Y. Miyazawa; Panax ginseng).
31882 Cylindrocarpon destructans f. sp. panacis
HISTORY: SUF 859 (T. Matuo) -- Nagano Veg. & Ornam. Crops Exp. St., Japan (Y. Miyazawa; Panax ginseng).
31883 Discosia artocreas
HISTORY: IFO (T. Yokoyama) -- Tokushima Hort. Exp. St. (H. Yamato; Ume 1-2; Prunus mume).
31884 Pestalotiopsis acaciae
HISTORY: IFO (T. Yokoyama; Nagano 5) -- Nagano Pref. Nanshin Agr. Exp. St. (S. Imamura; 5; dry japanese persimon).
31885 Dichotomomyces cejpii var. cejpii
HISTORY: IFO (T. Ito; T. Yokoyama RE-1603-7; soil under Paeonia).
31886 Diplogelasinospora grovesii
HISTORY: IFO (T. Ito; T. Yokoyama RE-1473-2; gold mine soil).
31887 Emericella nidulans var. acristata
HISTORY: IFO (T. Ito; T. Yokoyama R42-1616-4; litter).
31888 Eupenicillium pinetorum
HISTORY: IFO (T. Ito; T. Yokoyama R70-1615-4; humus soil).
31889 Eupenicillium shearii
HISTORY: IFO (T. Ito; T. Yokoyama R70-1538-2; peaty humus of swamp).
31890 Eurotium chevalieri
HISTORY: IFO (T. Ito; T. Yokoyama RE-1603-4; soil under Paeonia).
31891 Eurotium repens
HISTORY: IFO (T. Ito; T. Yokoyama RE-1603-3; soil under Paeonia).
31892 Hamigera avellanea
HISTORY: IFO (T. Ito; T. Yokoyama RE-1602-6; soil under Begonia).
31893 Hamigera striata
HISTORY: IFO (T. Ito; T. Yokoyama RE-1500-2; soil under turnip).
31894 Nectria inventa
HISTORY: IFO (T. Ito; T. Yokoyama R-1602-23; soil under Begonia).
31895 Neosartorya fischeri var. fischeri
HISTORY: IFO (T. Ito; T. Yokoyama RE-1601-1; soil).
31896 Petriella setifera
HISTORY: IFO (T. Ito; T. Yokoyama R-1501-8; soil of green house side).
31897 Pseudeurotium ovalis
HISTORY: IFO (T. Ito; T. Yokoyama RE-1603-18; soil under Paeonia).
31898 Pseudeurotium zonatum
HISTORY: IFO (T. Ito; T. Yokoyama RE-1453-1; road side soil).
31899 Pseudogymnoascus roseus
HISTORY: IFO (T. Ito; T. Yokoyama R70-1484-2; sandy clay).
31900 Talaromyces byssochlamydoides
HISTORY: IFO (T. Ito; T. Yokoyama R42-1499-5; soil of cabbage field).
31901 Talaromyces emersonii
HISTORY: IFO (T. Ito; T. Yokoyama R42-1499-4; soil of cabbage field).
31902 Talaromyces flavus var. flavus
HISTORY: IFO (T. Ito; T. Yokoyama R70-1602-1; soil under Begonia).
31903 Talaromyces helicus var. helicus
HISTORY: IFO (T. Ito; T. Yokoyama RE-1603-14; soil under Paeonia).
31904 Talaromyces leycettanus
HISTORY: IFO (T. Ito; T. Yokoyama RE-1606-3; soil).
31905 Talaromyces luteus
HISTORY: IFO (T. Ito; T. Yokoyama RE-1603-13; soil under Paeonia).
31906 Talaromyces stipitatus
HISTORY: IFO (T. Ito; T. Yokoyama R70-1609-1; soil).

31907 Talaromyces trachyspermus
HISTORY: IFO (T. Ito; T. Yokoyama RE-1601-2; soil).
31908 Talaromyces ucrainicus
HISTORY: IFO (T. Ito; T. Yokoyama R70-1602-2; soil under Begonia).
31909 Talaromyces wortmannii
HISTORY: IFO (T. Ito; T. Yokoyama RE-1603-12; soil under Paeonia).
31910 Thermoascus aurantiacus
HISTORY: IFO (T. Ito; T. Yokoyama R42-1499-3; soil of cabbage field).
31911 Thielavia arenaria
HISTORY: IFO (T. Ito; T. Yokoyama R42-1616-6; forest litter).
31912 Trichophaea abundans
HISTORY: IFO (T. Ito; T. Yokoyama R70-1595-1; humus soil).
31913 Penicillium crustosum
HISTORY: IFO (T. Yokoyama) -- Sericult. Exp. St. (K. Kawakami; 916; silkworm foods).
31914 Penicillium lilacinum
HISTORY: IFO (T. Yokoyama) -- Sericult. Exp. St. (K. Kawakami; 919a; silkworm foods).
31915 Scopulariopsis brevicaulis
HISTORY: IFO (T. Yokoyama; T. Yokoyama 6009-1-8; wall material).
31916 Nectria gliocladioides
HISTORY: IFO (T. Yokoyama; T. Yokoyama 6009-1-3; Glycine max).
31917 Trichoderma aureoviride
HISTORY: IFO (T. Yokoyama; T. Yokoyama 6009-1-7; Glycine max).
31918 Trichoderma longibrachiatum
HISTORY: IFO (T. Yokoyama; T. Yokoyama 6009-1-5; Glycine max).
31919 Trichoderma longibrachiatum
HISTORY: IFO (T. Yokoyama; T. Yokoyama 6009-1-6; Glycine max).
31920 Trichoderma pseudokoningii
HISTORY: IFO (T. Yokoyama; T. Yokoyama 6009-1-4; Glycine max).
31921 Pythium okanoganense
HISTORY: Coll. Agr., Univ. Osaka Pref. UOP 10 -- Fukui Agr. Exp. St. (S. Takamatsu; H-82-69; leaf of Hordeum vulgare).
31922 Pythium okanoganense
HISTORY: Coll. Agr., Univ. Osaka Pref. UOP 14 -- Fukui Agr. Exp. St. (S. Takamatsu; H84209; leaf of Hordeum vulgare).
31923 Pythium vanterpoolii
HISTORY: Coll. Agr., Univ. Osaka Pref. UOP 3 -- Fukui Agr. Exp. St. (S. Takamatsu; H-82-13; leaf of Hordeum vulgare).
31924 Pythium vanterpoolii
HISTORY: Coll. Agr., Univ. Osaka Pref. UOP 4 -- Fukui Agr. Exp. St. (S. Takamatsu; H-82-21; leaf of Hordeum vulgare).
31925 Pythium vanterpoolii
HISTORY: Coll. Agr., Univ. Osaka Pref. UOP 16 -- Fukui Agr. Exp. St. (S. Takamatsu; H-82-18; leaf of Hordeum vulgare).
31926 Pythium volutum
HISTORY: Coll. Agr., Univ. Osaka Pref. UOP 18 -- Fukui Agr. Exp. St. (S. Takamatsu; W-82-59; leaf of Triticum aestivum).
31927 Pythium volutum
HISTORY: Coll. Agr., Univ. Osaka Pref. UOP 5 -- Fukui Agr. Exp. St. (S. Takamatsu; H-82-82; leaf of Hordeum vulgare).
31928 Pythium volutum
HISTORY: Coll. Agr., Univ. Osaka Pref. UOP 39 -- Fukui Agr. Exp. St. (S. Takamatsu; H-82-60; leaf of Hordeum vulgare).
31929 Sclerotinia minor
HISTORY: IFO -- Tochigi Pref. Agr. Exp. St. (S. Saito; stem of Lycopersicon esculentum).
31930 Sclerotinia minor
HISTORY: IFO (T. Yokoyama; T. Yokoyama 6009-1-1; Glycine max).
31931 Sclerotinia minor

HISTORY: IFO (T. Yokoyama; T. Yokoyama 6009-1-2; Glycine max).
31932 Trichoderma aureoviride
HISTORY: IFO (T. Yokoyama; T. Yokoyama 5907-15-1) -- Fac. Agr., Kagawa Univ. (T. Tani; tuber of Ipomoea batatas).
31933 Pythium periplocum
HISTORY: UOP 311 -- Fac. Agr., Kagawa Univ. (T. Tani; P-K_1; Zoysia tenuifolia).
31937 Anellaria semiovata
HISTORY: IFO (T. Yokoyama; T. Yokoyama 6004-20-1; horse dung).
31938 Phallus rugulosus
HISTORY: IFO (T. Yokoyama; T. Yokoyama 6007-13-1).
31939 Collybia iocephala
HISTORY: IFO (T. Yokoyama; T. Yokoyama 5809-23-5; decaying trunk).
31940 Amanita virgineoides
HISTORY: IFO (T. Yokoyama; T. Yokoyama 6007-13-2).
31941 Pythium okanoganense
HISTORY: Coll. Agr., Univ. Osaka Pref. UOP 2 -- Fukui Agr. Exp. St. (S. Takamatsu; H-82-66 ; leaf of Hordeum vulgare).
31942 Pythium sylvaticum
HISTORY: IFO (T. Yokoyama) -- Fac. Agr., Hirosaki Univ. (Y. Harada; Harada 384; tuber of Discorea batatas).
31943 Pythium sylvaticum
HISTORY: IFO (T. Yokoyama) -- Fac. Agr., Hirosaki Univ. (Y. Harada; Harada 388; tuber of Discorea batatas).
31944 Tilletia barclayana
HISTORY: IFO (T. Yokoyama) -- Fac. Agr., Hirosaki Univ. (Y. Harada; Harada 465; Pennisetum alopeculoides).
31945 Tilletia barclayana
HISTORY: IFO (T. Yokoyama) -- Fac. Agr., Hirosaki Univ. (Y. Harada; Harada 468; Pennisetum alopeculoides).
31946 Ustilago sphaerogena
HISTORY: IFO (T. Yokoyama) -- Fac. Agr., Hirosaki Univ. (Y. Harada; Harada 473; Echinochloa crus-galli).
31947 Ustilago crus-galli
HISTORY: IFO (T. Yokoyama) -- Fac. Agr., Hirosaki Univ. (Y. Harada; Harada 486; Echinochloa crus-galli).
31948 Acremonium fusidioides
HISTORY: IFO (T. Ito; T. Yokoyama R70-1623-5; litter).
31949 Acremonium persicinum
HISTORY: IFO (T. Ito; T. Yokoyama R70-1499-1; cabbage field soil).
31950 Arthrinium phaeospermum
HISTORY: IFO (T. Ito; T. Yokoyama R1607-10; soil).
31951 Aspergillus deflectus
HISTORY: IFO (T. Ito; T. Yokoyama R42-1623-4; litter).
31952 Aspergillus fumigatus
HISTORY: IFO (T. Ito; T. Yokoyama R42-1455-1; soil).
31953 Beauveria bassiana
HISTORY: IFO (T. Ito; T. Yokoyama R-1454-8; soil).
31954 Chrysosporium merdarium
HISTORY: IFO (T. Ito; T. Yokoyama RE-1500-4; turnip field soil).
31955 Cladosporium staurophorum
HISTORY: IFO (T. Ito; T. Yokoyama R70-1470-1; soil).
31956 Doratomyces microsporus
HISTORY: IFO (T. Ito; T. Yokoyama R-1500-9; turnip field soil).
31957 Doratomyces nanus
HISTORY: IFO (T. Ito; T. Yokoyama R-1607-21; soil).
31958 Gilmaniella humicola
HISTORY: IFO (T. Ito; T. Yokoyama RE-1602-7; soil under Begonia).
31959 Gliocladium virens
HISTORY: IFO (T. Ito; T. Yokoyama R-1602-2; soil under Begonia).

31960 Harposporium helicoides
HISTORY: IFO (T. Ito; T. Yokoyama R-1506-6; peat).
31961 Metarhizium anisopliae
HISTORY: IFO (T. Ito; T. Yokoyama R-1455-9; soil).
31962 Myceliophthora thermophila
HISTORY: IFO (T. Ito; T. Yokoyama R42-1601-2; soil).
31963 Oidiodendron echinulatum
HISTORY: IFO (T. Ito; T. Yokoyama R-1525-7; humus).
31964 Paecilomyces carneus
HISTORY: IFO (T. Ito; T. Yokoyama R-1473-11; swamp soil).
31965 Paecilomyces inflatus
HISTORY: IFO (T. Ito; T. Yokoyama R-1499-4; cabbage field soil).
31966 Paecilomyces marquandii
HISTORY: IFO (T. Ito; T. Yokoyama R-1622-18; humus).
31967 Paecilomyces variotii
HISTORY: IFO (T. Ito; T. Yokoyama R70-1488-1; peat).
31968 Penicillium funiculosum
HISTORY: IFO (T. Ito; T. Yokoyama R-1471-12; swamp soil).
31969 Penicillium janthinellum
HISTORY: IFO (T. Ito; T. Yokoyama R-1460-5; soil).
31970 Penicillium lilacinum
HISTORY: IFO (T. Ito; T. Yokoyama R-1454-6; soil).
31971 Penicillium nigricans
HISTORY: IFO (T. Ito; T. Yokoyama R-1622-14; humus).
31972 Penicillium piceum
HISTORY: IFO (T. Ito; T. Yokoyama R42-1611-1; litter).
31973 Phialophora alba
HISTORY: IFO (T. Ito; T. Yokoyama R-1463-17; soil).
31974 Scolecobasidium humicola
HISTORY: IFO (T. Ito; T. Yokoyama R-1454-15; soil).
31975 Tolypocladium niveum
HISTORY: IFO (T. Ito; T. Yokoyama R-1481-3; fine sandy humus).
31976 Trichoderma harzianum
HISTORY: IFO (T. Ito; T. Yokoyama R-1458-1; soil).
31977 Trichoderma pseudokoningii
HISTORY: IFO (T. Ito; T. Yokoyama R42-1611-4; litter).
31978 Arthroderma uncinatum
HISTORY: IFO (T. Ito; T. Yokoyama R-1606-11; soil).
31979 Verticillium psalliotae
HISTORY: IFO (T. Ito; T. Yokoyama R-1461-4; moss box).
31980 Wardomyces anomalus
HISTORY: IFO (T. Ito; T. Yokoyama R-1485-30; fine sandy clay).
31981 Wardomyces inflatus
HISTORY: IFO (T. Ito; T. Yokoyama R-1499-9; cabbage field soil).
31982 Fusarium oxysporum f. sp. fragariae
HISTORY: Veg. & Ornam. Crops Res. St. -- Nara Pref. Agr. Exp. St. (Ha-2; crown of strawberry).
31983 Fusarium oxysporum f. sp. fragariae
HISTORY: Veg. & Ornam. Crops Res. St. (S. Takeuchi; Nara ichigo-3; crown of strawberry).
31984 Fusarium oxysporum f. sp. fragariae
HISTORY: Veg. & Ornam. Crops Res. St. -- Nara Pref. Agr. Exp. St. (T. Kodama; A-3; soil).
31985 Fusarium oxysporum f. sp. fragariae
HISTORY: Veg. & Ornam. Crops Res. St. -- Nara Pref. Agr. Exp. St. (T. Kodama; B-1; soil).
31986 Fusarium oxysporum f. sp. fragariae
HISTORY: Veg. & Ornam. Crops Res. St. -- Nara Pref. Agr. Exp. St. (T. Kodama; B-3; soil).
31987 Rhizopus oligosporus

HISTORY: FIRDI 31631 -- DSM 1964 -- Meyer -- Steinkraus.
31988 Rhizopus microsporus
HISTORY: FIRDI 31630 -- DSM 1834 -- Meyer -- Steinkraus.
31989 Rhizopus azygosporus
HISTORY: FIRDI 31158 -- ATCC 48108 -- G. Harris -- J.N. Hedger.
31990 Pythium iwayamai
HISTORY: Coll. Agr., Univ. Osaka Pref., UOP 8 -- Fukui Agr. Exp. St. (S. Takamatsu; W-82-24; leaf of Triticum aestivum).
31991 Pythium iwayamai
HISTORY: Coll. Agr., Univ. Osaka Pref., UOP 11 -- Fukui Agr. Exp. St. (S. Takamatsu; W-82-50; leaf of Triticum aestivum).
31992 Pythium iwayamai
HISTORY: Coll. Agr., Univ. Osaka Pref., UOP 12 -- Fukui Agr. Exp. St. (S. Takamatsu; H-82-78; leaf of Hordeum sativus).
31993 Pythium paddicum
HISTORY: Coll. Agr., Univ. Osaka Pref., UOP 6 -- Fukui Agr. Exp. St. (S. Takamatsu; P-7; leaf of Hordeum vulgare).
31994 Pythium paddicum
HISTORY: Coll. Agr., Univ. Osaka Pref., UOP 15 -- Fukui Agr. Exp. St. (S. Takamatsu; W-82-15; leaf of Triticum aestivum).
31995 Pythium paddicum
HISTORY: Coll. Agr., Univ. Osaka Pref., UOP 83 -- Fukui Agr. Exp. St. (S. Takamatsu; S 83193; wheat field soil).
31996 Pythium graminicola
HISTORY: Coll. Agr., Univ. Osaka Pref., UOP 13 -- Fukui Agr. Exp. St. (S. Takamatsu; H-82-36; leaf of Hordeum vulgare).
31997 Pythium graminicola
HISTORY: Coll. Agr., Univ. Osaka Pref., UOP 20 -- Fukui Agr. Exp. St. (S. Takamatsu; H-82-35; leaf of Hordeum vulgare).
31998 Pythium graminicola
HISTORY: Coll. Agr., Univ. Osaka Pref., UOP 21 -- Fukui Agr. Exp. St. (S. Takamatsu; H-82-38; leaf of Hordeum vulgare).
31999 Gonytrichella olivacea
HISTORY: IFO (T. Ito; ISR 18-4; leaf of broad-leaved tree).
32000 Fusarium oxysporum f. sp. raphani
HISTORY: Veg. & Ornament. Crops Res. St., Japan (S. Takeuchi; 772121; root of Raphanus sativus).
32001 Verticillium nigrescens
HISTORY: Hokkaido Nat. Agr. Exp. St. (K. Kitazawa; VNP-720; stem of Solanum tuberosum).
32002 Rhizopus oligosporus
HISTORY: Nat. Food Res. Inst., L-26 -- Nat. Inst. Chem. Indonesia (D.L. Tanuwidjaja; L-26; starter for Tempeh).
32003 Rhizopus oligosporus
HISTORY: Nat. Food Res. Inst., L-36 -- Nat. Inst. Chem. Indonesia (D.L. Tanuwidjaja; L-36; starter for Tempeh).
32004 Penicillium argillaceum
HISTORY: IFO (T. Ito; III; sake brewery).
32005 Amblyosporium botrytis
HISTORY: IFO (T. Ito; T. Ito S58-18-1; humus).
32006 Chaetomium fusiforme
HISTORY: IFO (T. Ito; T. Ito S58-28-6; burnt soil).
32007 Chaetomium seminudum
HISTORY: IFO (T. Ito; T. Ito S58-9-5; urea treated soil).
32008 Gelasinospora calospora
HISTORY: IFO (T. Ito; T. Ito S58-28-1; burnt soil).
32009 Gelasinospora reticulospora
HISTORY: IFO (T. Ito; T. Ito S58-21-3; burnt soil).
32010 Gelasinospora reticulospora
HISTORY: IFO (T. Ito; T. Ito S58E-24-5; burnt soil).

32011 Neurospora tetrasperma
HISTORY: IFO (T. Ito; T. Ito S5870-28-1; burnt soil).
32012 Pleospora herbarum
HISTORY: Res. Cent., Nippon Beet Sugar Mfg. Co. (H. Uchino; SB 83; Beta vulgaris var. sacchrifera).
32013 Cristulariella moricola
HISTORY: Fac. Agr., Hirosaki Univ. (Y. Harada; 837) -- Aomori Field Crops & Hort. Exp. St. (S. Noro; Noro 83 G-10; grapevine leaf).
32014 Grovesinia pyramidalis
HISTORY: Fac. Agr., Hirosaki Univ. (Y. Harada; 838) -- Aomori Field Crops & Hort. Exp. St. (S. Noro; Noro 85 Kobus II-1; Magnolia kobus leaf).
32015 Grovesinia pruni
HISTORY: Fac. Agr., Hirosaki Univ. (Y. Harada; 839) -- Aomori Field Crops & Hort. Exp. St. (S. Noro; Noro 82 Aa-3; apricot leaf).
32016 Grovesinia pruni
HISTORY: Fac. Agr., Hirosaki Univ. (Y. Harada; 840) -- Aomori Field Crops & Hort. Exp. St. (S. Noro; Noro 85JAa-2; Prunus mume leaf).
32034 Acremonium strictum PP 58-K-857
HISTORY: IFO (T. Ito; ISR 6-3; Eucalyptus leaves).
32035 Aspergillus melleus PP 58-K-857
HISTORY: IFO (T. Ito; ISR 14-2; leaves of broad-leaved tree).
32037 Chaetomium elatum PP 58-K-857
HISTORY: IFO (T. Ito; ISR 39-1; twigs).
32038 Chaetomium olivaceum PP 58-K-857
HISTORY: IFO (T. Ito; ISR 52-2; leaves of broad-leaved tree).
32039 Cladosporium sphaerospermum PP 58-K-857
HISTORY: IFO (T. Ito; ISR 32-3; Pinus leaves).
32040 Doratomyces stemonitis PP 58-K-857
HISTORY: IFO (T. Ito; ISR 47-1; leaves of needle-leaved tree).
32041 Idriella lunata PP 58-K-857
HISTORY: IFO (T. Ito; ISR 36-4; Benzoin umbellatum leaves).
32042 Melanospora zamiae PP 58-K-857
HISTORY: IFO (T. Ito; ISR 31-3; Chrysanthemum leaves).
32043 Penicillium rubrum PP 58-K-857
HISTORY: IFO (T. Ito; ISR 26-3; leaves of broad-leaved tree).
32047 Arachniotus aurantiacus
HISTORY: IFO (T. Ito; ISR 37-1; twigs).
32048 Arachnomyces nitidus
HISTORY: IFO (T. Ito; ISR 2-4; twigs).
32049 Chaetomium murorum
HISTORY: IFO (T. Ito; ISR 29-2; leaves of broad-leaved tree).
32050 Zygosporium mycophilum
HISTORY: IFO (T. Ito; ISR 11-3; Nerium oleander leaves).
32051 Emericella variecolor
HISTORY: IFO (T. Ito; ISR 11-1; Nerium oleander leaves).
32052 Emericella variecolor
HISTORY: IFO (T. Ito; ISR 26-2; leaves of broad-leaved tree).
32053 Petriella setifera
HISTORY: IFO (T. Ito; ISR 24-2; Rosa leaves).
32054 Scolecobasidium humicola
HISTORY: IFO (T. Ito; ISR 64-3; Eucalyptus leaves).
32055 Armillaria bulbosa PP 61-K-1150
HISTORY: Inst. Natl. Recherche Agr. (J.J. Guillaumin; ME80-15.1; monospore).
32056 Armillaria cepaestipes PP 61-K-1150
HISTORY: Inst. Natl. Recherche Agr. (J.J. Guillaumin; T2-1; monospore).
32057 Armillaria obscura PP 61-K-1150
HISTORY: Inst. Natl. Recherche Agr. (J.J. Guillaumin; SF2.5; monospore).
32059 Crinipellis perniciosa PP 61-K-1870
HISTORY: Univ. Coll. Wales (G.M. Griffith; 226a; cacao tissue).
32060 Armillariella mellea PP 61-K-1647

HISTORY: Inst. Natl. Recherche Agr. (J.J. Guillaumin; PM 8 (MD 79.13.8); monospore).

50034 RBL-1
HISTORY: RTCI (S. Terao) -- Kyoto Univ.

50036 P388D1
HISTORY: RTCI (S. Iwasa) -- Flow Labs., Inc.

50037 RPMI 1788
HISTORY: IFO (M. Takeuchi) -- RTCI.

50038 U-937
HISTORY: IFO (M. Takeuchi) -- RTCI.

50039 NC-37
HISTORY: IFO (M. Takeuchi) -- RTCI.

50042 C3H/MCA clone 15
HISTORY: RTCI (S. Iwasa) -- Flow Labs., Inc.

50043 WiDr
HISTORY: RTCI (N. Suzuki) -- Flow Labs., Inc.

50046 Raji
HISTORY: IFO (M. Takeuchi) -- RTCI.

50067 LoVo
HISTORY: RTCI (K. Kondo) -- Flow Labs., Inc.

50068 COS-7
HISTORY: IFO (M. Takeuchi) -- RTCI.

50069 Alexander cells
HISTORY: IFO (M. Takeuchi) -- RTCI.

50070 Balb/3T3-A31-1-1
HISTORY: RIMD (T. Kakunaga).

50071 MDCK (NBL-2)
HISTORY: IFO (M. Takeuchi) -- ATCC (CCL 34).

50072 MRC-9
HISTORY: IFO (M. Takeuchi) -- ATCC (CCL 212).

50073 MRC-5
HISTORY: IFO (M. Takeuchi) -- ATCC (CCL 171).

50074 HFL1
HISTORY: IFO (M. Takeuchi) -- ATCC (CCL 153).

50075 WI-38
HISTORY: IFO (M. Takeuchi) -- ATCC (CCL 75).

50076 IT45R1
HISTORY: Tohoku Univ. (T. Itoh).

50077 IT45R91
HISTORY: Tohoku Univ. (T. Itoh).

50078 IT26R21
HISTORY: Tohoku Univ. (T. Itoh).

50079 Flow 7000
HISTORY: RTCI (R. Sasada) -- Dainippon Pharm. Co., Ltd.

50080 NCTC clone 1469
HISTORY: RTCI (M. Inoue) -- Dainippon Pharm. Co., Ltd.

50081 Neuro-2a
HISTORY: RTCI (M. Suno) -- ATCC (CCL 131).

50082 V79 379A
HISTORY: RTCI (M. Watanabe) -- Flow Labs., Inc.

50089 Flow 2000
HISTORY: RTCI (T. Kurokawa) -- Dainippon Pharm. Co., Ltd.

ABSTRACTS 1985-1986

Reclassification of "Flavobacterium arborescens" (Frankland and Frankland) Bergey et al. in the Genus Microbacterium (Orla-Jensen) Collins et al., as Microbacterium arborescens comb. nov., nom. rev.

K. Imai, M. Takeuchi and I. Banno
Curr. Microbiol. 11: 281-284 (1984)

"Flavobacterium arborescens" (Frankland and Frankland) Bergey et al. IFO 3750 (ATCC 4358) is a Gram-positive, coryneform bacterium and the only available reference strain of the species. The cell wall peptidoglycan of the organism possesses alanine, glycine, lysine, glutamic acid plus 3-hydroxyglutamic acid, and homoserine at a ratio of 1:3:1:1:1, and a possible peptidoglycan structure is the B1 type described by Schleifer and Kandler. Cell wall sugars are galactose, mannose, and 6-deoxy-L-talose, but not rhamnose. Major menaquinones are unsaturated MK-11 and MK-12. These findings and other taxonomic properties suggest that "F. arborescens" should be reclassified in the genus Microbacterium (Orla-Jensen) Collins et al., as Microbacterium arborescens comb. nov., nom. rev.

Studies on preservation of actinomycetes strains; Preservation of strain in a frozen state

T. Kusaka, K. Sato and I. Asano
JFCC Newsletter 12: 2-6 (1984)

Long-term preservation of actinomycetes strains by cryopreservation method was tested. One ml of whole cultured broth shaken for 4 or 6 days was poured into plastic tube directly without any protectant and frozen in the deep freezer at -80 C. They were thawed at 40 C for 3 min after one day, six months, one year and two years, respectively and their

survival as well as their characteristics were investigated. The thawed sample was frozen and stored for the succeeding examination. Periodical investigations about tested strains indicated that 98.9% of the 769 tested strains survived for two years under the conditions. The method seemed to be able to apply to the long-term preservation of actinomycetes.

(in Japanese)

The coenzyme Q system in strains of species in the genus Sterigmatomyces (Cryptococcaceae) and its teleomorphic genus Sterigmatosporidium

Y. Yamada* and I. Banno

Trans. mycol. Soc. Japan 25: 455-460 (1984)

Eighteen strains of Sterigmatomyces and Sterigmatosporidium species were examined for the Co-Q system. Six species of the genus Sterigmatomyces were divided into two groups; one is comprised of the Q_9-equipped strains assigned to S. elviae and S. halophilus, and the other is comprised of the Q_{10}-equipped strains assigned to S. nectairii, S. penicillatus, S. polyborus, and S. tursiopsis. In the genus Sterigmatosporidium, only the Q_{10}-equipped strains were found. These data are discussed from the taxonomic point of view.

* Department of Agricultural Chemistry, Shizuoka University

Fellomyces, a new anamorphic yeast genus for the Q_{10}-equipped organisms whose conidium is freed by an end-break in the sterigma

Y. Yamada* and I. Banno

J. Gen. Appl. Microbiol. 30:523-525 (1984)

A new genus Fellomyces was proposed for Q_{10}-equipped yeasts whose conidia were detached by a separation at the distal end of sterigmata.

Consequently the concept of the genus Sterigmatomyces has been emended.

* Department of Agricultural Chemistry, Shizuoka University

Transcriptional and post-transcriptional control of PHO8 expression by PHO regulatory genes in Saccharomyces cerevisiae

Y. Kaneko, Y. Tamai*, A. Toh-e**, and Y. Oshima*

Mol. Cell. Biol. 5: 248-252 (1985)

A DNA fragment bearing the PHO8 gene, which encodes repressible alkaline phosphatase of Saccharomyces cerevisiae, was cloned. Northern hybridizations with the PHO8 DNA as probe indicated that the PHO8 transcript is 1.8 kilobases in length and is more abundant in cells grown in low-phosphate medium than in high-phosphate medium. The pho9 mutant, whose phenotype is defective in the activity of repressible alkaline phosphatase, produced as much of the PHO8 transcript as did the PHO9$^+$ cells. Hence, the PHO9 product should act at the post-transcriptional level. The pho4 mutant could not derepress the PHO8 transcript, whereas the pho80 mutant could, irrespective of the amount of Pi in the medium, as has been suggested by genetic study.

* Department of Fermentation Technology, Osaka University
** Department of Fermentation Technology, Hiroshima University

Compounds protecting L-dried cultures from mutation. III. Effect of cysteine on prevention of mutation in L-dried E. coli cells

T. Sakane, K. Imai and I. Banno

Japan. J. Freez. Dry. 31: 27-35 (1985)

Protective effect of cysteine on L-dried E. coli cells was investigated.

When cysteine was added into the suspending medium at a concentration of 30 mM, increasing in survival value and decreasing in mutation frequency were observed in *uvrA*$^-$, *uvrB*$^-$, or *recA*$^-$ mutants, but not in *polA*$^-$ or *ung*$^-$ mutants. Moreover, cysteine did not prevent the transitional reversion from GC to AT in the *trp* gene. An agarose gel electrophoretic analysis showed that cysteine did not prevent directly DNA breakage by radicals arising in the dried cells. Those results suggest that cysteine stimulates the activity of DNA-repair system after rehydration of L-dried cells and/or acts protectively on the DNA-repair enzyme(s) during preservation of L-dried speciemens.

(in Japanese)

Conidiogenous cells of Fusarium wilt fungus of strawberry, *Fusarium oxysporum* f. sp. *fragariae*

S. Takeuchi*, Y. Okamoto**, T. Yokoyama and H. Hagiwara***

Trans. mycol. Soc. Japan 26: 343-348 (1985)

Conidiogenous cells of Fusarium wilt fungus of strawberry, *Fusarium oxysporum* Schlechtendahl emend. Snyder et Hansen f. sp. *fragariae* Williams, growing on potato maltose agar and carnation leaf agar were examined by a scanning electron microscope. Throughout the repeated examinations, polyphialides were noticed very rately, in about 3 out of 15 strains examined, but the condition which induces polyphialide formation was not elucidated. In the other cases, including 5 strains newly isolated from infested soils, practically all of the conidiogenous cells examined were the monophialide, and with the rest of them it was difficult to decide between the monophialide and polyphialide. On the basis of these observations, it is concluded that the fungus is to be classified into a group which produces only the monophialide.

(in Japanese)

* Vegetable and Ornamental Crops Research Station
** North Branch, Okayama Prefectural Agricultural Experiment Station
*** National Agriculture Research Center

Ubiquinone systems in fungi I. Distribution of ubiquinones in the major families of ascomycetes, basidiomycetes, and deuteromycetes, and their taxonomic implications

H. Kuraishi*, Y. Katayama-Fujimura*, J. Sugiyama**, and T. Yokoyama

Trans. mycol. Soc. Japan 26: 383-395 (1985)

The ubiquinone (coenzyme Q) systems of 218 species assigned to 195 teleomorph and 41 anamorph genera, 67 families and 4 anamorph sections in higher fungi, except yeasts and yeast-like fungi, have been determined by high performance liquid chromatography. Of these major isoprenologues detected are the sole ubiquinone structural types or a combination of two or three ubiquinone structural types: Q-(8+9), Q-9, Q-(9+10), Q-10, Q-(10+10(H_2)), Q-(10+10(H_2)+10(H_4)), Q-10(H_2), Q-(10(H_2)+10(H_4)), and Q-10(H_4). The majority of higher fungi examined possess the Q-9, Q-10, or Q-10(H_2) system as major isoprenologue. Various major ubiquinone systems are found in taxa of Pyrenomycetes and Loculoascomycetes have remarkably uniform ubiquinone profiles, with the Q-10(H_2) system predominating. Heterobasidiomycetous fungi possess the Q-9 or Q-10 system as their major isoprenologue, whereas all Homobasidiomycetous fungi are characterized by the Q-9 system. No taxa possessing major amount of hydrogenated Q-10 have been found in the Basidiomycotina. Data obtained suggest that ubiquinone systems are very useful in the classification of fungal taxa and in the elucidation of their genealogy.

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** Institute of Applied Microbiology, University of Tokyo

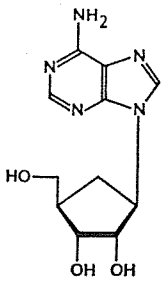
Isolation and characterization of a new aristeromycin analog

O. Miyashita,* F. Kasahara,* T. Kusaka and R. Marumoto*

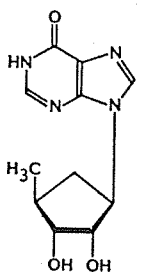
J. Antibiot. 38: 981-986 (1985)

The chemical structure of aristeromycin M, a new carbocyclic nucleoside, was elucidated by spectroscopic analysis and chemical

transformation from aristeromycin.



Aristeromycin



Aristeromycin M

* Central Research Division, Takeda Chemical Industries Ltd.

Zygophiala jamaicensis Mason, a causal fungus of flyspeck of grape, Japanese persimmon and apple

H. Nasu*, S. Fujii*, and T. Yokoyama

Ann. Phytopath. Soc. Japan 51: 536-545 (1985)

A new symptom of grape (Vitis vinifera L. cv. 'Muscut of Alexandria' and cv. 'Glos Colman'), the evanescence of a bloom which normally covers on the skin of berries, has been prevaling in greenhouses in Okayama Prefecture, Japan, since several years ago. No necrotic lesion nor rot of berries was seen. Similar symptom has also been found on Japanese persimmon and apple fruits in Okayama. The evenescence of the bloom on affected berries is visible to the naked eyes in accordance with the extention of subhyaline, superficial creeping mycelium which tends to form mycelial networks on berries. After a later stage of the symptom development, black microsclerotia-like bodies are produced in abundance, which give berries typical flyspeck symptom on them. Conidial structures, spiral tubular conidiophores and gourd-shaped conidia, are formed on the creeping mycelium, particularly around the microsclerotia-like bodies. Based on the morphological characteristics of the causal pathogens on grapevine, it was identified as Zygophiala jamaicensis Mason apud Martyn, a hyphomycete fungus. The causal fungus of flyspeck of Japanese persimmon and apple was also found

conspecific with this species. On the diseased fruits concerned, however, there was no evidence for Leptothyrium pomi (anamorphic state of Shizothyrium pomi) which was thought to be the causal agent of flyspeck of various kinds of fruits. Two types of colonies were segregated on the potato sucrose agar media. One (type A) is of fast growing type which produces both microsclerotia-like bodies and conidial structure in abundance. The other (type B) is of slow growing type which also produces the conidial structures well, but produces few microsclerotia-like bodies. The fungus can grow at a range of temperature at 6-28 C and of pH 3-11. The optimum temperature for growth was at 20-25 C. Inoculation tests by selected isolates from grapevine, Japanese persimmon and apple on non-injured fruits of grapevines, Japanese persimmons and apples revealed that these isolates are virulent to these fruits.

(in Japanese)

* Okayama prefectural Agricultural Experiment Station

Preservation of yeast cultures by freezing at -80 C

K. Mikata and I. Banno

Japan. J. Freez. Dry. 32: 58-63 (1986)

Freezing at -80 C has been applied for preservation of 141 yeast cultures, which were very sensitive to L-drying. The cells were frozen in 10% glycerol solution by electric deep-freezer, and survival values were determined immediately after freezing and after preservation of one, six and 48 months. All cultures except 7 strains showed high survival value. Viable counts of Candida bogoriensis IFO 1966, Candida slooffii IFO 0874, Leucosporidium nivale IFO 1852, Lipomyces starkeyi IFO 1289, Nadsonia commutata IFO 10029, Saccharomyces exiguus IFO 1169 and Torulopsis holmii IFO 0660 were less than 1% and insuficient for the frozen culture to survival for a long term.

Using the strain C. bogoriensis IFO 1966, L. starkeyi IFO 1289 and S. exiguus IFO 1169, effect of addition of 5 cryo-protectants and frequency of freeze-and-thawing was examined on viability of the frozen cells. Viability of the 3 cultures gradually decreased with repetition

of freeze-and-thawing. The degradation of the cells after 20 repetition for 30 days was the slowest rate when DMSO was used as a protectant.

(in Japanese)

Preservation of various bacteria by L-drying. Part II

T. Sakane and K. Imai

Japan. J. Freez. Dry. 32: 47-53 (1986)

Fifty-seven bacterial strains belonging to twenty-three genera were subjected to L-drying, and their viabilities were examined. Twenty-seven strains belonging to the genera Acidiphilium, Bacteroides, Bdellovibrio, Bifidobacterium, Clostridium, Desulfotomaculum, Desulfovibrio, Hyphomicrobium, Lactobacillus, Photobacterium, Propionibacterium, Streptococcus, Thermoanaerobacter, and Zymomonas showed high survival values between 9 and 98% when 0.1 M phosphate buffer containing 3% MSG, 1.5% adonitol and 0.05% cysteine was employed as the suspending fluid. Remaining thirty strains showed low survival values, and the suspending fluid was improved to obtain desirable viability after drying. Of thirty strains, twenty-eight strains gave the survival values of 1% or more using the following suspending fluids: 0.01 M phosphate buffer containing 0.3% MSG, 1.5% adonitol and 0.005% cysteine for four strains of Azotobacter; sea water containing 5% MSG, 1.5% adonitol and 0.05% cysteine for four halophilic bacteria belonging to the genera Cytophaga, Hyphomicrobium, and Photobacterium; PPLO broth supplemented with 3% sucrose and 10% horse serum for four strains belonging to the genera Mycoplasma and Acholeplasma, fluids composed of 0.5% cysteine in distilled water or sea water for eight strains belonging to the genera Aquaspirillum and Oceanospirillum, and 0.02 M phosphate buffer (pH 4 or 7) containing 0.3% MSG for eight strains belonging to the genera Nitrobacter, Nitrosomonas, and Thiobacillus. Other two strains, Oceanospirillum hiroshimense IFO 13616 and Oceanospirillum multiglobuliferum IFO 13614 still showed survival values less than 0.002%, even using improved suspending fluids.

(in Japanese)

Stability of plasmids in L-dried cells of E. coli strains

T. Sakane and K. Imai

Japan. J. Freez. Dry. 32: 83-88 (1986)

L-dried specimens of E. coli strains carrying one of three plasmids, pBR322, pSC138 or F'102, were prepared, and the plasmid stability in survivors was examined after rehydration and recovering on the nonselective medium. Immediately after drying, the plasmids were stably maintained in survivors. After preservation at 37 C for 6 weeks, survival values of $recA^-$ strains were decreased less than 0.1%, and the loss of plasmids was observed at the ratio of 0.5, 0.5, and 5.5% of survivors for pBR322, pSC138, and F'102, respectively. On the other hand, in $recA^+$ strains high survival values and no loss of the plasmids were found even after preservation at 37 C for 6 weeks. Addition of 30 mM thiourea or 100 mM adonitol and 3 mM cysteine into the suspending fluid remarkably increased survival values of the specimens of $recA^-$ strains and prevented the loss of plasmids. DNAs of pBR322 and pSC138 were extracted from L-dried cells and electrophoresed on agarose gels. The plasmid DNAs isolated from L-dried cells preserved at 37 C for 8 weeks were decreased in the CCC-form and increased in the OC-form, as compared the DNAs from cells preserved at 5 C. After rehydration with PY broth, cells were incubated at 37 C for 90 min, and the plasmid DNAs were analyzed by agarose gel electrophoresis. The increase in the CCC-form indicated that the injured plasmid DNAs were repaired during the incubation.

(in Japanese)

Isolation and amino acid sequence of a mating pheromone produced by mating type α cells of Saccharomyces exiguus

A. Sakurai*, H. Tanaka*, Y. Esumi*, N. Takahashi*, T. Hisatomi**, N. Yanagishima** and I. Banno

FEBS Letters 203: 285-288 (1986)

A peptide, termed α^{se} pheromone, was isolated as a mating pheromone

from culture filtrate of mating type α cells of Saccharomyces exiguus. The peptide showed both agglutinability-inducing activity to a cells of S. cerevisiae and shmoo-inducing action to a cells of S. cerevisiae, S. kluyveri and S. exiguus. The amino acid sequence of α^{se} pheromone was determined as H-Trp-His-Trp-Leu-Arg-Leu-Ser-Tyr-Gly-Gln-Pro-Ile-Tyr-OH by mass spectrometry, sequence analysis and enzymatic digestion.

* The Institute of Physical and Chemical Research
** Faculty of Science, Nagoya University

The electrophoretic comparison of enzymes in strains of species in the anamorphic yeast genera Sterigmatomyces and Fellomyces and in the teleomorphic yeast genus Sterigmatosporidium

Y. Yamada*, M. Watanabe*, M. Akita*, and I. Banno
J. Gen, Appl. Microbiol. 32: 157-163 (1986)

A taxonomic study below the generic or at the specific level was made of the electrophoretic pattern of seven enzymes in Sterigmatomyces, Fellomyces, and Sterigmatosporidium species. The seven enzymes were glucose-6-phosphate dehydrogenase, 6-phosphogluconate dehydrogenase, isocitrate dehydrogenase, malate dehydrogenase, hexokinase, phosphoglucomutase, and catalase. All examined strains of S. elviae, S. halophilus (=S. indicus), and S. polymorphum gave a uniform electrophoretic enzyme pattern within the respective species. Four strains of S. halophilus and three strains of S. halophilus (=S. indicus) were linked to each other with a similarity value of 43%. The similarity value between S. elviae and S. halophilus was calculated to be only 14%. The three Fellomyces species, F. nectairii, F. penicillatus, and F. polyborus and S. tursiopsis had quite different electrophoretic enzyme patterns. Their similarity values were all 0%. Between F. nectairii and S. polymorphum, the similarity value was only 14%. These data are discussed from the taxonomic point of view.

* Department of Agricultural Chemistry, Shizuoka University

Formation of 1-deoxy-ketoses by pyruvate dehydrogenase and acetoin dehydrogenase

A. Yokota and K. Sasajima*

Agric. Biol. Chem. 50: 2517-2524 (1986)

Cell-free extracts of Bacillus subtilis contain enzyme activities which catalyze an acyloin-type condensation reaction (carboligase reaction) resulting in the formation of 1-deoxy-ketoses. The reactions are deduced to proceed as follows:

pyruvate + aldose ⟶ CO_2 + 1-deoxy-ketose (I)

acetoin + aldose ⟶ acetaldehyde + 1-deoxy-ketose (II)

methylacetoin + aldose ⟶ acetoine + 1-deoxy-ketose (III)

Experiments with mutants of B. subtilis defective in pyruvate dehydrogenase (PDH) or acetoin dehydrogenase (AccDH) and with partially purified enzyme preparations revealed that PDH (EC 1.2.4.1) catalyzes reaction (I), and AccDH catalyzes reactions (II) and (III).

That the PDH purified from Escherichia coli and the PDC purified from bovine heart also catalyzed reaction (I) indicates that 1-deoxy-ketose-forming activities are widely distributed. One of the reactions catalyzed by these enzymes is the formation of 1-deoxy-D-threo-pentulose, a precursor of biosynthesis of thiazole ring of thiamine.

* Central Research Division, Takeda Chemical Industries, Ltd.

Induction of heterothallic strains and their genetic and physiological characterization in a homothallic strain of the yeast Saccharomyces exiguus

T. Hisatomi*, N. Yanagishima*, and I. Banno

Current Genetics 10: 887-892 (1986)

We isolated heterothallic strains from a homothallic strain of S. exiguus by mutagenization with UV or ethylmethanesulfonate (EMS). A gene, not linked to the mating-type locus, was found to control homothallism

in the yeast, as in S. cereviseae. α Pheromone of S. exiguus (α^{se} pheromone) induced formation of large pear-shaped cells (shmooing) in a strains of S. exiguus, S. cerevisiae, and S. kluyveri, and sexual agglutinability of an inducible a strain of S. cerevisiae. α^{se} Pheromone is a peptidyl substance a little different from α pheromone of S. cerevisiae. a Pheromone of S. exiguus acts only on α cells of S. exiguus. Contrary to the above results, neither sexual agglutination nor zygote formation occurred among these three Saccharomyces yeasts.

* Faculty of Science, Nagoya University

Secreted agglutinability-masking factors in Issatchenkia scutulata var. scutulata

S. Hasegawa*, K. Tanaka*, I. Banno, and N. Yanagishima*

Antonie van Leeuwenhoek 52: 371-379 (1986)

The agglutinability-masking factors (AMFs) of a and α mating types of Issatchenkia scutulata var. scutulata were prepared from culture fluids. AMFs masked the agglutinability of opposite mating-type cells sex-specifically, just like agglutination substances responsible for sexual cell agglutination. a AMF adsorbed to α cells was eluted by incubating the cells at 60°C for 10 min. α AMF was prepared directly from culture fluids of α cells by DEAE-cellulose ion exchange chromatography. The active part of the AMFs is thought to be a peptidyl moiety because of the sensitivity to subtilisin. The pretreatment of cells with AMF of the opposite mating-type was shown to promote zygote formation. α AMF slightly inhibited growth in a cells but not in α cells, while a AMF did not show any growth-inhibitory effect on either a or α cells.

* Department of Biology, Faculty of Science, Nagoya University

PRESENTATION OF PAPAERS AT SCIENTIFIC MEETINGS 1985-1986

Kansai Mycological Club (February, 1985, Kyoto)

T. Ito and T. Yokoyama
Filamentous fungi collected in the Far Eastern USSR.

K. Mikata and I. Banno
Morphology of ascospores of ascomycetous yeasts.

Japanese Society for Research of Freezing and Drying (April, 1985, Tokyo)

T. Sakane, K. Imai and I. Banno
Compounds protecting L-dried cultures from mutation. III. Effects of cysteine on prevention of mutation in L-dried E. coli cells.

International Institute of Refrigeration Commission C1 Meeting (May, 1985, Tokyo)

K. Imai and T. Sakane
Preservation of chemolithotrophic bacteria by L-drying.

T. Iijima, T. Sakane, K. Imai and I. Banno
Compounds which protect L-dried cultures from mutation.

Agricultural Chemical Society of Japan (July-August, 1985, Sapporo)

K. Imai
Isolation and preliminary characterization of Bacillus pumilus bacteriophages.

M. Takeuchi, A. Yokota, K. Imai and A. Misaki*1
Characterization of the polysaccharide isolated from the cell wall of Microbacterium imperiale IFO 12610.

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*1 Faculty of the Science of Living, Osaka City University

The Genetics Society of Japan (October, 1985, Kobe)

Y. Kaneko, I. Banno, A. Toh-e[*1] and Y. Oshima[*2]

The nucleotide sequence of the PHO8 gene encoding repressible alkaline phosphatase of Saccharomyces cerevisiae.

Society for Fermentation Technology, Japan (October, 1985, Tokyo)

I. Banno

Recent trend of taxonomy of microorganisms: Ascomycetous and basidiomycetous yeasts.

Y. Yamada, A. Yokota and K. Imai

Lipopolysaccharides isolated from iron-oxidizing bacteria (I). Chemical composition.

A. Yokota, Y. Yamada and K. Imai

Lipopolysaccharides isolated from iron-oxidizing bacteria (II). Identification of an unknown sugar in LPS of IFO 14262.

Agricultural Chemical Society of Japan (April, 1986, Kyoto)

K. Imai

Characterization of a temperate Bacillus pumilus bacteriophage NP-5.

Phytopathological Society of Japan (April, 1986, Nagoya)

S. Kasuyama[*3], T. Idei[*3] and T. Yokoyama

Teleomorphic state of grape black rot fungus (Guignardia bidwellii) newly found in Okayama Prefecture, Japan.

H. Nasu[*3], T. Yokoyama, H. Komatsu[*4] and M. Hatamoto[*3]

A new disease of peach caused by a new species of the genus Stenella.

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*1 Department of Fermentation Technology, Hiroshima University

*2 Department of Fermentation Technology, Osaka University

*3 Okayama Prefectural Agricultural Experiment Station

*4 Wakayama Prefectural Horticultural Experiment Station

Phytopathological Society of Japan (April, 1986, Nagoya)

M. Yoshikawa[*1] and T. Yokoyama

A new disease of Hemerocallis spp. caused by Aureobasidium microstictum.

Japanese Society for Research of Freezing and Drying (April, 1986, Tokyo)

K. Mikata and I. Banno

Preservation of yeast cultures by freezing at -80 C.

T. Sakane and K. Imai

Preservation of various bacteria by L-drying. II.

T. Sakane and K. Imai

Stability of plasmids in L-dried cells of E. coli strains.

Japanese Society of Mycoplasmology (May, 1986, Tokyo)

T. Yoshida, M. Kawase[*2], K. Sasaki[*2], H. Mizusawa[*2] and M. Takeuchi

Detection of mycoplasmal contamination in animal cell lines: Comparison of DNA-staining, culture and cytotoxicity methods.

Mycological Society of Japan (May, 1986, Hirosaki)

T. Ito and T. Yokoyama

Litter fungi collected in Israel.

T. Yokoyama and T. Ito

Emericella variecolor Berkeley et Broome and related species.

H. Kuraishi[*3], Y. Katayama-Fujimura[*3], J. Sugiyama[*4] and T. Yokoyama

Ubiquinone systems in the genus Aspergillus and its teleomorphs.

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*1 Kyoto Prefectural Agricultural Experiment Station

*2 Division of Mutagenesis, National Institute of Hygienic Sciences

*3 Faculty of Agriculture, Tokyo University of Agriculture and Technology

*4 Institute of Applied Microbiology, University of Tokyo

The genetic Society of Japan (December, 1986, Nagoya)

Y. Kaneko and I. Banno

Genetic analysis of the hybrids between Saccharomyces bayanus and Saccharomyces cerevisiae.

MISCELLANEOUS SCIENTIFIC PAPERS

I. Banno. 1986. Preservation of microorganisms. Biseibutsu 2: 97-108. (in Japanese)

T. Iijima, T. Sakane, K. Imai and I. Banno. 1985. Compounds which protect L-dried cultures from mutation. Fundamentals and applications of freeze-drying to biological materials, drugs and foodstuffs, P. 273-277. International Institute of Refrigeration, Paris.

K. Imai and T. Sakane. 1985. Preservation of chemolithotrophic bacteria by L-drying. Fundamentals and applications of freeze-drying to biological Materials, drugs and foodstuffs, P. 233-234. Internatonal Institute of Refrigeration, Paris.

T. Iijma. 1986. Maintenance of microorganisms. In Y. Okami et al. (ed.) Saishin Biseibutsu Handbook, p. 54-61. Science Forum, Tokyo. (in Japanese)

T. Iijima and K. Imai. 1986. Reagents for general characterization of microorganisms. In Y. Okami et al. (ed.) Saishin Biseibutsu Handbook, p. 529-531. Science Forum, Tokyo. (in Japanese)

T. Yokoyama. 1986. Conversion and utilization of forest biomasses. I. Molds. In Y. Okami et al. (ed.) Saishin Biseibutsu Handbook, p. 454-466. Science Forum, Tokyo. (in Japanese)

T. Kusaka. 1985. Preservation of actinomycetes strains (L-drying method). In A. Seino (ed.) Hohsenkin no Dotei, p. 196-203. Society for Actinomycetes, Japan, Tokyo. (in Japanese)

T. Kusaka and T. Iijima. 1986. Preservation of productive strains. In M. Kizumi and Y. Horiuchi (ed.) Ohyo Bunshi Iden Gaku, p. 93-97. Kodan-sha Scientific, Tokyo.

(in Japanese)

Y. Okami and T. Kusaka. 1986. Qualification of the international streptomyces project (ISP) strains deposited at the Institute for Fermentation, Osaka (IFO) by the Society for Actinomycetes, Japan (SAJ). The Actinomycetes 19: 2-10.

K. Sasajima, A. Yokota and M. Yoneda. 1985. Production of D-ribose by bacteria ---- Basic research and application of carbohydrate metabolism mutants of Bacillus species. Kagaku to Seibutsu 23: 240-248.

(in Japanese)

M. Kawase, H. Mizusawa, K. Sasaki, T. Yoshida, M. Takeuchi, R. Harasawa and M. Ishidate. 1986. Detection procedures for mycoplasmal contamination in cultured animal cells. Soshiki Baiyo 12: 298-303.

(in Japanese)

T. Yokoyama. 1985. Culture collections of microorganisms and distribution of cultures. Jouranal of Antibacterial and Antifungal Agents 13: 453-460.

(in Japanese)

T. Yokoyama. 1986. Paddy field soil (Molds). In K. Yamasato et al. (ed.) Biseibutsu no Bunrihou, p. 54-66. R & D Planing Co., Ltd., Tokyo.

(in Japanese)

CORRECTIONS

In the issue of IFO Research Communications No. 12, the following corrections should be made.

Page	Line	Type	Should read
19	18	comples	complex
27	21	2,3-dideoxy-ald-2-enonic	2,3-dideoxy-ald-4-enonic
	24	withgood	with good
46	13	10 M. longicollis R-1529-4	delete
	29	R-1548-1	RE-1548-1
	30	R-1527-1	RE-1527-1
	48	R42-1601-1	R42-1601-4
47	41	R-16033	R-1603-8
48	12	(IFO 31382)	(IFO 31392)
	13	(IFO 31383)	(IFO 31393)
	19	(IFO 31384)	(IFO 31394)
	20	(IFO 31385)	(IFO 31395)
	22	R1463-17	R-1463-17
	33	(IFO 31386)	(IFO 31396)
	36	(IFO 31386)	(IFO 31397)
53	2	Chlamidospore	Chlamydospore
59	9	T. Yokoyama 1533-2	T. Yokoyama R-1533-2
136	13	5022	50022
	25 - 27		delete
	31 - 33		delete

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