# THE GENUS MICROASCUS

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#### Abstract

Descriptions are given of 14 species of Microascus. M. pyramidus Barron and Gilman is described as a new species and M. trigonosporus var. macrosporus Orr as a new variety. The conidial stage and cultural characteristics are described for most of the species. The genus *Microascus* is separated from *Petriella* on the basis of the smaller, lighter-colored ascospores as well as the type of conidia. Most of the species have a *Scopulariopsis*-type of conidial stage. *M. variabilis* Massee and Salmon is placed in synonomy under *M. longirostris* Zukal. *Microascus stysanophorus* (Matt.) comb. nov. is proposed.

#### Introduction

The genus *Microascus* was established by Zukal (1885) with *M. longirostris* as the type species. In its gross morphology, the ascocarp of M. longirostris is perithecial-like, being dark, carbonaceous, and long-necked; the asci, however, are distributed at all levels within the centrum as in the genus Eurotium. Realizing the Plectomycetous nature of Microascus, Fischer (1896) placed it in the Plectascineae in Engler and Prantl. This is noteworthy as one of the earlier records of centrum organization taking precedence over gross morphology as a taxonomic criterion. Microascus was later placed in the Sphaeriales by Curzi (1931) but there is little evidence to support him in this treatment and it is not generally followed.

Since Zukal's publication, a number of *Microascus*-like fungi have been described under a variety of generic names. The affinities of many of these species to *Microascus* were recognized by Emmons and Dodge (1931), and by Curzi (1931), who transferred them to the latter genus. There is some disagreement with respect to these transfers and the controversies concerning them will be discussed in detail in the taxonomic section below. Curzi (1930b) separated out also a second series of *Microascus*-like species into a new genus *Petriella*. This separation was based largely on the setose condition of the perithecium in the latter genus. *Petriella* is regarded as quite distinct from Microascus and has recently been reviewed by Barron et al. (1961).

Apart from the taxonomic problems, Microascus is of especial interest because of the many areas of biologic activity with which its species are connected. Some have shown pathogenic tendencies against man, animals, plants, and even insects; others have been isolated frequently as coprophilous fungi and more recently as component organisms of the soil microflora. The relationship of *Microascus* in these associations is, for the most part, little understood

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# Morphology and Development of the Ascocarp

Critical cytological investigations on several species of the genus have been carried out by Emmons and Dodge (1931), Jones (1936), Moreau and Moreau (1953), and Doguet (1957). The results of these studies are outlined below.

The ascocarp is initiated by a favored vegetative cell giving rise to an ascogonial branch which is frequently coiled. The cells of the ascogonium are at first uninucleate but later become multinucleate. In *M. lunasporus* (sensu Jones), it was observed that antheridia are normally present and Jones (1936) noted the passage of the antheridial nucleus into the ascogonium with subsequent pairing of the nuclei. Antheridia were not found by Moreau (1953) in *M. doguetii* but in *M. trigonosporus*, Emmons and Dodge (1931) noted that antheridial-like branches are sometimes present.

The ascogonium is septate with multinucleate cells and becomes enveloped rapidly by sterile vegetative hyphae giving the young ascocarp a globular shape. With further development the ascogonium becomes surrounded by several layers of pseudoparenchymatous cells elongated in a tangential direction. The outermost cells become carbonized and pigmented while the inner layers remain thin-walled and hyaline. The perithecia enlarge and the innermost, hyaline, thin-walled cells surrounding the ascogonium grow into the cavity formed by this enlargement. The cells, at first tapering, become distinctly hyphal forming a matrix in the center of the ascocarp. These hyphae develop more rapidly below than above, resulting in a differential growth which causes the ascogonium to take up a position above the center of the perithecium. Concurrent with this series of developments is the initiation of the ostiolar and neck regions. Elongation of the outer layers to form a papilla or neck results in the formation of a schizogenous subostiolar cavity which extends up to the ostiole and is accompanied by the inward growth of periphyses to form a lining in the ostiolar channel.

The ascogenous hyphae grow out radially from the ascogonium, particularly in a downward direction, into the body of the centrum. The ascogenous hyphae branch freely and by mutual pressure become radially arranged.

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Croziers are not found consistently but have been observed infrequently by Emmons and Dodge in *M. trigonosporus*. Asci arise from the ascogenous hyphae either singly or in tufts from the terminal and lateral branches or they sometimes may arise in an intercalary manner and in rows as found by Moreau in *M. doguetii*.

The asci are produced first in the region around the ascogonium; further development follows the maturation of the ascogenous hyphae towards the periphery of the ascocarp. The orientation of the asci is similar to the ascogenous hyphae with the long axis in a radial direction.

The sterile matrix, into which the ascogenous hyphae grow, breaks down early. The ascus wall also breaks down very early in some species, allowing the ascospores to reach final maturity lying free in the matrix of the perithecial cavity; in other species, the ascus wall persists until maturity of the ascospores. In either case, at maturity, the perithecial cavity is filled with the free ascospores which are extruded in a sticky ball at the mouth of the perithecium or as a long reddish-brown cirrhus which may exceed 5 mm in length.

The ascospores are single-celled, uninucleate, and germinate either at one or at both ends depending on the species. Single-spore cultures of some species indicate homothallism within the genus.

# **Occurrence and Distribution**

The habitat and geographical distribution of individual species is discussed under the species descriptions below; it is interesting, nevertheless, to consider the group as a whole in this respect.

#### Plant Pathogens

Microascus schumacheri, M. cincreus, M. cirrosus, M. intermedius, and M. longirostris have been isolated frequently by Lichtwardt et al. (1958) from stored corn. While their presence is of little significance from the point of view of deterioration, they have been isolated so consistently that their function in this association is worthy of more serious study, especially since many of them are present in the apparently intact kernels independently of the presence of other organisms.

*M. trigonosporus* has been isolated by Whitehead *et al.* (1948) from surfacesterilized cereal and legume seed from Alabama, South Dakota, North Dakota, Michigan, Illinois, Minnesota, Iowa, Wisconsin, and Wyoming. From their experiments they report that the fungus persists within the enveloping tissue in dormant seeds of several crop plants and that blossom inoculation establishes the fungus in the pericarp tissues. They suggest that natural infection occurs in the same way.

*M. intermedius* was first recorded as part of a complex associated with strawberry root rot.

#### Animal Pathogens

Microascus manginii, M. cinereus, M. lunasporus (sensu Jones), and M. trigonosporus have all been isolated either from dermal lesions or from cases of onychomycosis in man. Furthermore, several of the 14 species of Scopulariopsis listed by Dodge (1935) as being pathogenic on man and animals have been

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described as producing abortive perithecia; under certain conditions such species may produce ascosporic forms. Orr<sup>5</sup> has isolated several species from mammalian lung tissue in California although he does not regard them as pathogenic on lung tissue.

#### Soil Fungi

In recent publications, M. cirrosus and M. trigonosporus have been listed by Guillemat and Montégut (1956) and Routien (1957), respectively, as component organisms of the soil microflora. In recent years Emmons<sup>5</sup> has isolated several hundred strains of Microascus species from the soil, particularly M. intermedius but also M. nidicola. In studies of the desert soils of Arizona and California, Orr<sup>5</sup> has found M. trigonosporus, M. cirrosus, M. manginii, and other species. Indications are that, as in the case of many other fungi, the soil 'pool' of Microascus may be much greater than is at present supposed.

#### Coprophilous Fungi

The type isolate of *M. longirostris* was described by Zukal from mammalian dung. *M. longirostris* and *M. schumacheri* were also described from this habitat by Massee and Salmon (1901) and Hansen (1877), respectively. *M. doguetii* has been isolated from dog dung in the United States by Thaxter.

#### Methods

Initially, the cultures were grown on a variety of media including Difco corn meal agar, Difco potato dextrose agar, Czapek's synthetic medium with 20% and 3% sucrose, potato dextrose agar (P.D.A.), and corn meal agar (C.M.A.). The latter two media proved to be most suitable and were used consistently throughout the studies. The dextrose concentration of the P.D.A. was observed to have considerable effect on colony habit and 0.6% dextrose was the concentration used.

The ascospores of a number of species of *Microascus* were found to be very similar in size and shape; for comparative purposes, therefore, all photographs of ascospores were enlarged to the same degree.

# Taxonomy

The genus *Microascus* was erected by Zukal (1885, 1890), who described two species, *M. longirostris* and *M. sordidus*. The type species, *M. longirostris*, has a black, carbonaceous ascocarp with a well-developed neck. The asci, which form a firm gelatinous ball within the perithecium, deliquesce early and liberate the small, red-brown, crescent-shaped ascospores into the perithecial cavity. The ascospores are extruded at maturity as a gelatinous ball at the mouth of the perithecium. *M. sordidus* has been transferred to the genus *Petriella* by Barron *et al.* (1961) on the basis of ascospore and conidial characters.

Thus the generic concept of *Microascus* is fairly well defined from Zukal's descriptions. Emmons and Dodge (1931), Curzi (1931), and others note that a number of species belonging properly to *Microascus* have been described

<sup>5</sup>Private communication.

under a variety of generic names of which the following were transferred to Microascus by Curzi: Sphaerella schumacheri Hans., Melanospora stysanophora Matt., Peristomium desmosporum Lech., Nephrospora manginii Loub., Scopulariopsis cinerea Emilé-Weil and Gaudin, Acaulium nigrum Sopp, and Acaulium albo-nigrescens Sopp. Unfortunately, Curzi gave neither reasons for these transfers nor descriptions of the species transferred.

Fuentes and Wolf (1956a, b) do not support Curzi in the transfer of Sphaerella schumacheri and Melanospora stysanophora and regard the evidence as unconvincing. Lichtwardt et al. (1958) recently isolated a fungus from stored corn which corresponds so well with Hansen's original descriptions of Sphaerella schumacheri that they must be considered the same; this species undoubtedly belongs in Microascus. From the description of M. stysanophorus given by Mattirolo and outlined herein, the writer is in agreement with Curzi that this species should also be assigned to Microascus. A recent study by Doguet (1957) of a nonconidial isolate of this species indicated that it belongs in Microascus.

#### The Relationships between Microascus and Petriella

The genus *Petriella* is very close to *Microascus* in a number of characters. The perithecia, papillate to long-necked, are characteristically invested with hairs. The asci, as in *Microascus*, are produced at all levels within the centrum and are evanescent. The ascospores, moreover, are red-brown, frequently asymmetric, and extruded at maturity in long cirrhi. Curzi separated *Petriella* from *Microascus* principally on the setose condition of the perithecium. He regarded *Microascus* as typically lacking such hairs, and where they occur he regards them as being sparse and rhizoidal in function. The present work fails to support this distinction in that a number of species recognized as being typical *Microascus* may show an abundant development of hairs. Observations show that the setose character is also considerably affected by environment and is therefore an undesirable character for taxonomic purposes.

Barron et al. (1961) showed that Microascus and Petriella could be better distinguished on the basis of ascospore and conidial characters. Microascus has small straw-colored ascospores and Petriella larger red-brown ascospores. With the single exception of M. stysanophorus, all species of Microascus so far connected have a Scopulariopsis conidial stage. Petriella species on the other hand have Graphium and Sporotrichum as their conidial stages.

*M. stysanophorus* is of particular interest in its somewhat intermediate characters between *Microascus* and *Petriella*. It possesses a *Stysanus* conidial stage which is essentially a coremial form of *Scopulariopsis* and thus shows relationships to *Microascus*. It also has a *Sporotrichum* conidial stage typical of *Petriella*. Observations on the ascospores of this species also show that it is intermediate in character; while approximating the size range of *Petriella*, the ascospores lack the intensity of pigmentation and bulk and appear close, morphologically, to *M. schumacheri* and *M. nidicola*. *M. stysanophorus* is therefore regarded by the writers as belonging properly in *Microascus*. Schmidt (1912) recognized only one genus, *Microascus*.

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Taxonomy of the Conidial Stages

The genus *Scopulariopsis* was established by Bainier (1907) with *S. brevicaulis* (*Penicillium brevicaule* Sacc.) as the type species. This genus includes a series of forms which differ from the true *Penicillium* in spore type, conidiophore, and colony characteristics. Apparently unaware of Bainier's monograph, Sopp (1912) included essentially the same group of organisms in a new genus *Acaulium*. While *Scopulariopsis* is the prior name, *Acaulium* is of particular interest in that the ascospore phase with small carbonaceous perithecia provided with minute ostioles was described for certain species. Biourge (1923) identifies this group of organisms with *P. anomalum* and places these forms in the subsection *Anomala* of the genus *Penicillium*. Most workers agree that, as stated by Thom (1930, p. 512): "No one who has studied many strains of this group (i.e. *Scopulariopsis*) in comparison with the usual types of *Penicillium* pretends to believe in a close relationship between them."

The genus *Phaeoscopulariopsis* was established by Ota (1928) to include two dematiaceous species of *Scopulariopsis*-like fungi, *P. paisii* and *P. bestae*. The erection of this new genus to include these species of *Scopulariopsis* with dark conidia is supported by Fuentes and Wolf (1956 *a*, *b*). A new genus, *Masoniella* (=*Masonia* Smith), was established by Smith (1952*a*, *b*) with the type species *M. grisea*. This genus is regarded by Hughes (1953) as being synonymous with *Phaeoscopulariopsis*.

According to Hughes, in *S. brevicaulis* the sporogenous cells are anellophores. When young the anellophores are flask-shaped with short, almost cylindrical, necks; in older anellophores the necks are longer by virtue of the greater number of anellations. The conidia have a flattened base with a minute frill. Hughes notes that *Masoniella grisea* also produces chains of conidia through anellophores.

The conidia of both *Scopulariopsis* and *Phaeoscopulariopsis* (= *Masoniella*) are similar in shape and method of origin, being basally truncate and produced from anellophore-type conidiophores. The only significant difference between these two genera is the color of the spores. The inclusion of these species with dark conidia in a new genus *Phaeoscopulariopsis* does not, however, seem justified. Pigmentation of the spores is not, in this instance, regarded as a generic character and species which are similar in all other respects must be placed in the same genus, viz. *Scopulariopsis*, with *Phaeoscopulariopsis* and *Masoniella* to be regarded as synonyms. A review of *Scopulariopsis* is given by Raper and Thom (1949).

# Speciation within the Genus

In the taxonomy of the Ascomycetes and their related conidial stages, a number of characters have been used to distinguish species. Ascocarp, ascospore, cultural, and conidial characters have all been adjudged suitable criteria for speciation. Characters associated with sexuality are less subject to the effects of strain differences and environmental effects, and are therefore much more desirable as taxonomic criteria. In the present study, the ascospore is regarded as the fundamental taxonomic unit for speciation. Observations show that the ascospores are the most constant character, not only in that they are little influenced by substrate or environmental

conditions but also in that isolates from widely different geographical regions show good agreement in ascospore characters.

# The Systematic Position of Microascus

*Microascus* was placed by Nannfeldt (1932) close to *Ophiostoma* in the family Ophiostomataceae of the Plectomycetes. The relationships between *Microascus* and *Ophiostoma* were also appreciated by Luttrell (1951); he, however, referred them to separate families, the Microascaceae and the Ophiostomataceae, respectively, of a new order, the Microascales.

*Microascus* has been well established since Zukal characterized *M. longi*rostris and has remained a stable unit despite numerous revisions and controversies associated with related forms. *Ophiostoma*, on the other hand, has a complex history and has undergone a number of taxonomic and nomenclatorial reappraisals. Bakshi (1950) would regard *Ophiostoma* as being synonymous with *Ceratocystis*, with the latter name having priority. Although this revision has been accepted and extended by Hunt (1956), it is not yet generally accepted and the systematics of the genus are still in a state of flux. It would seem appropriate to take advantage of the stability of *Microascus* and include it in the Microascaceae of the Microascales after the fashion of Luttrell.

*Microascus* is included by Moreau and Moreau (1953), Emmons and Dodge (1931), Doguet (1957), and others in the Eurotiales on the basis of close similarities in the development and organization of the perithecial centrum. Undoubtedly the Microascales comprise a transitional group bearing close morphological resemblances to the Pyrenomycetes, yet having the primitive centrum organization typical of the Plectomycetes. While the more fundamental characters of centrum organization would certainly include these forms in the Plectomycetes, the black, carbonaceous, ostiolate, frequently long-necked perithecia are sufficient, in the opinion of the writers, to exclude it from the Eurotiales.

The basic difference between *Microascus* and *Ophiostoma* is in the centrum morphology. In *Microascus* the sterile matrix of the centrum is mycelioid whereas in *Ophiostoma* it is pseudoparenchymatous. Whether this difference is absolute is not yet known since only a few of the species of the two genera have been studied critically in this regard.

#### **Generic Diagnosis**

Microascus Zukal. Verhandl. zoolo.-botan. Ges. Wein, 35, 333-342 (1885).

Perithecia superficial or immersed, produced singly or in clusters, mostly black and carbonaceous, sometimes membranous, spherical to pyriform, papillate to long-necked, ostiolate, fringe of ostiolar hairs sometimes present, glabrous to extremely hairy; setae hyaline to dark-brown, stiff or flexuous, smooth or covered with warty incrustations; paraphyses absent, periphyses usually present; asci globose to elliptic sometimes clavate, sessile or shortstalked, disposed at all levels within the centrum, evanescent, eight-spored; ascospores nonseptate, smooth, oval, concavo-convex or plano-convex, sometimes triangular or tetrangular, disposed irregularly in the ascus, pale redbrown in mass, extruded from the mature perithecium in the form of a long reddish-brown cirrhus or as a gelatinous ball at the ostiole.

Conidial stages are present in the majority of species and belong to the form genus *Scopulariopsis*; a single exception, *M. stysanophorus*, has both *Stysanus* and *Sporotrichum* as its conidial stages.

Type species: Microascus longirostris Zukal.

## Key to the Species

1. 2.	Ascospores less than $8 \mu$ long or if longer less than $4 \mu$ broad
3.	Ascospores triangular, concave on all three sides, $3.5-5 \mu$ long1. M. trigonosporus
	Ascospores $6-7 \mu \log \dots 4$ Ascospores triangular with broadly rounded ends, colonies restricted $1a$ . M. trigonosporus var. macrosporus
	Ascospores triangular or tetrangular in planar view with tapering ends, colonies spreading2. Microascus pyramidus
	Ascospores asymmetrically convex, plano-convex, or concavo-convex
6.	As cospores 5.5 by 3 $\mu$
	Ascospores 7 by 5 $\mu$
7.	Ascospores less than two times as long as broad, usually abruptly concave
8. 8	Ascospores long and narrow, at least two and one-half times as long as broad
9.	Ascospores 5–7 $\mu$ long
9. 10	Ascospores $8-10 \mu$ long
10.	Ascospores more convex on one side than the other, conidial stages Stysanus and
11.	Sporotrichum    7. M. stysanophorus      Conidial stage lacking    8. M. intermedius
11.	Conidial stage present
12.	Ascospores less than $3.5 \ \mu$ long
13.	Colonies white
	Colonies other than white
14.	Ascospores not guttulate, conidia globose
15.	Colonies brown, ascospores heart-shaped to broadly lunate, conidia striate
15.	Colonies some shade of gray, ascospores variable, more often as broad as long, perithecia smooth

## **Species Descriptions**

# 1. Microascus trigonosporus Emmons and Dodge. Mycologia, 23, 313–331 (1931). (Figs. 7, 23–27)

Perithecia black, carbonaceous, glabrous or with scattered hairs, flaskshaped, with spherical base, 125–250  $\mu$  in diameter; neck up to 250  $\mu$  long, cylindric or tapering, sometimes swollen at the trumpet-like tip, smooth or with well-marked protuberances giving a rough outline; asci subglobose to ovoid 6–9 by 9–12  $\mu$ , sessile; ascospores triangulate in planar view, concave on all three sides, 2.5–3.5 × 3.5–5.5  $\mu$ , broadly rounded at the ends, redbrown in mass.

Conidial stage *Scopulariopsis*; vegetative hyphae hyaline to dark-brown, anastomosing to form "ropes", funiculose habit more pronounced in some

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isolates; conidiophores mostly long and narrow,  $10-15 \times 2-3 \mu$ , broadest between the center and the base, tapering towards both ends, borne singly or in whorls, occasionally branching and approaching a penicillioid habit; conidia variable in size and shape, globose, subglobose, ovoid, or lemonshaped, narrowly truncate at the basal end, rounded to papillate at the apical end,  $2.5-3.5 \times 3.5-5.5 \mu$ .

*Collections.*—From dead standing tree of *Abies balsamea*, Nobleton, York Co., Ontario, by R. F. Cain, July 1, 1957 (TRTC 33450). From onion seeds, Gilroy, California, by J. W. Groves, 1942 (TRTC 33050). From mouse in Arizona by C. W. Emmons, 1941 (NRRL A 6901). From culture collection Peoria, Illinois, as from B.O. Dodge, Puerto Rico (NRRL 1570). From skin lesion, Puerto Rico, by B. O. Dodge (ATC 10131). Received from G. Smith, London, England, as from J. T. Duncan, 1948 (BB22A). From seed of Sumac sorghum, grown in Hodgeson Co., Kansas, by C. T. Rogerson, 1954 (4775a). From desert soil, California, by G. F. Orr (2). From soil, collected in El Salvador, by J. B. Routien (L M928). From rat dung collected in Geary Co., Kansas, by T. E. Brooks, March, 1948 (1102).

Culturally this species shows the greatest variation of any species of *Microascus* so far studied; wide differences occur between isolates from different substrates and geographical locations. Growth may be restricted or spreading, zonate or azonate, regular or irregular in outline. Colonies from different isolates also vary considerably in color, from dirty white through light-gray to dark-gray; some distinctly brownish. Conidial production varies considerably from sparse to abundant.

Morphological variations between isolates are also in evidence. In some strains the perithecia are extremely long-necked while in others they are papillate to short-necked. Ascospores may be equilateral or distinctly longer on one side. Conidia show variability both within and between isolates; in some isolates they may be mostly globose while in others mostly lemonshaped.

Despite the variations shown by these strains in both morphological and cultural characters they all have the same basic similarity in triangulate ascospores and are considered merely as different facets of the same species.

1a. M. trigonosporus Emmons and Dodge var. macrosporus Orr var. nov. (Fig. 5)

Ascosporis 5–6.5  $\times$  5.5–7.5  $\mu$ ; conidiis 4–5  $\times$  5–7  $\mu$ .

This strain is basically the same as M. trigonosporus in many of its characters but is distinct in having ascospores and conidia which are half as large again in linear dimensions. The ascospores measure 5-6.5  $\times$  5.5-7.5  $\mu$ with the asci being proportionately larger. The perithecia of this large-spored strain are similar in size and shape to the normal. Conidia measure  $4-5 \times 5-7\mu$ and are narrowly truncate at the base and rounded to papillate at the apical end. Conidiophores are narrow and out of proportion to the conidia which they bear, being  $2-3 \mu$  broad and cylindrical to irregularly flask-shaped. Certain conidiophores and some of the conidia appear abortive and devoid of protoplasmic contents. The vegetative hyphae exhibit vesicular swellings in numerous places along their length.

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*Collection*.—From desert soil, California, by G. F. Orr and received as culture No. 4.

Colonies show a restricted growth on P.D.A.; at first white, later turning brownish-gray with the production of conidia, particularly around the perimeter of the colonies. Colonies are raised in the center with radial wrinkling of the agar. Perithecia are produced abundantly; the central regions turn black with perithecia and later red-brown with the extrusion of numerous cirrhi.

## 2. Microascus pyramidus Barron and Gilman sp. nov. (Fig. 8)

Peritheciis pyriformibus vel subglobosis, 125–250  $\mu$  diam., nigris, carbonaceis, raro pilosis: collis 100–200  $\mu$  longis, cylindraceis, asperatis, pilosis regione circumdata ostiolis praeditis. Pilis hyalinis vel pallide brunneis, 100  $\mu$  longis, levibus, septatis. Ascis subglobosis, 9–12  $\times$  13–18  $\mu$ . Ascosporis triangulis vel quadrangulis, pallide aureo-flavis, per saturam rubro-brunneis, 5–6.5  $\times$ 5.5–7  $\mu$ . Conidiis truncatis, 3–4  $\times$  4.5–5.5  $\mu$ , pallide cano-brunneis.

Perithecia flask-shaped with a spherical base,  $125-250 \ \mu$  in diameter, black, carbonaceous, sparingly setose; neck  $100-200 \ \mu$  long, cylindrical, frequently trumpet-shaped at the top with a tuft of ostiolar hairs, rough in outline; setae hyaline to dilute-brown colored, up to  $100 \ \mu$  long, smooth, septate; asci subglobose to ovoid,  $9-12 \ \times 13-18 \ \mu$ ; ascospores triangular or tetrangular in planar view, frequently with thickenings on the sides extended to points to give the ascospores a rough diamond shape with concave sides narrowly tapering to rounded ends, pale golden yellow, red-brown in the mass,  $5-6.5 \ \times 5.5-7 \ \mu$ .

Conidial stage *Scopulariopsis;* vegetative hyphae anastomosing to form ropes; pale to dark-brown pigmented; conidiophores borne on strands and hyphae, singly or in groups, rarely approaching a penicillioid habit; conidiophores long and narrow, broadest at the center, tapering towards both ends, sometimes broadest near the base and flask-shaped; vegetative hyphae and conidiophores frequently finely roughened; conidia markedly truncate, papillate, or rounded at the apical end, frequently with a distinct waist,  $3-4 \times 4.5-5.5 \mu$ , pale gray-brown.

*Collection.*—Isolated once from desert soils in California by G. F. Orr and received as culture No. 12.

On P.D.A., colonies reach a diameter of 4–5 cm in 4 weeks. Growth is strongly funiculose with hyphal strands being orientated in a radial direction. Colonies gray to violaceous gray, turning brownish in age, with a raised button in the center, and accompanied by radial wrinkling of the agar. A clear exudate is produced, particularly near the center. Perithecia are produced abundantly, at first in the aerial hyphae and later in and on the agar around the perimeter of the colonies.

This species is closest to M. trigonosporus var. macrosporus from which species it is distinguished by the distinctive ascospores, the smaller conidia, and the growth habit.

3. Microascus desmosporus (Lech.) Curzi. Boll. staz. patol. vegetale, Roma, 11, 60 (1931)

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# Syn. Peristomium desmosporum Lechmère. Bull. soc. mycol. France, 39, 303-331 (1913)

Perithecia dark, membranous, spherical in shape,  $160-200 \ \mu$  in diameter, papillate, glabrous to slightly hairy; asci ovoid, evanescent,  $6-7 \ \times \ 13-14 \ \mu$ ; ascospores ovoid,  $4-5 \ \times \ 3 \ \mu$ , slightly pointed at the extremities, reddishbrown in color, extruded at maturity of the ascocarp in the form of a long cirrhus.

Two distinct forms of this species were described by Lechmère. The first of these produces abundant conidia of the *Scopulariopsis* type; the second lacks a conidial stage but produces numerous thick-walled chlamydospores.

Conidial form.—Conidial stage belonging to Scopulariopsis; vegetative hyphae hyaline to lightly pigmented, bearing conidiophores along their length; conidiophores produced singly or in whorls, mostly 5–10  $\mu$  long, cylindrical or more often flask-shaped; conidia with a well-marked truncate basal end and rounded or papillate at the apical end,  $3.5-4.5 \times 2.5-3.5 \mu$ , pale gray in color.

Chlamydospore form.—Vegetative hyphae mostly subsurface, at first white, later acquiring a blackish color; certain cells enlarging, becoming thick-walled, and rounding off to form chlamydospores; chlamydospores dark-brown colored, terminal or intercalary, single or in chains, measuring on the average  $8 \times 5 \mu$ .

*Collections.*—From the Ivory Coast, Africa, by F. Lechmère. No other recording of this species has been noted.

Peristomium desmosporum was described as a new genus and species by Lechmère (1913), who associated his isolates with both a Verticillium and an Oidium imperfect stage. Curzi (1931) and Emmons and Dodge (1931) noted correctly that what Lechmère figured as a Verticillium might well be a Scopulariopsis, since the figures show the conidia arising in short chains. According to modern terminology what Lechmère describes as oidia must be regarded as chlamydospores. The type cultures no longer produce ascospores, but from the descriptions of the ascosporic phase as given by Lechmère, there is little doubt that Curzi was correct in transferring the genus to Microascus.

Only two species of Microascus have been described with ovoid ascospores, the other being M. niger. M. desmosporus is readily distinguished from the latter species in having smaller ascospores and a distinct conidial stage.

4. Microascus niger (Sopp.) Curzi. Boll. staz. patol. vegetale, Roma, 11, 60 (1931)

Syn. Acaulium nigrum Sopp. Videnskapsselskapets-Skrifter. I. Mat.-naturv. Kl. 11 (1912)

"Colonies brown toward black, color potato and gelatine blue-black, in gelatine cultures remaining long-submerged in the liquefied brown mass, ultimately producing conidial areas about the margin and over most of the surface which is irregular or rough and wrinkled; in very old colonies successive new growths overlay each other to form deep masses; hyphae delicate upon the usual media almost *Steptothrix*-like; conidiophores often wanting, occasion-

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ally present and *Penicillium*-like, especially in young cultures upon potato, branching at the summit one, two, or three times to produce a tangled often winding mass of sterigmata, bearing long chains of conidia; sterigmata frequently sessile on mycelial hyphae, especially in old cultures; conidia large, irregularly echinate, angular, almost polygonal, thick walled, at first violet brown then various shades of red, chocolate, and coffee brown to black-brown,  $7-8 \mu$  in diameter; perithecia produced most readily on potato, at first olivegreen, then black, sunk in the mycelium, with definite ostiole; ascospores eight to the ascus, smooth, oval, almost sharp-pointed, brown,  $7 \times 5 \mu$ ."

"The species was parasitic upon insect larvae especially *Gastrophacha pini* and recoverable from earth samples in the infected area; it was reported from various places in Norway; upon the larvae, colonies form a thin close woven mycelial layer, which is densely covered with sessile sterigmata."

The above description is from Thom's monograph (1930, p. 534) of the *Penicillia*. The species as described is closest to M. *desmosporus* but differs in having large ascospores and a distinct conidial phase.

Curzi (1931) refers to this as a new species but gave no validating description. He did, however, refer to the earlier descriptions of *Acaulium nigrum* Sopp given by Sopp (1912) and Thom (1930) which include perithecia and ascospores. The species is therefore considered to be validly published in respect to the perfect stage. It seems, therefore, more reasonable to regard it as a transfer by Curzi rather than as a new species. *Acaulium* is usually regarded as a synonym of *Scopulariopsis* but Sopp did include several species for which he reports perithecia with well-defined ostioles. In addition to *A. nigrum* he described *A. albo-nigrescens* Sopp and *A. flavum* Sopp.

5. Microascus nidicola Massee and Salmon. Ann. Botany, **15**, 313–357 (1901). (Figs. 11, 33, 41)

Perithecia black, carbonaceous, glabrous, nearly spherical in shape, 75–150  $\mu$  in diameter, with a very short cylindrical neck; asci ovoid to clavate, sessile, 10–17  $\times$  5–8  $\mu$ ; ascospores long and narrow, 6.5–8  $\times$  1.5–2  $\mu$ , plano-convex to markedly concavo-convex with rounded ends, pale straw-colored. No conidial stage known.

*Collections.*—From a disinterred wasp's nest, Royal Gardens, Kew, England, by G. Massee and E. S. Salmon. From *Dipodomys merriami*, Utah, by C. W. Emmons, 1955 (NRRL A 6894 and NRRL 6895). From soil in Utah, by C. W. Emmons, 1955 (NRRL 6913).

On P.D.A. the colonies are restricted, reaching a diameter of only 2–3 cm in several weeks. At first white, they rapidly turn black with the production of numerous perithecia over the entire colony save for a narrow white fringe of sterile floccose hyphae. There is a raised button at the center of the colonies accompanied by radial wrinkling of the agar. Perithecia are produced in profusion, being several layers deep in older parts.

This species shows a close morphological resemblance to M. schumacheri in both the shape of the ascospores and the perithecia. In the latter species, however, the ascospores are distinctly larger, guttulate, and less markedly concave. None of the three cultures of M. nidicola examined by the writers

produced a conidial stage, nor was one described by Massee and Salmon in the original paper.

6. Microascus schumacheri (Hans.) Curzi. Boll. staz. patol. vegetale, Roma, 11, 60 (1931). (Figs. 12, 40)

Syn. Sphaerella schumacheri Hans. Videnskab. Medd. Kopenhagen, 1876, 37-71, 207-354 (Jan. 1877)

Rosellinia schumacheri (Hans.) Sacc. Sylloge Fungorum, 1, 276 (1882)

Perithecia black, carbonaceous, 150–300  $\mu$  in diameter, nearly spherical with very short truncate necks, glabrous or covered with scattered hairs; asci ovoid, sessile, 13–20  $\times$  7–9  $\mu$ ; ascospores, long and narrow, 7.5–9  $\times$  2.5–4  $\mu$ , plano-convex to slightly concavo-convex with rounded ends, guttulate with numerous small oil droplets, pale straw-colored, reddish-brown in mass. No conidial stage known.

*Collections.*—From the dung of rabbits, rats, and mice in Denmark by E. C. Hansen, 1876. From stored corn, Ames, Iowa, by G. L. Barron, 1956 (531, 532, 533). From decaying hay, Nashville, York Co., by R. F. Cain, June 2, 1956 (TRTC 33463).

*M. schumacheri* grows very poorly on all media tried. On C.M.A. a stromatic-like crust is formed reaching a diameter of 1/2-1 cm in 4 to 6 weeks. Perithecia are produced in abundance in the crustose colony. Vegetative hyphae are irregular, extremely vesiculose along much of their length, and contain numerous oil globules. No conidial stage is found associated with any of the isolates obtained.

The isolates from corn differ from Hansen's original descriptions in having larger setose perithecia and slightly smaller ascospores. The relationship of this species to M. *nidicola* is discussed under the latter species. M. *schumacheri* is close to M. *stysanophorus* but is distinct in having narrower, slightly concavo-complex ascospores.

7. Microascus stysanophorus (Matt.) comb. nov. (Fig. 13)

Syn. Melanospora stysanophora Mattirolo. Atti Reale accad. sci. Torino 21, 273–282 (1886); Nuova giorn. botan. ital. 18, 121–154 (1886)

Microascus stysanosporus Curzi. Boll. staz. patol. vegetale, Roma, 11, 60 (1931)

Perithecia dark-brown to black, short-necked, spherical to pyriform, 180–200  $\mu$  in diameter; asci ovoid; ascospores yellowish, transparent, more convex on one side, 9–10  $\times$  5–6  $\mu$ .

Conidial stages belonging to the form genera Stysanus and Sporotrichum; vegetative hyphae branching freely, producing numerous conidia on short conidiophores of the Sporotrichum type; conidia acrogenous, hyaline, borne singly on short spicules, wedge-shaped,  $9-12 \times 5-6 \mu$ . Stysanus stage produced readily; lateral-growing branches perpendicular to the surface, becoming corticated to form coremia; coremial hyphae branch profusely at the top to produce an elongate sporiferous head; conidia hyaline, lemon-shaped, borne in chains. In some cases conidia borne directly on the vegetative hyphae as in Scopulariopsis.

This species was redescribed by Gueguen (1903), who noted that the peri-

the cial neck may be one-quarter the length of the base, sometimes contorted, and frequently with a fringe of ostiolar setae. Gueguen found both small  $(7-8 \times 4 \mu)$  cinnamon-brown ascospores and larger  $(10 \times 5 \mu)$  paler ascospores, variation in size being dependent on the position of the perithecium in the colonies. He noted the *Stysanus* conidia measured  $9 \times 6 \mu$ .

Two isolates were received as M. stysanophorus from Mme. Nicot of the Laboratoire Cryptogamie, Paris. The original source of these isolates is not known; it is assumed that they must have possessed Stysanus and Sporotrichum conidial stages when first isolated. These forms agree very well with the descriptions and figures of Gueguen for perithecial and ascospore characters. Neither produced a conidial stage. Culturally these isolates were very similar to M. schumacheri and M. nidicola in growth rate and colony habit, being somewhat restricted on P.D.A. The ascospores agree with other reports for M. stysanophorus but frequently spores were found up to 12.5  $\mu$  long.

*M. stysanophorus* is of particular interest because of its intermediate position between *Microascus* and *Petriella*. It has a *Stysanus* conidial stage, which being a coremial form of *Scopulariopsis* would relate it to *Microascus*. Its *Sporotrichum* conidial stage, however, is indicative of the *Petriella* group. While its ascospores are the largest of the *Microascus* group, they are palecolored distinguishing them from the darkly pigmented *Petriella* type. There are, however, conflicting reports on this character and ascospores have been reported as cinnamon-brown for this species.

The character and position of *M. stysanophorus* would merit further study but at present it seems to belong properly in the genus *Microascus* and together with *M. schumacheri* and *M. nidicola* to represent a distinct series within the genus.

This species is similar to M. schumacheri but differs in having a longer perithecial neck, larger ascospores, and a conidial stage. This species remains confused as to its limits and deserves further study.

Curzi (1931) actually used the names *Melanospora stysanospora* Matt. and *Microascus stysanosporus* (Matt.) Curzi.

# 8. Microascus intermedius Emmons and Dodge. Mycologia, 23, 313-331 (1931). (Fig. 10)

Perithecia black, carbonaceous, spherical to pyriform,  $75-150 \mu$  in diameter, mostly papillate, sometimes with a short neck, glabrous; asci spherical to ovoid,  $5-8 \times 10-13 \mu$ ; ascospores straw-colored, plano-convex to concavo-convex, with rounded ends,  $2-3 \times 4.5-6 \mu$ . No conidial stage known.

Growth is restricted on all media used; colonies reach a diameter of 1.5-2.5 cm in 4 weeks on C.M.A. Vegetative growth is mostly subsurface with little aerial mycelium. Perithecia are produced abundantly over the colonies scattered or in concentric rings.

Ten isolates of this species have been examined in culture and the characters as outlined above are very constant. No conidial stage has been described for this fungus and it seems unlikely that such a stage exists. In ascospore dimensions it approaches *M. cinereus*, from which it is distinguished by its narrower, more convex, paler, ascospores and by the absence of a conidial stage.

Can. J. Bot. Downloaded from www.nrcresearchpress.com by UNIV WINDSOR on 11/14/14 For personal use only. *Collections.*—Isolated by mouse passage from pigeon intestines by C. W. Emmons (5490) in 1956 (NRRL A-6914). From soil, Virginia, by mouse passage by C. W. Emmons (5499) in 1956 (NRRL A-6915). From soil, Utah, by mouse passage by C. W. Emmons (6062) received from Peoria as NRRL 6916. From soil, Georgia, by C. W. Emmons (5501) in March, 1953 (NRRL A-6900). From soil, Georgia, by C. W. Emmons (5510), 1953 (NRRL A-6902). From soil, New York, by C. W. Emmons (E7423), 1953 (NRRL A-6912). From stored corn, Ames, Iowa, by R. W. Lichtwardt and G. L. Barron in 1956 (521, 522, 523). From diseased strawberry roots, Charlbourn, North Carolina, by C. W. Emmons and B. O. Dodge 1930 (A.T.C.). From rat dung, Edwards Co., Kansas, by T. Brooks in March, 1948 (1055).

 Microascus cinereus (Emilé-Weil and Gaudin) Curzi. Boll. staz. patol. vegetale, Roma, 11, 60 (1931). (Figs. 9, 38)

Syn. Scopulariopsis cinerea Emilé-Weil and Gaudin. Archiv. méd. exptl. anat. pathol. Paris, 28, 452-467 (1919)

Perithecia black, carbonaceous, scattered or crowded, globose, 100–250  $\mu$  in diameter, papillate, glabrous, or with scattered hairs; hairs septate, pigmented, encrusted with wart-like protuberances; asci sessile, mostly elliptical, sometimes globose, rarely clavate, 10–14  $\times$  7–10  $\mu$ ; ascospores liberated early into the central cavity, plano-convex to slightly concavo-convex with rounded ends, pale red-brown, 5–6.5  $\times$  3–4  $\mu$ .

Conidial stage *Scopulariopsis;* conidiophores arising from the vegetative hyphae singly or in groups, frequently in a complex penicillioid arrangement; funiculose habit strongly marked with hyphal ropes bearing numerous conidiophores along their length; conidiophores broader at the center, tapering towards the extremities, usually  $6-12 \mu$  long, sometimes almost obsolete; conidia borne in long chains, markedly truncate at the basal end, obtusely rounded at the the tip, sometimes papillate,  $3.5-5 \times 3-4 \mu$ , pale grayish-brown in color.

*Collections.*—From oat seed variety Nemaha, grown in Kingman Co., Kansas, 1950, by C. T. Rogerson, 1953 (0–44–4). From oat seed variety Neosho, grown in Barton Co., Kansas, 1951, by C. T. Rogerson, 1953 (0–52–1). From oat seed variety Nemaha, grown in Cloud Co., Kansas, 1952, by C. T. Rogerson, 1953 (0–110–2). From stored corn, Iowa, by R. W. Lichtwardt, G. L. Barron, and L. H. Tiffany, 1956 (501, 502, 503, 504, 505, 506, 507). From infected big toe, France, 1919, by P. Emilé-Weil and L. Gaudin. From chromoblastomycosis infection in association with *Hormodendrum pedrosi*, Cuba, in 1956 by C. Fuentes and F. A. Wolf (631, 1885, 832).

This species grows well on P.D.A., reaching a diameter of 4-6 cm in 4 weeks. Colonies are grayish to olivaceous-gray when young, becoming progressively browner in age. The funiculose habit is well marked on this medium, giving the cultures a mealy appearance. Younger colonies produce a clear exudate in the form of small droplets over the surface of the colonies and particularly towards the center. Perithecia are produced abundantly within a few weeks; obscured at the center by the thick vegetative growth, they appear as small black dots towards the outside of the colonies where the

turf is thinner. The perithecia may be scattered evenly over the colony but are frequently produced in concentric rings.

Certain isolates of this species fruited on the cotton wool plugs of the culture tube. The perithecia produced under these circumstances tend to be long-necked, with the necks variously bent and misshapen. Perithecia have never been observed to form necks under normal cultural conditions.

*M. cinereus* is one of the most common species in the genus. It is of wide geographical distribution and substrate range. All cultures examined conform well with the description given above. Strains from quite different substrates and locations are fairly constant in ascosporic, perithecial, and conidial characters.

The ascosporic phase of this species was erroneously connected to *Hormo*dendrum pedrosoi by Fuentes and Wolf (1956), who later reassigned it as M. cinereus. As given by these workers the ascosporic dimensions are somewhat exaggerated (8.4-10  $\times$  3.2-3.8  $\mu$ ), and the range given by Emilé-Weil and Gaudin (1919) agrees more closely with the cultures examined. The possible relationship between M. cinereus and M. lunasporus is discussed under the latter species. M. cinereus closely resembles M. intermedius in ascospore shape and size; the ascospores of the latter species are somewhat slimmer, more markedly concave, and paler in color.

Since the descriptions given by Emilé-Weil and Gaudin (1919) as well as Thom (1930) refer to the perithecia and ascospores produced by this species, the simplest solution is to regard the specific name as validly published and to treat it as a valid transfer by Curzi to *Microascus*. Curzi himself wrote "sp. nov.", but unfortunately gave no validating description. He did, however, give the references to the earlier descriptions, here considered as valid for the perfect structures, even though the species was included in the genus *Scopulariopsis*.

10. Microascus longirostris Zukal. Verhandl. zoolo.-botan. Ges. Wein, 35, 333-342 (1885). (Figs. 2, 17, 22, 32, 39)

Syn. Microascus variablis Massee and Salmon. Ann. Botany, 15, 313 (1901)

Perithecia flask-shaped, with a spherical base up to 300  $\mu$  diameter, black, carbonaceous, extremely setose; neck variable in length, up to 600  $\mu$  but usually between 100 and 200  $\mu$  long by 20–30  $\mu$  broad, sometimes papillate, somewhat trumpet-shaped at the tip with a fringe of hyaline ostiolar hairs; perithecial hairs stiff, straight, numerous wart-like protuberances; asci sessile or produced in short chains, small, globose to subglobose, 8–10  $\mu$  in diameter; ascospores uniform, small, 3–4  $\times$  2.5  $\mu$ , abruptly concave on one side, dilute-brown colored.

Conidial stage *Scopulariopsis;* vegetative hyphae hyaline, anastomosing to form hyphal ropes; conidiophores borne along the length of hyphae and ropes, simple or branched dichotomously, cylindrical, straight or somewhat contorted, slightly swollen collar at the tip; conidia hyaline, well-marked truncate basal end, rounded or tapered at the apical end, sometimes with a well-marked waist,  $4-6 \times 3-4 \mu$ . This represents the first recording of the conidial stage of *M. longirostris*.

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# BARRON ET AL.: THE GENUS MICROASCUS

*Collections.*—From dry dung and rotting wood in contact with faecal masses. Germany, 1885, by H. Zukal. From dung, England, by C. Massee and E. S. Salmon in 1901 (NY). From stored corn, Iowa, by R. W. Lichtwardt, G. L. Barron, and L. H. Tiffany, 1956 (541, 542, 543, 544, 545, 546, 547). From C.B.S., Baarn, Holland, as from M. Curzi, Italy. From U.S.D.A., Peoria, as Harvard Univ. No. 51 ex. wasp's nest. From Kittery Point, Maine. 1929, by R. Thaxter (N.R.R.L. 1717). From rat dung, Edward Co., Kansas. by T. Brooks, March, 1948 (1052). From rat dung, Edward Co., Kansas, by T. Brooks, April, 1948 (1057). From Farlow Herbarium as collected by R. Thaxter (818). From dog dung, Buenos Aires, Argentina, 1906, by R. Thaxter (FH 4212). From dog dung, Cambridge, Mass., April 19, 1894, by R. Thaxter (FH). From Album graecum, Buenos Aires, by R. Thaxter, March, 1906 (NY 1518). From dog dung, Cambridge, Mass., by R. Thaxter, Jan., 1891 (NY). From pea seeds; Howick, Quebec, by J. W. Groves (42--514 A), 1942 (TRTC 33044). From pea seeds, Howick, Quebec, by J. W. Groves, 1942 (TRTC 33045). From dung, Sturgeon R., near Beardmore, Thunder Bay, Ontario, by R. F. Cain, 1949 (TRTC 33600). From bird dung SE. of Hatchley, Brant Co., Ontario, by R. F. Cain, 1949 (TRTC 33599).

*M. longirostris* grows slowly on C.M.A., reaching a diameter of 2-3 cm in 3-4 weeks. Vegetative growth is mainly subsurface with aerial hyphae scant; conidial production is largely suppressed on this medium. Perithecia begin to appear in a few weeks and are produced in abundance at first at the point of inoculation, later in concentric zones around the point of inoculation. On P.D.A. the fungus grows slowly; a floccose white mycelium is produced around the point of inoculation composed largely of concentric zones with a clear zone of subsurface hyphae around the perimeter of the colonies.

This species shows considerable variation in culture. Some isolates grow well and fruit abundantly, others may fail to fruit or sector into sterile and fertile regions. A number of cultures which fruited abundantly when first isolated lost the ability to do this after a few transfers; such cultures produced small black sclerotial-like bodies which were apparently abortive perithecia. A few isolates produced large numbers of perithecia below the surface of the agar, particularly on 20% Czapek's agar. Such perithecia were characterized by extremely long necks (up to 1 mm) and were devoid of hairs.

M. longirostris has the smallest ascospores of any species of *Microascus* so far studied. It is close to M. albo-nigrescens in ascospore shape and size but differs from this species in its smaller, less-variable, nonguttulate ascospores and its long-necked, setose perithecium.

*M. longirostris* is of special significance in that it is the type species of the genus; unfortunately neither a type culture nor herbarium specimen is available for study.

Massee and Salmon's drawings and descriptions of M. variabilis correspond well to the fungus described herein as M. longirostris. The two descriptions differ particularly in the degree of development of the neck, which, as described above, is long and setose with frequently a fringe of ostiolar setae very similar in appearance to Zukal's figure of M. longirostris, whereas Massee and Salmon describe a papillate to short-necked perithecium. Zukal, however, described his ascospores as crescent-shaped with a gelatinous sheath. No other species of *Microascus* possess a gelatinous sheath and the sheath described by Zukal may well be an artifact. If such be the case, the ascospores of the two species would be very close in shape and dimensions. Thus the isolate described herein as M. *longirostris* is neither M. *longirostris* as described by Zukal nor M. *variabilis* as described by Massee and Salmon but something of a composite of the two reflecting the opinion of the writers that the two descriptions are of the same fungus, descriptions which were essentially limited since neither of the workers studied the species in pure culture but rather from the original substrate.

11. Microascus albo-nigrescens (Sopp) Curzi. Boll. staz. patol. vegetale, Roma, 11, 60 (1931)

Syn. Acaulium albo-nigrescens Sopp. Videnskapsselskapets Skrifter. I. Mat.naturv. Kl. 11, 70-76 (1912)

Perithecia black carbonaceous, spherical to pyriform, papillate to shortnecked, 200-500  $\mu$  in diameter, with scattered setae; setae mostly short, up to 40  $\mu$  long, dark, smooth; asci subglobose to ovoid, 8-12  $\times$  6-10  $\mu$ , eightspored, walls disappearing very early; ascospores lunate with rounded ends,  $3.5-5 \times 2-3.5 \mu$ , guttulate, red-brown in mass, extruded at maturity in the form of a gelatinous ball at the mouth of the perithecium or as a long cirrhus.

Conidial stage *Scopulariopsis*; vegetative hyphae hyaline, containing numerous oil droplets; conidiophores mostly simple, sometimes branching, cylindrical or tapering from the middle to the apex, variable in length, mostly between 15 and 30  $\mu$  long, sometimes almost obsolete; conidia long and narrow, 7–9.5  $\times$  2–3.5  $\mu$ , rectangular in shape, basally truncate, hyaline; borne in short, fragile chains.

Colonies grow rapidly on P.D.A., reaching a diameter of 6–8 cm in 4 weeks. The turf is thin, white, slightly floccose, sometimes sectoring into fertile and sterile regions. Perithecia are produced within a few weeks, at first scattered, later evenly and thickly over the whole surface giving the white colonies a broad black border in age.

The above description holds for the culture number NRRL 1571 at Peoria. It was received by them as Harvard University No. 50 deposited by R. Thaxter as *M. vesparius*.

M. albo-nigrescens is close to M. longirostris but differs from it in having a short-necked, sparingly setose perithecium, larger guttulate ascospores, and a distinctive conidial stage.

*Collections.*—This species, originally described by Sopp (1912) in his monograph as *Acaulium albo-nigrescens*, was first found as a chalk-white growth upon milk in a cellar in Norway and later in a compost heap and in old goat cheese. It was one of two isolates deposited at Peoria as *Microascus vesparius* by R. Thaxter. The first of these, Harvard University No. 51, was found to be identical with *M. longirostris*; the second (NRRL 1571) is described herein. Thaxter gave no formal descriptions of *M. vesparius* and the name was apparently assigned for herbarium purposes.

The culture of this species from C.B.S. no longer produces ascospores and the vegetative growth is atypical, thus no attempt has been made to give

cultural details on this isolate. There is, however, a dried culture deposited by Dodge at the New York Herbarium from a culture of *Acaulium albonigrescens* received from C.B.S., April 14, 1931. The ascospores from this culture correspond exactly with those of Thaxter's isolate and the two are undoubtedly the same. Sopp's original description of the species as reported in Thom's monograph (1930, p. 516–517) of the *Penicillia* shows certain discrepancies from the writers', notably in the degree of development of the conidial apparatus. It may well be that Thaxter's isolate upon which our description is based has undergone some degeneration of the conidial stage similar to that of the C.B.S. isolate. Thom's description of this species is therefore included below.

"Colonies chalk-white with coremium-like white hyphal bundles bearing conidial masses either on short conidiophores or sessile, and giving a mealy white appearance followed by the progressive development of black perithecia until the whole colony appears as a coal-black wrinkled mass of black bodies with areas of white showing among them; the underside is also black at this stage; conidiophores produced typically as branches from ropes, bundles, or coremia, forming Stysanus-like columns, branching of conidiophores in several superposed verticils with elements progressively smaller in diameter; sterigmata long and tapering to a very narrow apex at the conidium-bearing tube, from which arise long diverging conidial chains; conidia white, recorded as  $10 \times 10 \mu$ . but figured as narrowly elliptical, possibly  $10 \times 5 \mu$ ; which become globose in germinating; perithecia arising as characteristic coils of hyphae involving the tips of several adjacent branches; olive-green then coal-black produced in great abundance ripening in several weeks and extruding their ascosporic masses through fine pores as a pale coffee brown powder (ascospores) covering of the whole surface; some perithecia are superficial, others immersed in mycelial masses; as cospores about  $6 \times 4 \mu$ , fairly thick-walled, oblique or concave on one side, brown, 8 to the ascus, extruded in slimy masses from the perithecia, and germinating slowly."

Sopp (1912) included a description of the perfect stage.

12. Microascus manginii (Loub.) Curzi. Boll. staz. patol. vegetale, Roma, 11, 60 (1931). (Figs. 6, 30)

Syn. Nephrospora manginii Loub. Thèses présentées à la Faculté des Sciences de Paris, Ser. A, No. 982, 1–94 (1924)

Scopulariopsis albo-flavescens Zach. Osterr. Botan. Z. 82, 173–186 (1934) Perithecia spherical, dark-brown to black, carbonaceous, papillate, glabrous, 100–175  $\mu$  in diameter; asci spherical to ovoid, sessile, 8–12 × 12–16  $\mu$ ; ascospores uniform in size and shape, almost as broad as long, somewhat heart-shaped, 4–6 × 5–6  $\mu$ , markedly concave, pale red-brown.

Conidial stage *Scopulariopsis*, vegetative hyphae hyaline, bearing numerous conidiophores; conidiophores long and cylindrical, simple or sparingly branched, mostly  $10-20 \mu$  long; conidia large, thin-walled, hyaline, globose to subglobose, markedly truncate and sometimes with a waist at the basal end, rounded to papillate at the apical end,  $5-6 \times 6-8 \mu$ .

Colonies grow rapidly on P.D.A., reaching a diameter of 5–6 cm in a few weeks; colony white, mycelium of which becomes thicker and exudes droplets.

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of amber or colorless fluid at the surface. In age, colonies appear slightly yellowish and mealy. Some isolates show a sparse production of conidia, most of the vegetative growth being subsurface and conidial production restricted to the center. Perithecia are produced abundantly and tend to be zonate in concentric rings.

Collections.—From diseased skin, Austria, by F. Zach, 1934. From cheese, France, by E. Lechmère, 1924. From desert soil, California, by G. F. Orr (7). A culture with well-developed but abortive perithecia was received in June, 1957, from G. Smith, London, as from soil. The conidial characters are so distinctive that the writers have no hesitation in assigning this isolate to M. manginii.

*M. manginii* is readily distinguished from all other species by the uniform heart-shaped ascospores, the papillate perithecium, and the distinctive conidial phase. The species was originally described by Loubière (1924) in the genus *Nephrospora*.

13. Microascus doguetii Moreau. Rev. mycol. 18, 165–180 (1953). (Figs. 3, 31)

Perithecia spherical, black, carbonaceous, 150–300  $\mu$  in diameter, covered with short hairs or nearly glabrous, usually with a well-developed neck; neck 25–150  $\mu$  long, more or less hairy; asci oval to elliptical, 10–20  $\times$  7–15  $\mu$ ; ascospores heart-shaped to broadly lunate, 5–7  $\times$  4–7  $\mu$ , with rounded ends, red-brown in the mass.

Conidial stage *Scopulariopsis;* conidiophores usually short, mostly 5–10  $\mu$  long, borne singly or in groups, cylindrical to flask-shaped, frequently contorted; conidia 4–7  $\times$  3.5  $\mu$ , truncate basal end well-marked, rounded at the apical end, brown in color, with one to five narrow bands running at an angle along the length of the conidia giving them a striate appearance.

Collections.—Found as a plate contaminant, Caen, France, by G. Doguet in 1953. On barrel bottom, Kittery Point, Maine, by R. Thaxter (837); this is probably the source of Harvard University No. 47 which is labelled M. longirostris and deposited under this name at Peoria as NRRL 1715.

Colonies grow slowly on P.D.A., reaching a diameter of 3-4 cm in 4 weeks. Growth at first mostly subsurface, showing markedly zonate habit of several concentric rings.

This species resembles M. *cirrosus* somewhat in shape of ascospores but those of M. *doguetii* are less variable and slightly larger. The shape of the perithecia and the character of the conidial stages also serve to distinguish readily between the two species. The American culture has a higher frequency of heart-shaped ascospores than the European isolate, the spores of which tend to be broadly lunate shape. The two isolates, however, agree well in other respects, particularly in the brown color of the colonies and the similarities of the conidial stages.

14. Microascus cirrosus Curzi. Boll. staz. patol. vegetale, Roma, 10, 302-310 (1930). (Figs. 4, 34, 35, 36)

Perithecia black, carbonaceous, glabrous to sparingly setose, about 100-250  $\mu$  in diameter, flask-shaped with a spherical to subglobose base and a well-

developed neck; neck somewhat swollen at the tip, measuring up to 200  $\mu$  long by 20-40  $\mu$  broad; hairs smooth, pigmented; asci globose to ovoid in shape, measuring 8-11  $\times$  10-15  $\mu$ ; ascospores variable in size and shape, 3.5-5.5  $\times$  4-6  $\mu$ , concavo-convex, sometimes irregularly angular, rarely triangular in shape, dilute brown in color.

Conidial stage *Scopulariopsis;* conidiophores arising singly or in groups, forming whorls of three to five at the end of short lateral branches, occasionally penicillioid, sometimes almost obsolete; vegetative hyphae anastomosing to form dark 'ropes' bearing conidiophores along their length; conidiophores broader at the center, narrowing towards both ends; conidia produced in long chains, basally truncate character not as well seen in this species; globose to subglobose in shape, rounded, sometimes slightly pointed at the ends, measuring  $3.5-4.5 \times 3-4 \mu$ , pale gray-brown in color.

Collections.—From decayed Prunus leaves, Italy, by M. Curzi, 1930 (C.B.S.). From stored corn, Iowa, by J. C. Gilman. From soil, France, by J. Guillemat and J. Montegut, 1956 (AS75). From seed of sorghum variety Atlas grown in Jewell County, Kansas, during 1951, isolated by C. T. Rogerson, 1954 (4821A). From oat seed grown in Reno County, Kansas, during 1952, isolated by C. T. Rogerson, 1953 (0-94-4). From flax seed, Ottawa, Canada, by J. W. Groves (43-668D), February 26, 1944 (TRTC 33047). Received from Mme. Nicot, Laboratoire de Cryptogamie, Paris (T7). From desert soil, California, by G. F. Orr (1).

This species grows rather slowly on C.M.A., reaching a diameter of 3-4 cm in a few weeks at room temperature. At this time the colonies are dark olivaceous-gray in color, turning to olivaceous-brown and finally brown in age. Young cultures exhibit a clear exudate as small droplets over the surface, particularly around the perimeter. Colonies have a mealy appearance due to the funiculose habit of the growth, and may be markedly zonate, uniform, or sector into light (conidial) and dark (nonconidial) areas. Older colonies become dotted with small white tufts of floccose sterile hyphae. Perithecia are produced abundantly in a few weeks, either scattered evenly over the surface, in concentric rings, or confined to the nonconidial sectors of sectoring isolates. Ascospores may be produced in such abundance as to give a reddish tinge to the entire colony.

On P.D.A. the colonies have a much thicker turf with the funiculose habit more strongly pronounced. The colonies have a raised button in the center and frequently cause radial wrinkling of the agar. Culturally this species is very close to M. *cinereus* but differs in the darker color and less-stable habit.

 $M.\ cirrosus$  shows a wide variability in ascospore shape and size within and between isolates; many of the ascospores are irregular and some approach the triangulate condition of  $M.\ trigonosporus$ . The species as outlined above would cover an assemblage of forms basically similar in that their variable ascospores cover the same range of size and shape but differing in the proportions of the various types.

#### Excluded Species

Microascus lunasporus Jones. Mycologia, 28, 503-509 (1936)

"On Sabauraud's medium forming a smooth whitish colony, becoming

grayish and mealy as conidia develop, then black with the formation of ascocarps, becoming wrinkled and raised above the surface of the agar; mycelium of branched septate hyphae  $2-3 \mu$  in diameter; no growth on dextrose - tartaric acid media. Conidia produced directly on the mycelium, or on simple or branched conidiophores with sterigmata 5-12  $\mu$  long; conidia oval to lemon shape, with a collar at the base,  $2-4 \times 4-7 \mu$ . Perithecia developing abundantly on Sabauraud's medium and in Knop's solution, 175–300  $\mu$ in diameter, beaked and with a papillate ostiole, the wall consisting of an outer layer 5 to 8 cells thick, with heavily carbonized walls, and an inner layer of thin-walled colorless cells; asci oval, 7-12  $\times$  7-14  $\mu$ , irregularly distributed, eight spored, deliquescing at an early stage; spores lunate,  $4-7 \times 8-14 \mu$ , smooth, extruded in light reddish-brown cirrhi 30-50  $\mu$  in diameter and reaching a length of one mm."

Type culture isolated from an infection on a human hand. Slides from subcultures deposited at the New York Botanical Garden.

The ascospore measurements as given by Jones (1936) are apparently in error. Measurements of the ascospores on the type slides show them to be  $2.5-3.5 \times 4.5-6.5 \mu$ . This would bring the species into the range of M. *cinereus* to which it appears similar in many respects. Unfortunately, there is not enough material on the type slides to come to a definite conclusion regarding this, and the species must be considered doubtful. An isolate received from Peoria as *M. lunasporus* failed to produce ascospores in culture and its vegetative growth was atypical. This isolate was of little value in establishing the possible validity of Jones' species.

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NOTE: Figures 1-42 follow.

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FIGS. 1-8. Ascospores of Microascus spp.: 1, M. albo-nigrescens; 2, M. longirostris; 3, M. doguetii; 4, M. cirrosus; 5, M. trigonosporus var. macrosporus; 6, M. manginii; 7, M. trigonosporus; 8, M. pyramidus.

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FIGS. 9-13. Ascospores of Microascus spp.: 9, M. cinereus; 10, M. intermedius; 11, M. nidicola; 12, M. schumacheri; 13, M. stysanophorus. FIGS. 14-16. Ascospores of Petriella spp.: 14, P. setifera (syn. M. setifer); 15, P. guttulata; 16, P. sordida (syn. M. sordidus).

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PLATE III



FIGS. 17-22. *M. longirostris:* 17, perithecium; 18, top of perithecial neck showing ostiolar setae and remnant of cirrhus; 19, ascospores; 20, test tube slant showing normal perithecia on agar surface and abnormal submerged forms; 21 and 22, *Scopulariopsis* conidial stage.

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FIGS. 23–27. *M. trigonos porus:* 23, perithecium; 24, ascospores; 25 and 26, conidiophores; 27, conidia.

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FIGS. 28-33. Four-week-old cultures of Microascus spp.: 28, M. trigonosporus; 29, M. albo-nigrescens; 30, M. manginii; 31, M. doguetii; 32, M. longirostris; 33, M. nidicola. Barron et al.—Can. J. Botany



FIG5. 34 and 35. Conidiophores of *M. cirrosus*. FIG. 36. Perithecium of *M. cirrosus*. FIG. 37. Perithecia of *M. schumacheri*. FIG. 38. Conidiophore, *M. cinereus*.

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FIGS. 39-42. Original figures of *Microascus* spp.: 39, *M. longirostris* after Zukal; 40, *M. schumacheri* after Hansen; 41, *M. nidicola* after Massee and Salmon; 42, *M. variabilis* after Massee and Salmon.

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