# Persoonia Vol. 15 - 1992-1994

## CONTENTS

Arnolds, E.: Notulae ad Floram agaricinam neerlandicam. XX. A revision of <i>Dermotoma</i> (J. Lange)	
Sing. – 2	187
Aronsen, A.: Two new Mycenas of section Fragilipedes from southern Norway	531
Aronsen, A. & R.A. Maas Geesteranus: Mycena terena, a new member of section Polyadelphia from southern Norway	105
Bas, C. & A. A. R. de Meijer: Amanita grallipes, a new species in Amanita subsection Vittadiniae	
from southern Brazil	345
Bas, C. & M.E. Noordeloos: Notulae ad Floram agaricinam neerlandicam. XXII. New taxa in Maras-	0.10
miellus	351
Billekens, P.: Octospora rubens and Octospora rustica in the Netherlands (Pyronemataceae, Asco-	
mycetes)	55
nellaea	197
(Coelomycetes). III	431
Brummelen, J. van: Ultrastructure of the ascus and the ascospore wall in Scutellinia (Pezizales, Asco-	
mycotina)	129
Brummelen, J. van & M.E. Noordeloos: Editorial	1
Dennis, R.W.G. & B.M. Spooner: The fungi of North Hoy, Orkney. II	169
Gruyter, J. de & M. E. Noordeloos, M. E.: Contributions towards a monograph of <i>Phoma</i> (Coelomycetes). I – 1. Section <i>Phoma</i> : Taxa with very small conidia in vitro	71
Gruyter, J. de, M.E. Noordeloos & G.H. Boerema: Contributions towards a monograph of <i>Phoma</i> (Coelomycetes). I – 2. Section <i>Phoma</i> : Additional taxa with very small conidia and taxa with	
conidia up to 7 µm long	369
Häffner, J.: Rezente Ascomycetenfunde. XI. Sterigmate Formen in der Gattung Peziza (2. Teil)	179
Hoog, G.S. de: Some Ramularia-like fungi on Malvaceae	63
Keizer, PJ. & E. Arnolds: Taxonomical notes on macrofungi in roadside verges planted with	
trees in Drenthe (The Netherlands), I	489
Kelderman, P.H.: Lepiota cingulum spec. nov., a new species in section Stenosporae	537
Kullman, B. & J. van Brummelen: Studies on the character variability in the Ramsbottomia crec'h	
queraultii complex (Pezizales)	93
Læssøe, T.: Xylaria digitata and its allies – delimitation and typification. II	149
Maas Geesteranus, R. A.: Some Myceneae of the Himalayan foothills	33
Maas Geesteranus, R.A. & E. Horak: Mycena acrocephala, a new member of section Adonideae	
from Sikkim	341
Maas Geesteranus, R.A. & T. Læssøe: Mycena dasypus, a new member of section Polyadelphia	101
Marchetti, M.: A new species in section Hydrophilae of the genus Psathyrella	249
Noordeloos, M.E.: Studies in Clitopilus (Basidiomycetes, Agaricales) in Europe	241
Noordeloos, M.E. & V. Liiv: New taxa of Entoloma (Basidiomycetes, Agaricales) from Estonia and Karelia	23
Noordeloos, M.E., M. Tabarés & A. Rocabruna: A new species of Entoloma subgenus Pouzarella	
from Spain	123
Schild, E.: Erläuterungen zu Ramaria obtusissima und zu Ramaria schildii	109
Schn-Irlet, B.: Type studies in Crepidotus. II	155
Genia-med, D 17pc studies in Creptuonis. II	

Stijve, T.: Psilocin, psilocybin, serotonin and urea in Panaeolus cyanescens from various origin.	117
Stijve, T. & C. Blake: Bioconcentration of manganese and iron in Panaeoloideae Sing	525
Cife, C.D. & C. Das. Some new species of coprising from the recommendation	357
sion and terision of subsection first emens,	257
uon bienosporae	223
Verkley, G.J.M.: Ultrastructure of the apical apparatus of asci in Ombrophila violacea, Neobulgaria pura and Bulgaria inquinans (Leotiales)	3
Verkley, G.J.M.: Ultrastructure of the ascus apical apparatus in <i>Hymenoscyphus</i> and other genera of the Hymenoscyphoideae (Leotiales, Ascomycotina)	303
Verkley, G.J.M.: Ultrastructure of the ascus apical apparatus in <i>Leotia lubrica</i> and some Geo- glossaceae (Leotiales, Ascomyctina)	405
Books received by the Rijksherbarium Library:	541
Errata Persoonia 15 (1) (1992) 71–92:	221

## Dates of publication:

Part 1, pp. 1-128, 6 Oct. 1992

Part 2, pp. 129-256, 5 April 1993 Part 3, pp. 257-404, 6 Dec. 1993

Part 4, pp. 405-550, 23 Sept. 1994

#### PERSOONIA

Published by Rijksherbarium / Hortus Botanicus, Leiden Volume 15, Part 1, p. 1 (1992)

### EDITORIAL

With the publication of volume 15 (1) Persoonia has undergone a number of changes, both in the team of managing editors and in the lay-out.

Dr. C. Bas retired in March 1991 from his post at the Rijksherbarium, and turned over his editorial duties to his successor, Dr. M.E. Noordeloos, after finishing volume 14, part 4, the 'Festschrift' dedicated to Dr. R.A. Maas Geesteranus. Dr. Bas has been an editor of Persoonia since 1972, and the present editors wish to thank him most sincerely for his efforts to maintain and improve the standard of our journal.

The cover, typography, and lay-out of the present issue have undergone changes in order to achieve more unity of style in the journals published by the Rijksherbarium / Hortus Botanicus.

In the light of the above mentioned changes, concise information on the scope of Persoonia and instructions to the authors are given on the cover.

J. van Brummelen, M.E. Noordeloos Editors

#### PERSOONIA

Published by Rijksherbarium / Hortus Botanicus, Leiden Volume 15, Part 1, pp. 3-22 (1992)

# ULTRASTRUCTURE OF THE APICAL APPARATUS OF ASCI IN OMBROPHILA VIOLACEA, NEOBULGARIA PURA AND BULGARIA INQUINANS (LEOTIALES)

### GERARD J. M. VERKLEY

Rijksherbarium / Hortus Botanicus, P.O. Box 9514, 2300 RA Leiden, The Netherlands

The ultrastructure of the apical apparatus in asci of three species of the Ombrophiloideae (Leotiaceae) is compared. *Ombrophila violacea* and *Neobulgaria pura* show similarities in morphology and maturation pattern of the apical apparatus, suggesting a close relation between these species. In these respects and in development of the ascospore wall *Bulgaria inquinans* differs considerably from *O. violacea* and *N. pura*.

#### INTRODUCTION

The order Leotiales (Carpenter, 1988; Helotiales sensu auct.) is basically defined by the structure of the ascus. Its current classification is still largely based upon the system that Nannfeldt (1932) proposed for the inoperculate discomycetes. Students of Leotiales have disagreed on the arrangement of families for a long time now (Dennis, 1956, 1978; Korf, 1973; Barr, 1976; Hawksworth et al., 1983; Eriksson, 1983). Apparently it is difficult to create a natural system by applying the more conventional taxonomic criteria based on ascocarp anatomy, morphology of hymenial elements and ecology.

Light microscopic studies performed by Chadefaud (1964, 1973) and more recently by Baral (1987) provided valuable diagnostic data on the ascus morphology of various groups of inoperculate discomycetes. For studies on the structural variation of the ascus apex in taxa with small asci like the Leotiales the use of electron microscopy is necessary.

Bellemère (1977) made a comparative ultrastructural study of a selection of representatives of the Leotiales and proposed six types of apical apparatus. Occasionally, others studied some species as well (e.g. Corlett & Elliott, 1974; Schoknecht, 1975; Benny et al., 1978). Unfortunately these authors used different techniques and terminologies and had different ways of interpreting their electron micrographs. This has made the data less accessible to other mycologists. In order to reveal more about the variation in ultrastructure of the apical apparatus of asci within the Leotiales and its possible significance for the taxonomy of the group, a comparative study on selected species was initiated. In this first report data on three species of the Leotiaceae (Corda, 1842) are presented: Ombrophila violacea Fr., Neobulgaria pura (Fr.) Petr., and Bulgaria inquinans (Pers.) Fr.

Korf (1973) and Dennis (1978) treated Ombrophila Fr. in a restricted sense, arranging only a few species around O. violacea. Korf (l.c.) followed Gamundi & Dennis (1969)

and treated *Neobulgaria* Petrak, typified by *N. pura*, also in a restricted sense, separated from *Ascotremella* Seaver. More recently, Baral & Krieglsteiner (1985) proposed to place *Neobulgaria* in the genus *Ombrophila*. *Bulgaria* inquinans is the type species of a small genus showing a combination of characters that is quite unusual in Leotiaceae: pigmentation and size-differentiation of the ascospores within a single ascus and large ascocarp size.

The arrangement of Ombrophila, Neobulgaria, and Bulgaria together within a single tribe, the Ombrophiloideae sensu Dennis, suggests a close relationship of these genera. The classification of this tribe is largely based on anatomical features of the ascocarp, especially those concerning gelatinized tissues. Differences in other characters of the species assembled in this group are apparently considered less important. The Ombrophiloideae therefore seems to form a heterogeneous taxon and an interesting subject for a comparative study on the ultrastructure of the apical structures in the asci. Little is known about the ultrastructure of ascospore walls in the Leotiales. For this reason data concerning ascospore wall development are also included in the present paper.

### MATERIALS AND METHODS

Fresh material was collected in the field. Parts of fruit-bodies were fixed for 3 hours using 1% glutaraldehyde in 0.1 M cacodylate buffer (pH 7.2) at 4°C, washed in buffer, and postfixed for 1 hour using 1% osmium tetroxide in cacodylate buffer at room temperature. Material was then dehydrated in a graded series of ethanol and embedded in Epon. During dehydration material was stained with 1% uranyl acetate for 10 minutes. Ultrathin sections were cut using a diamond knife on a Reichert Jung Ultracut E ultratome.

For PA-TCH-SP (periodic acid-thiocarbohydrazide-silver-proteinate), sections were picked up on uncoated 200 mesh golden grids and treated at room temperature as follows (modified from Thiéry, 1967): 1) 1% aqueous periodic acid (PA), 30 min.; 2) three rinses of water, 10 minutes each; 3) 0.2% thiocarbohydrazide (TCH) in 20% acetic acid, 60 min.; 4) rinses of 10%, 5%, and 2.5% acetic acid, 5 min. each; 5) three rinses of water, 10 min. each; 6) 1% aqueous silver proteinate (SP) (Prolabo, Paris), 25 min. in the dark; 7) three rinses of water, 10 min. each.

In other cases sections were contrasted with Reynolds' lead citrate and uranyl acetate, or, occasionally, with barium permanganate. Preparations were examined shortly after treatment, using a Philips EM 300 at 60 kV.

In the following list details are given about the origin of the collections.

Ombrophila violacea Fr. Bunderbos, Limburg, the Netherlands, on soil and plant debris, April 1990, H. Huyser.

Neobulgaria pura (Fr.) Petr. Fôret de St. Prix, Bois de la Canche, Morvan, France, on decaying wood, Oct. 1990, J. van Brummelen.

Bulgaria inquinans (Pers.) Fr. Mooshof, Dungou near Roith, Ober Pfaltz, Germany, on decaying wood, Sept. 1990, J. van Brummelen 7942.

The identification of specimens was performed according to Dennis (1978).

#### TERMINOLOGY

Clarification of the terminology employed

Corresponding terms of Bellemère (1977) and Bellemère et al. (1987) are given in brackets, as [1] and [2] respectively.

Apical apparatus: apical region of ascus wall that forms the functional spore-shooting apparatus. It includes the apical thickening and the central cylinder ('dôme apical' or 'appareil apical' [1], [2]).

Central cylinder (CC): central region in the apical apparatus. It operates as a pore by tearing or breaking during ascospore discharge (it includes 'coussinet apical', 'pseudomanubrium', 'cylindre axial' [1] and 'coussinet apical', 'pseudomanubrium', 'corps ombiliqué' [2]).

Annulus (A): annular (cylindrical) structure surrounding the central cylinder, from which it can be distinguished by differences in ultrastructure. Its outer limits are usually less distinct because of a more gradual change in structure, and therefore the annulus is considered to be a part of the apical thickening ('anneau inférieur', 'anneau supérieur' [1], [2]).

Annular protrusion (AP): part of the wall material in the apical thickening that is associated with the annulus and protrudes downwards into the ascus lumen. It may enclose the apical chamber to a variable extent ('pendentif' [1], [2]).

Apical chamber (AC): amount of epiplasm enclosed to a variable extent by the annular protrusion ('oculus' [1]; 'évagination médiane du épiplasme' [2]).

Apical thickening (AT): total region of increased thickness of the ascus wall, excluding the central cylinder, in which an annulus may be embedded.

Outer layer (OL): outer part of the ascus wall that does not increase in thickness in the apical apparatus.

Inner layer (IL): inner part of the ascus wall that increases in thickness in the apical apparatus.

Apex development: proces of apex formation and maturation.

Apex formation: addition of wall material to the apical region of the ascus wall, thus forming the apical apparatus.

Apex maturation: all changes in the ultrastructure of the apical apparatus that occur after apex formation is completed.

## Stages in ascus development

Young ascus – The ascus initial is formed and elongates, meiosis and mitosis take place, resulting in (usually) eight ascospores delimited by two unit membranes. In the species under study the apex formation is completed within this stage.

Immature ascus – Ascospore maturation (Beckett, 1981b) occurs. The ascosporoplasm is enriched with organelles and lipid bodies, and the primary and secondary ascospore wall are formed. Towards the end of this stage, the various organelles decrease in number, while the number of vacuoles in the epiplasm increases. In the species under study most of the changes that characterize the apex maturation take place during this stage.

Mature ascus – Maximum vacuolization of the epiplasm is reached and no organelles remain in the epiplasm, ascospores are fully mature; the internal pressure increases rapidly, eventually leading to dehiscence.

Dehisced ascus - After the discharge of ascospores the ascus wall disintegrates rapidly.

#### RESULTS

Observations on the lateral ascus wall, the apical apparatus and ascospore wall are based on PA-TCH-SP material. Conventional staining procedures with uranyl and lead salts proved to be less suitable, because they did not provide the amount of contrast needed for a study on wall substructure. The applied procedure modified from Thiéry (1967) gave satisfactory results.

The electron micrographs presented here show the result of the uranyl staining during the ethanol dehydration series plus the specific binding of silver to the reactive (i.e. 'PAS-positive') polysaccharides in PA-TCH-SP procedure. When the periodic acid reaction step is omitted from this procedure, the binding of silver to the fungal wall is practically inhibited, indicating that levels of endogenic aldehydes or aldehydes introduced during fixation are very low. Thus, in the walls – and this can be stated for the walls only – nearly all contrast is in fact reactivity and this term will therefore be used rather than electron density.

Longitudinal median sections of young, immature, mature, and dehisced asci were studied. The lateral ascus wall and the apical apparatus are described. Some observations on the development of the ascospore wall are included.

## Ombrophila violacea

The ascus wall

The shape of the ascus apex varies from truncate-rounded to rounded. In the lateral ascus wall two layers are observed. The outer layer 65–90 nm thick consists of two strata: a very reactive outer stratum which is associated with the reactive material of an extra-ascan periascus and a moderately reactive inner stratum with a rather rough granular appearance (Fig. 1C, 2C). The inner layer 180–250 nm thick of the lateral wall has an inner part that is somewhat more reactive and varies considerably in thickness (arrow, Fig. 2D). The thickness of the inner layer increases abruptly in the apical apparatus (Fig. 1A). The outer layer does not increase in thickness here. There seems to be a decreased reactivity in it at the tip of the apparatus and, especially in the mature ascus, the outer stratum may even be partly absent (arrows, Fig. 1C).

Young ascus – An apical apparatus with some distinct characters is already present shortly after ascospore delimitation. The apical thickening, which is mainly formed by an abrupt increase of the inner layer shows a random network of very fine reactive material (Fig. 1A). The central cylinder shows a similar substructure. The narrow, very reactive but discontinuous annulus is associated with an annular protrusion surrounding an apical chamber (Fig. 1A). The greatest annular reactivity is found in the annular protrusion. The

Abbreviations used in Figures 1–9: A, annulus; AC, apical chamber; AP, annular protrusion; AS, ascospore; AT, apical thickening; AW, ascus wall; CC, central cylinder; E, epiplasm; ER, endoplasmatic reticulum; G, glycogen; IL, inner layer of ascus wall; im, investing membrane; is, inner stratum of OL; iz, inner zone of pw; L, lipid body; m, mitochondrion; N, nucleus; OL, outer layer of ascus wall; os, outer stratum of OL; oz, outer zone of pw; P, periascus; Pa, paraphysis; pw, primary wall of SW; SP, sporoplasm; SW, ascospore wall; sw, secondary wall of SW; V, vacuole; Ve, vesicle.



Fig. 1. Ombrophila violacea. Longitudinal median sections of apices in asci at different stages of development (bar equals 1 µm). A. Young ascus, shortly after ascospore delimitation; B. immature ascus, ascospore wall development approximately halfway (compare Fig. 2C); C. mature ascus.

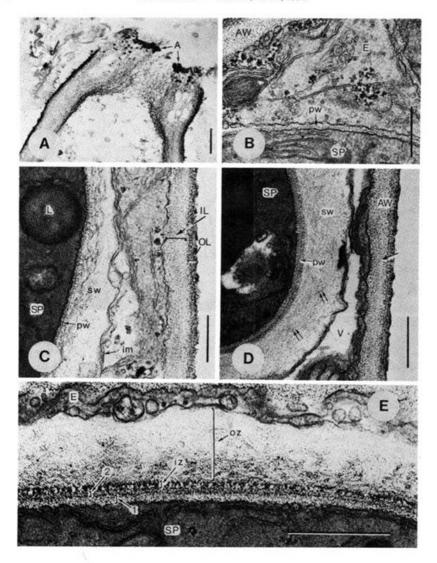


Fig. 2. Ombrophila violacea. A. Dehisced ascus (bar equals  $1~\mu m$ ); B-E. development of ascospore wall (bar equals  $0.5~\mu m$ ); B. development of primary wall; C, D. development of secondary wall; E. differentiation of primary and secondary wall.

patches of reactive material seem to spread out in the upper part of the annulus and are not found in the outer layer. Frequently some large vesicles are observed in the upper part of the apical chamber at this stage (arrows, Fig. 1A).

Immature ascus - The overall reactivity in the central cylinder slightly decreases. The boundary of the central cylinder with the epiplasm flattens (Fig. 1B). The irregularity of the boundary of the apical thickening with the epiplasm is probably caused by swelling.

Mature ascus — The apical apparatus is considerably compressed by the time the ascospore maturation is completed and the epiplasm is largely filled with vacuoles. The microfibrils in the central cylinder align in a 90° angle to the axis of the annulus. The annulus is a narrow, compact ring that seems to end abruptly on its upper side at a lower level than at the young and immature stage (Fig. 1C).

Dehisced ascus – During discharge of the ascospores the annulus is everted over an angle of approximately 90°, hence giving the ascus apex a flattened appearance. Very little remnants of the central cylinder can be found. The remaining wall material swells and disintegrates very rapidly after dehiscence (Fig. 2A).

### Ascospore wall development

At first the investing membrane and spore plasma membrane lie closely together and the first wall material appears in between these membranes shortly afterwards (Fig. 2B). This wall material is designated the primary wall. On further development the investing membrane is lifted from the primary wall by the deposition of a matrix substance. In this matrix 400–500 nm thick numerous reactive fibrils are found, at first mainly in the direct vicinity of the primary wall (Fig. 2C), later also in the outer parts of this matrix (double arrows, Fig. 2D). Finally, the primary wall (about 100 nm thick) becomes differentiated into two layers (1, 2 in Fig. 2E), while in the matrix of the secondary wall an inner zone (about 35 nm thick) of highly reactive bands is formed directly on the outer surface of the primary wall. It appears that this zone is formed from the deposition of the reactive material in the matrix (Fig. 2E). Shortly after discharge of the spores the secondary wall is still present, no longer surrounded by the investing membrane.

### Neobulgaria pura

The ascus wall

Young asci could not be observed in the material available for this study. The shape of the ascus apex shows a distinct circular depression in the apical surface just over the annulus at the immature stage (Fig. 3A). In mature asci this depression is absent and the shape is rounded with a small flattened zone just over the central cylinder (Fig. 3B). The lateral ascus wall consists of two layers. In the outer layer approximately 60 nm thick two strata are observed. The outer stratum about 15 nm thick is very reactive and gives the surface of the ascus wall a rough appearance. The inner stratum about 45 nm thick is less reactive with a granular appearance (Fig. 4B). The outer layer is already considerably disintegrated towards the apex in the immature ascus (arrow, Fig. 3B). The inner layer 150–180 nm thick shows a fine granular reactivity. Its inner two-third to half is somewhat more reactive (Fig. 4B). The inner layer gradually thickens towards the apex to form the apical apparatus (Fig. 3A, B).

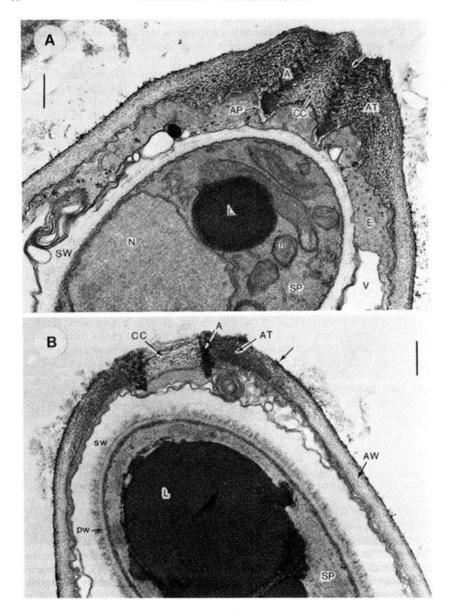


Fig. 3. Neobulgaria pura. Longitudinal median sections of apices of asci at different stages of development (bar equals  $1~\mu m$ ). A. Immature ascus; B. mature ascus.

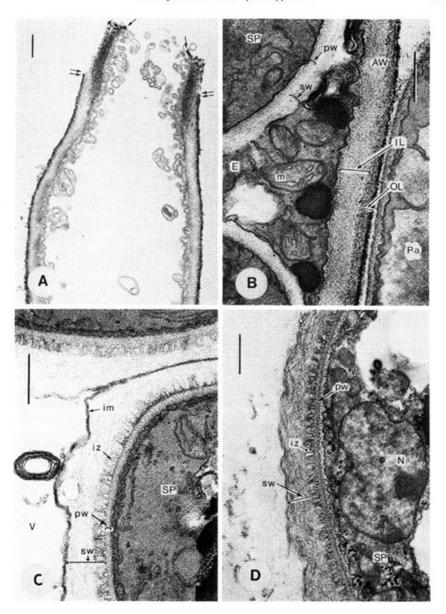


Fig. 4. Neobulgaria pura. A. Dehisced ascus (bar equals 1  $\mu$ m); B–D. development of ascospore wall (bar equals 0.5  $\mu$ m); B. development of primary and secondary wall; C. differentiation of secondary wall; D. ascospore after discharge.

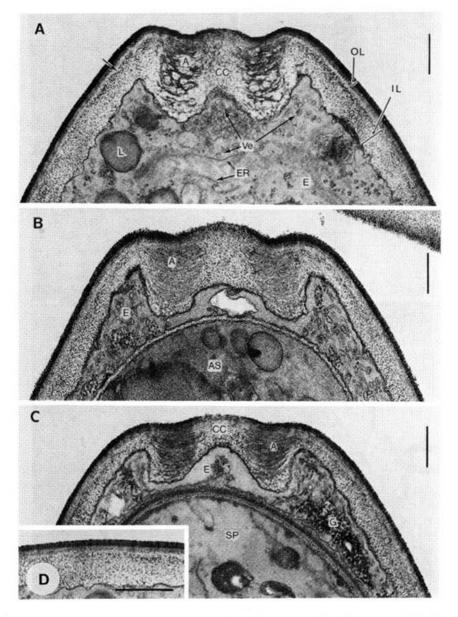


Fig. 5. Bulgaria inquinans. Longitudinal median sections of apices in asci at different stages of development (bar equals 1  $\mu$ m). A. Young ascus, before ascospore delimitation; B. immature ascus; C. as B, but more advanced state; D. detail of lateral ascus wall showing transverse microfibrils.

Immature ascus – The apical thickening which is mainly formed by a gradual increase of the thickness of the inner layer towards the apex contains several layers of very reactive material parallel to the ascus surface. This material increases in density to a variable degree in the annulus, giving it a rather irregular appearance (Fig. 3A). Also towards the inner side of the annulus the boundary with the central cylinder is less clear, because of the gradual decrease in reactivity here. The reactivity in the central cylinder is somewhat lower than in the apical thickening. Especially in the upper part of the annulus, where it increases in width the layered character is distinct (arrow, Fig. 3A), contrasting with the more compact mass of reactive material in the annular protrusion. The outer layer in the apical apparatus seems rather eroded. It does not fully cover the annulus and the central cylinder. The difference in reactivity that can be observed between an outer and inner zone of the inner layer in the subapical wall disappears in the apical thickening.

Mature ascus — When the ascospore walls are fully differentiated and the epiplasm is almost completely replaced by vacuoles the apical apparatus is strongly compressed (Fig. 3B). Furthermore changes in ultrastructure are observed: the reactive fibrils in the apical thickening lie closely together, parallel to the ascus surface. The annulus is more compact and the annular protrusion points straight downwards. The central cylinder has lost most of its reactivity and contains only a few distinctly reactive fibrils. The boundary of the central cylinder with the ascus lumen has flattened.

Dehisced ascus – In longitudinal section the inner layer points upwards surrounding a relatively large opening. The annulus is everted over an angle of approximately 90° during ascospore discharge. Its remnants are still clearly visible (arrows, Fig. 4A). The outer layer covers only the lower one-third part of the apical thickening (double arrows, Fig. 4A).

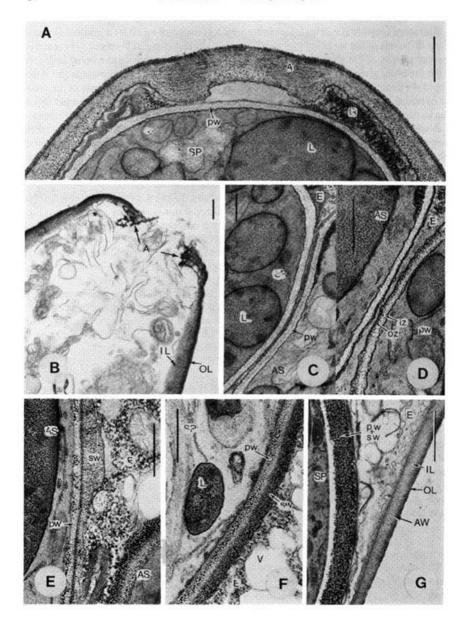
## Ascospore wall development

In the youngest asci that could be studied there is already a considerable distance between the investing membrane and spore plasma membrane. A weakly reactive granular primary wall about 90 nm thick borders the spore plasma membrane and it is surrounded by an early secondary wall consisting of a matrix with a narrow non-reactive inner zone approximately 50 nm thick and an outer zone 100-200 nm thick containing moderately reactive fibrils (Fig. 4B). Later, in the inner zone of the secondary wall bristle-like reactive fibrils are deposited perpendicularly to the outer surface of the primary wall (Fig. 4C). The reactivity in the outer zone increases. Shortly after discharge of the ascospores the secondary wall is still present, no longer surrounded by the investing membrane (Fig. 4D).

## Bulgaria inquinans

#### The ascus wall

Throughout the ascus development the circular depression in the apical surface over the annulus characterizes the shape of the ascus apex. In the lateral wall two layers can be observed. The outer layer about 120 nm thick is characterized by fibrils orientated perpendicularly to the surface of the ascus wall (particularly distinct in young and immature asci) (Fig. 5D). It seems to decrease markedly in thickness over the central cylinder, but it is difficult to find a clear boundary line with the inner layer there. The outer layer consists of a very reactive outer stratum and a less reactive inner stratum, both approximately 60 nm



thick (Fig. 5A). The inner layer 330–370 nm thick of the lateral wall shows a differentiation in reactivity in the part adjacent to the apical apparatus. There, a thin, lowly reactive outer zone and a considerably higher reactive inner zone ('strate annellogène' of Chadefaud; arrow, Fig. 5A) can be distinguished. The reactivity of the inner zone decreases again inwards.

Young ascus – The apical thickening is formed by a rather abrupt increase of thickness of the inner layer. It is almost fully occupied by a broad annulus. Strictly, there is no annular protrusion (see definitions). In the upper part of the annulus the reactive material lies in parallel layers, while towards the base it is found in patches distributed at random (Fig. 5A). In the epiplasm near the apical apparatus high concentrations of vesicles containing reactive material are found in some asci (Fig. 5A). The central cylinder shows a fine granular reactivity, much like the subapical part of the inner layer (Fig. 5A).

Immature ascus – After the ascospore delimitation has been completed more of the reactive material in the annulus becomes oriented in fine fibrillar layers (Fig. 5B, C).

Mature ascus — The annulus now consists of very fine fibrils, oriented parallel to the ascus surface. The apical apparatus is compressed (Fig. 6A). Asci at corresponding stages of ascospore maturation sometimes show different stages of apex maturation (compare Figs. 5B and C).

Dehisced ascus – After dehiscence little remnants of the central cylinder can be found near the annular material. In longitudinal section the apical thickening and annulus show no eversion or any other marked change in position (Fig. 6B).

### Ascospore wall development

In the earliest stage of ascospore wall development that could be observed wall material is deposited between the spore plasma membrane and investing membrane. This primary wall approximately 160 nm thick is of low reactivity and shows a fine granular line exactly in the middle separating two zones of about equal thickness (Fig. 6C). The outer zone soon differentiates into a layer of moderate to high reactivity (Fig. 6D). After this the investing membrane is irregularly lifted from the primary wall. In the resulting space secondary wall material 90–180 nm thick with a fine granular reactivity is deposited. In the beginning the reactivity in the secondary wall is lower than that of the outer zone of the primary wall (Fig. 6E). Some particles of reactive material are now present in the inner zone of the primary wall and soon in the secondary wall too. For some time the outer zone of the primary wall is practically devoid of these particles. The secondary wall now has a constant thickness of about 120 nm (Fig. 6F).

Finally, just before ascospore discharge the whole wall is incrustrated with these large reactive particles (Fig. 6G).

Fig. 6. Bulgaria inquinans. A, B. Longitudinal median sections of ascus apices (bar equals 1  $\mu$ m); A. mature ascus; B. dehisced ascus; C–G. development of ascospore wall (bar equals 0.5  $\mu$ m, except G, 1  $\mu$ m); C. early development of primary wall; D. advanced development of primary wall; E. development of secondary wall and differentiation of primary wall; F. advanced differentiation of primary wall and advanced development of secondary wall and accordary wall completed.

#### DISCUSSION

Although developed as a specific reaction for PAS-positive polysaccharides by Thiéry (1967), the PA-TCH-SP procedure has been used mainly for improvement of contrast in a number of studies on ascus wall ultrastructure of various taxonomic groups (Bellemère, 1977; Beckett, 1981a). Technical problems concerning the specificity of this reaction were not directly relevant to these studies.

In light microscopy, the ascus apices of *Ombrophila violacea*, *Neobulgaria pura*, and *Bulgaria inquinans* show regions blueing in Lugol's iodine or other iodine solutions (Dennis, 1978; Baral, 1987; and own observations). These regions correspond to the highly reactive annular structures observed in electron microscopy. In studies of the ascus apex one must be careful comparing details of light microscopic images with those of electron microscopic images.

The terminology used here for the description of the lateral ascus wall is in accordance with the one used in several other studies on operculate and inoperculate discomycetes (Schrantz, 1970; Griffiths, 1971; van Brummelen, 1981). Basically, the lateral ascus wall of the species under study contains an inner and outer layer in which strata can be designated. If the differentiation within the layers is not continuous in larger parts of the walls it is preferred to speak of zones. In his ultrastructural study of Helotiales Bellemère (1977) used a concept in which four layers in the ascus wall are distinguished, assuming the outer two, a and b, to correspond to the 'exoascus' and the inner two, c and d, to correspond to the 'endoascus' as described for light microscopy by Chadefaud (1973). The present results do not allow such a strict division to be utilised objectively.

A partly new terminology for the apical apparatus is introduced in the present study. It is a preliminary one and it may be adapted or extended in future studies if necessary. The terminology applied for the ascospore wall agrees with the work by Carroll (1966), Merkus (1976), Beckett (1981b), and van Brummelen (1986, 1989).

In the three species under study the apex formation, i.e. addition of wall material to the apical wall, is completed before ascospore delimitation. In a detailed study of this process in *Xylaria longipes* Beckett & Crawford (1973) described an apical body and a surrounding vesicle system. In *Bulgaria inquinans* no apical bodies were observed, but the high concentration of vesicles (Fig. 5A) resembles the vesicle system of *Xylaria longipes*.

Once apex formation has been completed, the wall ultrastructure in the apical apparatus continually changes on further ripening of the ascus. As yet little attention has been paid to the process of apex maturation. It is found that details of this process of maturation are essential for a comparative study of the Ombrophiloideae.

The relative reactivity and the interpretation of wall substructure in the apical apparatus of the asci are depicted in diagrammatic schemes (Fig. 7-9). Two patterns of apex maturation can be distinguished. In *Ombrophila violacea* and *Neobulgaria pura* the annulus condenses to a homogeneous, very reactive narrow ring occupying only a small part of the apical thickening. In *Bulgaria inquinans* the pattern is characterized by a reduced reactivity and an ordered arrangement of the microfibrils in the annulus occupying most of the apical thickening. In *O. violacea* and *N. pura* the structure of the outer layer of the ascus

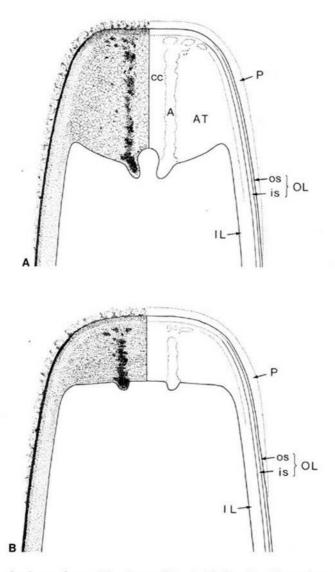


Fig. 7. Diagrammatic schemes of young (A) and mature (B) asci of *Ombrophila violacea*, demonstrating relative PA-TCH-SP reactivity on the left half and corresponding interpretation of layers on the right half of each scheme. See also the next two pages.

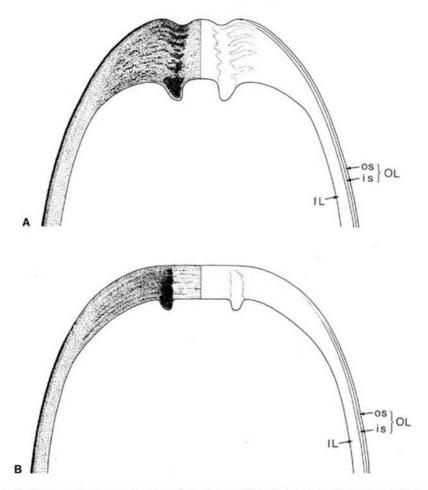


Fig. 8. Diagrammatic schemes of immature (A) and mature (B) asci of *Neobulgaria pura*, demonstrating relative PA-TCH-SP reactivity on the left half and corresponding interpretation of layers on the right half of each scheme.

wall is relatively simple, but in *B. inquinans* a rather remarkable outer layer with a transverse orientation of its constituents occurs. In *O. violacea* and *N. pura* the dehisced ascus is characterized by a 90° eversion of the annulus, while in *B. inquinans* no eversion is found. Furthermore, *O. violacea* and *N. pura* show similarities in development of the ascospore wall, while they both differ in this respect from *B. inquinans*. So, the ultrastructural data justify the conclusion that *O. violacea* and *N. pura* are closely related, while *B. inquinans* has no affinities that close to either of these two species.

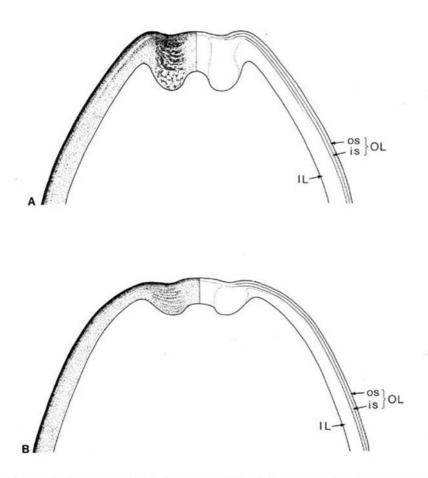


Fig. 9. Diagrammatic schemes of young (A) and mature (B) asci of *Bulgaria inquinans*, demonstrating relative PA-TCH-SP reactivity on the left half and corresponding interpretation of layers on the right half of each scheme.

The only data available on the apical ultrastructure in *N. pura* were published by Bellemère (1977). He shortly mentioned the species, stating it to be of the *Sclerotinia*-type. It is difficult to form an opinion about the substructure in the apical apparatus from the only electron micrograph shown. In the present study of *N. pura* it was not possible to clearly indicate in the apical apparatus a line of demarcation between two possible strata in the inner layer that could correspond to Bellemère's layers c and d. The eversion of the annulus during ascus dehiscence in *N. pura* found in the present study was also reported by

Bellemère (1977). This eversion is considered to be one of the distinctive characters of Bellemère's *Sclerotinia*-type. In *O. violacea*, not previously studied ultrastructurally, the inner stratum of the inner layer seems to be the most important one in the apical thickening. If it is assumed that this inner stratum corresponds to Bellemère's d layer, the apical apparatus would belong to the *Sclerotinia*-type sensu Bellemère.

Students of the ultrastructure of asci in species of *Sclerotinia* (*S. sclerotiorum*: Codron, 1974; Bellemère, 1977, and *S. tuberosa*: Schoknecht, 1975) did not report on changes related to apex maturation and it is not clear which stages are shown in the micrographs they published in their papers. Therefore it is difficult to make a good comparison.

The outer and inner strata of the outer layer in the ascus wall of *B. inquinans* are likely to correspond to the layers a and b as designated by Bellemère (1977). He clearly showed a "brosse apicale" over the apical apparatus that could not be observed in the specimens used in the present study. Bellemère did not mention the peculiar substructure of the outer layer in *B. inquinans*, although he stated earlier that as a general feature of the b-layer of the ascus wall "ses constituants sont orientés perpendiculairement à la surface de l'asque" (Bellemère, 1975). His interpretation of the layers c and d (inner layer?) cannot be confirmed here. There is a distinct differentiation in the inner layer in the subapical part of the ascus wall. The reactive zone (arrow, Fig. 5A) was called "strate annellogène" by Chadefaud (1973) on the basis of Bellemère's micrographs. It is not clear why Bellemère (1977) excluded this zone from the scheme of his *Bulgaria*-type.

At this stage it can be concluded that *B. inquinans* certainly has a type of apical apparatus of its own. This was also recognized by Bellemère (1977), but additional features especially those concerning apex maturation need to be included in a schematic characterization of the type of apical apparatus in asci of *B. inquinans*.

Dennis (1978) mentioned the strong resemblance between Ombrophila and Neobulgaria, expecting the genera to be reunited in the future. Yet he did treat them separately, emphasizing differences in ascocarp anatomy. The proposal of Baral & Krieglsteiner (1985) to fuse Neobulgaria and Ombrophila is interesting in this respect. The present ultrastructural data confirm a close relation between O. violacea and N. pura, but it is too early to draw final conclusions concerning the taxonomic status of the two genera. The variation in ultrastructure of the apical apparatus and the development of the ascospore wall at the generic and family level within the Leotiales is still unsufficiently known. For this reason a reconsideration on the position of Bulgaria within the Leotiaceae (Helotiaceae sensu auct.) will have to wait as well. Ultrastructural features of the ascus and features pertaining to ascocarp ontogeny and anatomy should be integrated into a modern system for the Ascomycotina. The data on ascus ultrastructure presented here do not confirm the relatedness suggested by the present arrangement of Bulgaria together with Ombrophila and Neobulgaria within the tribe Ombrophiloideae.

#### ACKNOWLEDGEMENTS

The author is greatly indebted to Dr. J. van Brummelen for his support and valuable criticism throughout this study, to Prof. C. Kalkman, Dr. G.M. Lokhorst, and Dr. Nick D. Read for critical reading of the manuscript. He also wishes to thank Mrs. P. van Spronsen at the Botanical Laboratory (Leiden) and Mr. W. Star for their technical advice and help in electron microscopy and Prof. A. Bellemère at St. Cloud, France, for stimulating discussions.

#### REFERENCES

- Baral, H.O. 1987. Der Apikalapparat der Helotiales. Eine Lichtmikroskopische Studie über Arten mit Amyloidring, Z. Mykol. 53: 119-135.
- Baral, H.O. & G.J. Krieglsteiner. 1985. Bausteine zu einer Askomyzeten-Flora der B.R. Deutschland: In Süddeutschland gefundete inoperculate Discomyceten mit taxonomischen, ökologischen und chronologischen Hinweisen. Beih. Z. Mykol. 6: 1–226.
- Barr, M.E. 1976. Perspectives in the Ascomycotina. Mem. N.Y. bot. Gdn. 28: 1-8.
- Beckett, A. 1981a. The ascus with an apical pore: development, composition and function. In: D.R. Reynolds (ed.), Ascomycete Systematics. The Lutrellian Concept. New York.
- Beckett, A. 1981b. Ascospore formation. In: G. Turian & H.R. Hohl (eds.), The Fungal Spore: morphogenetic controls. Proc. third internat. fungal spore symp.: 107–129.
- Beckett, A. & R.M. Crawford. 1973. The development and fine structure of the ascus apex and its role during spore discharge in Xylaria longipes. New Phytol. 72: 357–369.
- Bellemère, A. 1975. Étude ultrastructurale des asques: la paroi, l'appareil apical, la paroi des ascospores chez des Discomycètes inoperculés et des Hystériales. Physiol. végét. 13: 393–406.
- Bellemère, A. 1977. L'appareil apical de l'asque chez quelques discomycètes: Étude ultrastructurale comparative. Rev. Mycol. 41: 233–264.
- Bellemère, A., M.C. Janex-Favre& A. Parguey-Leduc. 1987. Marius Chadefaud et les asques: donées inédites. Études ultrastructurales complémentaires. Bull. Soc. bot. Fr. 134, Lettres bot. 3: 217–246.
- Benny, G.L., D.A. Samuelson & J.W. Kimbrough. 1978. Ultrastructural studies on Orbilia luteorubella (Discomycetes). Can. J. Bot. 56: 2006–2012.
- Brummelen, J. van. 1981. The operculate ascus and allied forms. In: D.R. Reynolds (ed.), Ascomycete Systematics. The Lutrellian Concept. New York.
- Brummelen, J. van. 1986. Ultrastructure of the ascus top and the ascospore wall in Fimaria and Pseudombrophila (Pezizales, Ascomycotina). Persoonia 13: 213–230.
- Brummelen, J. van. 1989. Ultrastructure of the ascus and the ascospore wall in Eleutherascus and Ascodesmis (Ascomycotina). Personnia 14: 1–17.
- Carpenter, S.E. 1988. Leotiales, a name to replace Helotiales (Ascomycotina). Mycologia 80: 127–130.
  Carroll, G.C. 1966. A study of the fine structure of ascosporogenesis in Saccobolus kerveni and Ascodesmis sphaerospora. Ph.D. Thesis. University of Texas, Austin.
- Chadefaud, M. 1964. Sur l'origine et la structure des asques du type annellascé. C. r. hebd. Séanc. Acad. Sci., Paris (Sér., D) 244: 299–301.
- Chadefaud, M. 1973. Les asques et la systématique de Ascomycetes (1). Bull. trimest. Soc. mycol. Fr. 89: 127–170.
- Codron, D. 1974. Étude ultrastructurale de quelques points du développement des asques du Sclerotinia sclerotiorum (Lib.) de Bary. Annls. Sci. nat. Bot., Biol. végét. 15: 255–276.
- Corda, A.K.J. 1842. Icones fungorum 5. Praha.
- Corlett, M. & M.E. Elliott. 1974. The ascus apex of Ciboria acerina. Can. J. Bot. 52: 1459-1463.
- Dennis, R.W.G. 1956. A revision of the British Helotiaceae in the Herbarium of the Royal Botanical Gardens, Kew, with notes on related European species. Mycol. Pap. 62: 1–216.
- Dennis, R.W.G. 1978. British Ascomycetes. Vaduz.
- Eriksson, O. 1983. Outline of the Ascomycetes. Systema Ascomycetum 2: 1-37.
- Gamundi, I.J. & R.W.G. Dennis. 1969. The status of Ascotremella Seaver (Fungi, Helotiales). Darwinia 15: 14-21.
- Griffiths Bronwen, H. 1971. The structure of the Pyrenomycete ascus and its relevance to taxonomy. In: G.C. Ainsworth & J. Webster (eds.), Abstr. first int. mycol. Congr., Exeter: 37–38.
- Hawksworth, D.L., B.C. Sutton & G.C. Ainsworth. 1983. Ainsworths & Bisby's Dictionary of the Fungi (including the Lichens). Kew.
- Korf, R.P. 1973. Discomycetes and Tuberales. In: G.C. Ainsworth, F.K. Sparrow & A.S. Sussman (eds.), The Fungi, An Advanced Treatise 4A: 249-319.
- Merkus, E. 1976. Ultrastructure of the ascospore wall in Pezizales (Ascomycetes). IV. Morchellaceae, Helvellaceae, Rhizinaceae, Thelebolaceae and Sarcoscyphaceae. General discussion. Persoonia 9: 1–38.

- Nannfeldt, J. A. 1932. Studien über die Morphologie und Systematik der nicht-lichenisierten inoperculaten Discomyceten. Nova Acta R. Soc. Sci. upsal. IV, 8 (2): 1–368.
- Schoknecht, J.D. 1975. Structure of the ascus apex and ascospore dispersal mechanisms in Sclerotinia tuberosa. Trans. Br. mycol. Soc. 64: 358-362.
- Schrantz, J.P. 1970. Étude cytologique, en microscopie optique et électronique, de quelques Ascomycètes. II. La paroi. Rev. Cytol. et Biol. végét. 33: 111-168.
- Thiéry, J.P. 1967. Mise en évidence des polysaccharides sur coupes fines en microscopie électronique. J. Microsc. 6: 987–1018.

### PERSOONIA

Published by Rijksherbarium / Hortus Botanicus, Leiden Volume 15, Part 1, pp. 23-31 (1992)

## NEW TAXA OF ENTOLOMA (BASIDIOMYCETES, AGARICALES) FROM ESTONIA AND KARELIA

MACHIEL E. NOORDELOOS1 & VELLO LIIV2

Nine new species of Entoloma are described from the Islands of Saaremaa and Vormsi, Estonia, viz. E. conocybecystis, E. leochromus, E. mutabilipes, E. ochromicaceum, E. politoflavipes, E. rhynchocystidiatum, E. roseotinctum, E. viiduense, and E. violaceozonatum, Entoloma lactarioides is described as new from Karelia.

The present paper is the result of many years of collecting and painting *Entoloma* species by the second author, who lives on the Island of Saaremaa, Estonia in an area extremely rich in *Entoloma* species. Saaremaa has the same geological origin as some other islands in the gulf of Bothnia, viz. Öland and Gotland, and is characterized by highly basic calcareous bedrock. On the island various vegetation types occur, among others also the Alvar type of open grassy vegetation with scattered Juniper trees, and a range of deciduous and coniferous or mixed forests on dry to fairly damp soil. Many taxa of *Entoloma* have been observed in these habitats, and studied and depicted over the years. The present paper gives diagnoses and full descriptions of ten new species of *Entoloma*, collected by Vello Liiv in Estonia and Karelia. Full descriptions and illustrations of all new taxa will be published in the European monograph of the whole genus *Entoloma* (Noordeloos, 1992), that will be accompanied by coloured plates of about 120 species, made by Vello Liiv.

#### 1. Entoloma violaceozonatum Noordel, & Liiv, spec, nov.

Pileus 20-55 mm latus, truncato-conicus vel conicus, umbilicatus, haud hygrophanus, haud translucido-striatus, coeruleo-violaceus vel violaceobrunneus, zonatus, toto fibrilloso-squamulosus, opacus. Lamellae adnatae vel decurrentes, sordide brunneae demum brunneo-roseae acie concolore. Stipes  $40-95\times 4-8$  mm, violaceo-griseus, glaber, politus. Carne violaceo-griseus. Odore saporeque subfarinaceis. Sporae  $10.0-11.5\times(7.0-)7.7-8.5~\mu m$ , Q=1.25-1.4-1.5, heterodiametricae, 5-6-angulatae. Basidia  $35-54\times 8-12(-14)~\mu m$ , 4-sporigera, fibulata. Acies lamellarum sterilis. Cheilocystidia,  $60-125\times 5-14~\mu m$ , clavata. Pileipellis trichodermatis elementis inflatis,  $40-70(-120)\times 8-20~\mu m$  pigmentis intracellulosis. Fibulae abundantae. Habitat in graminosis ad terram calcaream. Holotypus: Vello~Liiv~275, 29-VII-1990, Viidumae, Saaremaa, Estonia (L; isotypus TAM).

Pileus 20-55 mm broad, truncate conical to convex with slightly to distinctly umbilicate centre and involute margin at first soon expanding to applanate or concave with irregularly undulating marginal zone, not hygrophanous, not translucently striate, when young blue-violaceous, then violaceous grey-brown, very strongly and coarsely radially fibrillose, in older specimens fibrillose-squamulose with uplifted squamules, especially in

<sup>1)</sup> Rijksherbarium / Hortus Botanicus, P.O. Box 9514, 2300 RA Leiden, The Netherlands.

Oismae 44-25, Tallinn EE 0035, Estonia.

central part, dull or shining. Lamellae adnate to decurrent, segmentiform to ventricose, sordid beige-brown when young then reddish brown (chocolate) with fimbriate, concolorous edge. Stipe  $40-95 \times 4-8$  mm, usually distinctly broadened towards base, violaceous-grey more or less concolorous with expanded pileus, smooth, finely striate. Context violaceous-grey like surface. Smell en taste not distinctive to subfarinaceous.

Spores  $10.0-11.5 \times (7.0-)7.7-8.5~\mu m$ , average spore  $10.8 \times 7.8~\mu m$ , Q=1.25-1.4-1.5, heterodiametrical, 5-6-angled in side-view with rather pronounced angles. Basidia  $35-54 \times 8-12(-14)~\mu m$ , 4-spored, clamped. Lamella edge entirely sterile with dense clusters of very long, (sub)cylindrical cheilocystidia,  $60-125 \times 5-14~\mu m$ . Hymenophoral trama regular, made up of subcylindrical elements,  $130-300 \times 5-15~\mu m$ . Pileipellis a cutis with transitions to a trichoderm, made up of septate, cylindrical hyphae,  $6-15~\mu m$  wide with cylindrical to narrowly clavate terminal elements,  $40-70(-120) \times 8-20~\mu m$ . Pigment very abundant, brown, diffusely and granulose-intracellular in pileipellis. Pileitrama regular, made up of long, cylindrical to slightly inflated elements, up to  $500 \times 4-30~\mu m$ . Clamps abundant.

Habitat. In grassy places on calcareous soil.

Collection examined. Estonia, Saaremaa, Viidumae, 29 July 1990, Vello Liiv 275 (holotype, L; isotype, TAM).

Entoloma violaceozonatum belongs to section Leptonia on account of the clamped septae and pileipellis that is made up of long, septate hairs. The fibrillose-zonate aspect of the pileus is very distinctive and reminds of the pileal surface of E. undatum. Entoloma dichroum and E. allochroum are very close, but differ among other things by the pale lamellae and a heterogeneous lamella edge with differently shaped cheilocystidia. Entoloma allochroum differs furthermore by having encrusting pigments in the pileipellis.

# Entoloma viiduense Noordel. & Liiv, spec. nov.

Pileus 20–70 mm latus, truncato-conicus vel convexus, margine involutus, centro depressus vel umbilicatus, haud hygrophanus, haud translucido-striatus, coeruleo-griseus vel violaceo-griseus, demum violaceo-brunneus, toto squamulosus. Lamellae adnato-decurrentes, sordide roseae acies concolorae. Stipes 40–130 × 5–12 mm, cylindraceo-flexuosus vel compressus, coeruleus vel griseus, demum violaceo-griseus vel violaceo-brunneus, glaber, politus. Sporae 9.3–12.5(–15) × 7.2–9.3 μm, 5–7(–8) angulatae. Basidia 44–65 × 14–17 μm, 4-sporigera, efibulata. Acies lamellarum fertilis. Pileipellis cutis vel trichoderma hyphis ad 25 μm lata, pigmentis intracellulosis. Fibulae adsunt. Habitat ad terram in silvis. Holotypus: Vello Liiv 178, 263, Viidu, Saaremaa, Estonia (L; isotypus in TAM).

Pileus 20–70 mm broad, truncate conico-convex or convex with involute margin at first then irregularly applanate with undulating marginal zone, with depressed to umbilicate centre, not or only weakly hygrophanous, blue-grey to violaceous grey, fading to violaceous brown with age, not translucently striate, strongly radially fibrillose becoming squamulose with more or less concentric circles of suberect, pointed squamules. Lamellae adnate or with small decurrent tooth, segmentiform to broadly ventricose, up to 12 mm broad, sometimes strongly curled, sordid pink with concolorous, entire edge. Stipe 40–130  $\times$  5–12 mm, flexuose, often compressed with longitudinal groove, often gradually broadened towards base, in upper part initially blue, lower part blue-grey, often turning violaceous-blue or violaceous-brown with age, smooth, glabrous, polished, base white tomentose. Context blue-grey in cortex, inner parts pale. Smell and taste indistinct.

Spores  $9.3-12.5(-15)\times 7.2-9.3~\mu m$ , average spore  $11.5\times 8.7~\mu m$ , Q=1.2-1.4-1.8, rather pronouncedly 5-7(-8)-angled in side-view. Basidia  $44-65\times 14-17~\mu m$ , 4-spored, clampless. Lamella edge fertile, rarely some scattered cylindrical sterile cells found, no real cheilocystidia present. Hymenophoral trama regular, made up of long, inflated elements,  $140-250\times 10-22~\mu m$ . Pileipellis a transition between a cutis and a trichoderm, made up of inflated hyphae with subcylindrical to clavate terminal elements,  $55-140\times 15-25~\mu m$ . Pigment abundant, greyish, intracellular in pileipellis. Brilliant granules abundant. Clamps absent.

Habitat. Terrestrial in large groups in mixed forest on calcareous soil.

Collections examined. Estonia, Saaremaa, Viidu, nature reserve, 14 Aug. 1985, Vello Liiv 178 (L); ditto, 27 July 1990, Vello Liiv 263 (holotype, L; isotype, TAM).

Entoloma viiduense is a fairly robust species of Leptonia, sect. Cyanula. A very distinctive feature of this taxon is the colour that changes from blue or blue grey in young and fresh specimens to violaceous-brown in old and weathered specimens. Old stages may be confused with E. nigroviolaceum, but that species is violaceous-black or violaceous brown already from the beginning, and its stipe is fibrillose-striate, often also apically flocculose. Also the ecological range of E. nigroviolaceum is different, as it grows under fairly acid conditions in meadows and Sphagnum-bogs. Entoloma scabrosum, that also has been found in the forests of Viidu, has a grey-brown, translucently striate pileus, and a innately fibrillose stipe.

## 3. Entoloma rhynchocystidiatum Noordel. & Liiv, spec. nov.

Pileus 30–60 mm latus, conicus demum applanatus, subumbilicatus, margine involutus demum deflexus, haud hygrophanus, haud translucido striatus, griseo-brunneus tinctu ochraceus, centro obscurioir, margine tincto violaceo, toto fibrilloso squamulosus. Lamellae confertae, emarginatae, ventricosae, sordide roseae acie concolores. Stipes 30–75 × 3–6 mm, cylindraceus, griseo-violaceus, politus. Odore saporeque inconspicuis. Sporae (8.0–)8.5–11.0(–11.5) × 7.0–8.5 μm, 5–7-angulatae. Basidia 24–40 × 8–14 μm, 2- vel 4-sporigera, efibulata. Acies lamellarum sterilis. Cheilocystidia, 25–45(–70) × 7–15(–20) μm, clavata apice mucronata vel rostrata vel lageniformibus. Pileipellis trichoderma elementis inflatis, 30–80 (–110) × 10–25 μm pigmentis intracellulosis. Fibulae nullae. Habitat in vegetatione nominatur 'Alvar'. Holotypus: Vello Liiv 220, 9-IX-1986, Svybi, Saaremaa, Estonia (L; isotype TAM).

Pileus 30–60 mm broad, convex to applanate, slightly depressed at centre, with involute then deflexed margin, with undulating marginal zone with age, not hygrophanous, grey-brown with ochraceous tinge, darker at centre and margin with distinct violaceous tinge, entirely radially fibrillose-squamulose with fine, pointed squamules. Lamellae moderately crowded, adnate-emarginate, ventricose, sordid pink with concolorous edge. Stipe  $30-75\times3-6$  mm, cylindrical, flexuose, slightly broadened at base, grey with distinct violaceous tinge, smooth, polished, base white tomentose. Context grey. Smell and taste indistinct.

Spores  $(8.0-)8.5-11.0(-11.5) \times 7.0-8.5 \,\mu\text{m}$ , average spores  $9.4 \times 8.2 \,\mu\text{m}$ , Q = 1.1-1.2-1.3(-1.5), 5–7-angled in side-view with rather weak angles, thin-walled. Basidia  $24-40 \times 8-14 \,\mu\text{m}$ , 2- and 4-spored, clampless. Lamella edge entirely sterile with masses of densely clustered cheilocystidia,  $25-45(-70) \times 7-15(-20) \,\mu\text{m}$ , clavate with mucronate to rostrate apex or lageniform. Hymenophoral trama regular, made up of medium-sized, subcylindrical to inflated elements,  $150-220 \times 6-25 \,\mu\text{m}$ . Pileipellis a

trichoderm of inflated terminal elements,  $30-80(-110)\times 10-25~\mu m$ . Pigment brownish, intracellular in pileipellis. Pileitrama regular, made up of inflated elements,  $100-230\times 4-30~\mu m$ . Clamps absent.

Habitat. In Juniperus-heath on calcareous soil.

Collections examined. Estonia, Vormsi, near the village Svybi, 9 Sept. 1986, Vello Liiv 220 (holotype, L; isotype, TAM).

Entoloma rhynchocystidiatum belongs to subgenus Leptonia, sect. Rhamphocystotae (Largent) Noordel. on account of its large, rostrate cheilocystidia. No similar species is known from Europe. Entoloma subcorvinum from North America differs by having deep blue colours in pileus and stipe, and larger cheilocystidia.

### 4. Entoloma leochromus Noordel. & Liiv, spec. nov.

Pileus 10-40 mm latus, truncate-conicus vel convexus, margine inflexus, centro depressus vel umbilicatus, haud hygrophanus, haud translucido striatus, moderate flavo-griseus vel griseo brunneus, toto velutinus vel squamulosus. Lamellae adnatae vel decurrentes, albidae demum roseae vel roseo-brunneae acie concolore. Stipes  $50-90\times2-4$  mm, cylindraceus, pileo concolor, politus. Sporae  $9.0-13.0\times7.0-8.0(-9.5)$  µm, 5-9 angulatae. Basidia 4-spored, efibulata. Acies lamellarum fertilis. Pileipellis trichoderma vel hymeniderma elementis late clavatis vel globosis; 20-30 µm latis pigmentis intracellulosis. Fibulae nullae. Habitat in silvis frondosis (*Fraxinus, Alnus*). Holotypus: *Vello Liiv 269*, Viidu, Saaremaa, Estonia (L; isotype TAM).

Pileus 10–40 mm broad, truncately conical to convex with slightly depressed to umbilicate centre, with involute then deflexed margin, not hygrophanous, not translucently striate, pale to moderately dark yellowish grey to greyish brown, entirely granulose-velutinous to distinctly squamulose with small, semi-erect squamules, then usually also with distinct calotte, dull or somewhat shining. Lamellae adnate to decurrent, arcuate to segmentiform or subventricose, white then pink finally reddish-brown, with entire, concolorous edge. Stipe  $50-90\times 2-4$  mm, cylindrical, usually slightly curved and broadened towards base, yellowish grey, paler or almost concolorous with pileus, smooth, glabrous, polished, base white tomentose, but slowly staining pinkish red when picked. Context pale yellowish-grey in cortex, pallid in inner parts. Smell none. Taste bitter.

Spores  $9.0-13.0\times7.0-8.0(-9.5)~\mu m$ , average spore  $11.1\times7.5~\mu m$ , Q=1.3-1.5-1.7, irregularly heterodiametrical, 5-9-angled in side-view with rather pronounced angles. Basidia 4-spored, clampless. Lamella edge fertile. Cystidia absent. Hymenophoral trama regular, made up of cylindrical to slightly inflated elements,  $100-300\times4-25~\mu m$ . Pileipellis a trichoderm at margin, a hymeniderm at centre, made up of broadly clavate to subglobose elements,  $35-80\times20-30~\mu m$ . Pigment abundant, brown, intracellular in pileipellis. Brilliant granules very abundant in trama of pileus. Clamps absent.

Habitat. In deciduous forest (Fraxinus, Alnus) with dense undergrowth of Rubus.

Collections examined. Estonia, Saaremaa, Viidu, nature reserve, 27 July 1990, Vello Liiv 265 and 28 July 1990, Vello Liiv 269 (holotype, L; isotype, TAM).

The most distinctive features of *Entoloma leochromus* are the yellowish grey fruitbodies, opaque, granulose to squamulose, not translucent pileus, polished stipe and fertile lamella edge. *Entoloma turci* is similar, but has much darker colours and a sterile, often brown-coloured lamella edge. *Entoloma longistriatum* differs in colour, translucently striate pileus, and sterile lamella edge.

### 5. Entoloma ochromicaceum Noordel. & Liiv, spec. nov.

Pileus 12–35 mm latus, truncato-conicus demum convexus margine involutus, subhygrophanus, ad marginem subtiliter translucido striatus, pallide ochraceo-griseus, toto granuloso-micaceus. Lamellae adnatae, albidae demum roseae acie concolore vel in parte brunneolus. Stipes  $35-65\times1-3$  mm, cylindraceus, pileo concolorus vel pallidiorus, glaber, politus. Contextus albidus. Odore saporeque nullis. Sporae  $9.0-11.5\times7.0-8.5$  µm, 5-6-angulatae. Basidia 4-sporigera, efibulata. Acies lamellarum sterilis. Cheilocystidia  $20-50\times7-20$  µm, clavata vel globosa, utriformia vel lageniformia. Pileipellis trichoderma elementis 8-22 µm lata pigmentis intracellulosis. Fibulae nullae. Habitat in locis graminosis in silvis. Holotypus:  $Vello\ Liiv\ 295,\ 22-VIII-1991,\ Viidu,\ Piskuna-house,\ Saaremaa,\ Estonia\ (L;\ isotypus\ TAM).$ 

Pileus 12-35 mm broad, truncate conical at first then convex finally irregularly planoconvex with involute then deflexed margin, with undulating marginal zone when old, slightly hygrophanous, when moist tender beige-ochraceous, only very slightly translucently striate at margin only, granulose on limb, minutely squamulose in central part, shining, micaceous. Lamellae narrowly adnate, narrowly ventricose, white then purely pink with in part brownish edge. Stipe  $35-65 \times 1-3$  mm, cylindrical, slightly curved and bent towards base, concolorous with pileus or paler, smooth, glabrous. Context purely white. Smell and taste not distinctive.

Spores  $9.0-11.5 \times 7.0-8.5~\mu m$ , average spore  $10.1 \times 7.5~\mu m$ , Q=1.2-1.35-1.5, rather pronouncedly 5-6-angled in side-view. Basidia  $26-40 \times 9-13~\mu m$ , 4-spored, clampless. Lamella edge entirely sterile of serrulatum-type; cheilocystidia  $20-50 \times 7-20$ , clavate to globose, utriform to lageniform, thin-walled, sometimes with brown, intracellular pigment, especially in lower strand of hyphae on which the cheilocystidia are born. Pileipellis a cutis with transitions to a trichoderm, made up of inflated hyphae,  $8-15~\mu m$  wide with clavate terminal elements,  $45-120 \times 8-22~\mu m$ . Pigment abundant, brown, intracellular in pileipellis. Pileitrama regular, made up of more or less cylindrical hyphae,  $9-22~\mu m$  wide. Brilliant granules very abundant in pileitrama. Clamps absent.

Habitat. In groups on grassy spot in deciduous forest.

Collection examined. Estonia, Saaremaa, Viidu, near the Piskuna house, 22 Aug. 1990, Vello Liiv 295 (holotype, L; isotype, TAM).

Entoloma ochromicaceum clearly belongs in the group of E. longistriatum, from which it differs by the pale coloured, hardly striate, entirely granulose-micaceous pileus, and structure of the lamella edge. The poorly known species Rhodophyllus caliginosus Romagn. & Favre is also close, but has a darker brown, striate, smooth or slightly granulose pileus, brown lamellae and a grey stipe.

# Entoloma lactarioides Noordel. & Liiv, spec. nov.

Habitus Lactarii species revocat. Pileus 15–35 mm latus, convexus demum concavus, umbilicatus, paulisper hygrophanus, haud translucido striatus, modice griseo-brunneus roseo-tinctus, fibrillosus. Lamellae moderate distantes, adnato-decurrentes, pallide griseae demum griseo-roseae. Stipes 20–40 × 2–5 mm, cylindraceus, roseus griseo-tinctae, politus. Odore saporeque nullae. Sporae 7.5–9.2 × 7.5–8.5 μm, 5–6- angulatae. Basidia 30–46 × 8–15 μm, 4-sporigera, fibulata. Acies lamellarum fertilis. Cystidia nulla. Pileipellis cutis hyphis cylindraceis, 2–7 μm latis interdum cum elementis terminalis clavatis, 7–21 μm latis, pigmentis intracellulosis interdum in additione incrustatis. Fibulae abundantes. Habitat in pratis. Holotypus: *Vello Liiv 217*, 16 IX-1991, Otradnoje, Karelia (L; isotypus in TAM).

Pileus 15–35 mm broad, convex to concave, umbilicate to infundibuliform, with involute then deflexed margin, slightly hygrophanous, when moist moderately dark greybrown with pinkish tinge, not translucently striate, entirely covered with fine fibrillose patches, slightly pallescent on drying. Lamellae, L = 25–35, 1 = 3–7, moderately distant, adnate-decurrent, segmentiform to ventricose, greyish white then sordid pink with concolorous, entire edge. Stipe  $20-40\times2-5$  mm, cylindrical, sometimes flexuose or curved at base, pinkish with grey or brown tinge, smooth, glabrous. Context pallid. Smell and taste none.

Spores  $7.5-9.2\times7.5-8.5~\mu m$ , average  $8.2\times7.7~\mu m$ , Q=1.0-1.15-1.25, 5-6-angled in side-view with rather pronounced angles. Basidia  $30-46\times8-15~\mu m$ , 4-spored, clamped. Lamella edge fertile. Cystidia absent. Hymenophoral trama regular, made up of short, inflated elements,  $30-170\times7-21~\mu m$ . Pileipellis a cutis of narrow, cylindrical hyphae,  $2-7~\mu m$  wide, with scattered tufts of inflated, repent or ascending terminal elements,  $30-90\times5-15(-20)~\mu m$ . Pileitrama regular, made up of short elements,  $60-150(-200)\times10-25~\mu m$ . Pigment pale brown, intracellular, in addition finely encrusting on a few hyphae in pileipellis and upper pileitrama. Clamps abundant.

Habitat. In meadow.

Collection examined. Russian Federation, Karelia, Otradnoje, 16 Sept. 1986, Vello Liiv 217 (holotype, L; isotype, TAM).

The distinctive characters of *Entoloma lactarioides* are the omphalioid fruit-bodies with distinct pinkish tinges, the not striate, fibrillose pileus, and the smooth and glabrous stipe. The fibrillose patches on the pileal surface consist of tufts of inflated elements, arising from a rather simple cutis-like structure which is normally in subgenus *Entoloma*. Also the structure of the trama, with short elements, place this species in subgenus *Entoloma*. Therefore *Entoloma lactarioides* is accommodated in this subgenus. The habit and smooth stipe are distinctive for section *Polita*, in which it is provisionally accommodated, since the aspect and pigmentation of the pileal surface is rather aberrant for section *Polita*.

# 7. Entoloma conocybecystis Noordel. & Liiv, spec. nov.

Pileus 45 mm latus, plano-convexus, umbonatus, margine deflexus, haud hygrophanus, haud translucido-striatus, violaceo-ochraceus, radialiter fibrillosus. Lamellae moderate distantes, liberae, ventricosae, roseo-brunneae. Stipe 95 × 5 mm, cylindraceus, ochraceus, argenteo-striatus. Odore saporeque nullae. Sporae 8.2–10.5 × 6.2–7.7(–8.5) μm, 6–7 angulatae. Basidia tetrasporigera, fibulata. Acies lamellarum heterogeneis. Cheilocystidia 35–55 × 8–12 × 1.5–5 × 4–8 μm, lecithiformia, abundantia. Pileipellis trichoderma elementis inflatis 10–25 μm latis pigmentis intracellulosis. Fibulae in hymenio presentes. Habitat in graminosis in associacione Juniperi. Holotypus: *Vello Liiv 124*, 14-VIII-1984, Kipi, Saaremaa, Estonia (L; isotypus in TAM).

Pileus 45 mm broad, plano-convex with low umbo, with deflexed margin, not hygrophanous, not translucently striate, ochraceous with slight violaceous tinge, radially fibrillose. Lamellae, L= about 40, l=3-5, moderately distant, free, ventricose, brown with pink tinge. Stipe  $95 \times 5$  mm, cylindrical with slightly broadened base, ochraceous, paler than pileus, densely silvery fibrillose all over. Context white, in base of stipe yellowish. Smell and taste not distinctive.

Spores  $8.2-10.5 \times 6.2-7.7(-8.5)$  µm, average spore  $9.2 \times 6.7$  µm, Q = 1.3-1.4-1.7, heterodiametrical, 6-7-angled in side-view. Basidia  $30-36 \times 10-16$  µm, 4-spored,

clamped. Lamella edge (almost) sterile. Cheilocystidia  $35-55\times8-12$  (base)  $\times$  1.5-5 (neck)  $\times$  4-8 (capitulum), lecythiform, often with slimy cap round capitulum; numerous. Hymenophoral trama regular, made up of very long, fusoid elements, up to  $600\times18-35$   $\mu m$ . Pileipellis a cutis with transitions to a trichoderm, made up of long, inflated elements,  $60-260\times10-25$   $\mu m$ . Pigment brown, intracellular in pileipellis. Pileitrama regular, made up of long, inflated elements,  $350-550\times25-45$   $\mu m$ . Clamps present in hymenium, not seen in other tissues.

Habitat. In dry Juniperus-heath on calcareous soil.

Collection studied. Estonia, Saaremaa, Kipi, 14 Aug. 1984, Vello Liiv 124 (holotype, L; isotype, TAM).

The distinctive characters of *Entoloma conocybecystis* are the relatively pale ochraceous-violaceous colour of the pileus, pale lamellae, pallid, ochraceous stipe, and perfectly tibii-form cheilocystidia. It belongs to subgenus *Trichopilus*, and is close to *E. elodes*, that has darker fruit-bodies, larger spores, and a completely different habitat in peatbogs among *Sphagnum*. *Entoloma jubatum* is also close, but has much darker fruit-bodies, and less distinctly lecythiform cheilocystidia. *Entoloma porphyrophaeum* has a very different porphyraceous-brown colour and larger spores. *Entoloma fuscotomentosum* has a squamulose, grey-brown pileus, pale greyish-pink lamellae, and less distinctly lecythiform cheilocystidia.

Entoloma conocybecystis is only known with certainty from Saaremaa but has also been observed in the Netherlands.

## 8. Entoloma politoflavipes Noordel. & Liiv, spec. nov.

Pileus 10–35 mm latus, semiglobosus vel convexus margine deflexus vel rectus, umbilicatus, hygrophanus, translucido striatus, flavo-brunneus vel brunneus centro obscurioir, toto fibrillosus vel velutinus, centro subtiliter squamulosus. Lamellae adnato-decurrentes, segmentiformia vel ventricosae, albidae demum roseae. Stipes 30–50 × 1–2 mm, cylindraceus, flavidus, politus. Odore saporeque nullae. Sporae 7.0–9.5 × 6.0–8.5 μm, Q = 1.1–1.3–1.5, heterodiametricae, 5–7-angulatae. Basidia 4-sporigera, fibulata. Acies lamellarum fertilis. Pileipellis cutis vel trichoderma elementis cylindraceus vel inflatis, ad 20 μm latis pigmentis intracellulosis. Fibulae presentes. Habitat in pratis. Holotypus: Vello Litv 171, 11-VIII-1985, Viidu, Saaremaa, Estonia (L; isotype in TAM).

Pileus 10-35 mm broad, semiglobose to convex with deflexed or straight margin, umbilicate, hygrophanous, when moist translucently striate, yellow-brown to brown with dark brown centre, slightly pallescent on drying, entirely finely fibrillose-felted, centre minutely squamulose. Lamellae broadly adnate-decurrent, broadly segmentiform to subventricose, not extending under pileus, white then pink finally brownish pink, with concolorous, entire edge. Stipe  $30-50\times 1-2$  mm, cylindrical, straight or flexuose, often slightly curved at base, yellow, much paler than pileus, smooth, glabrous, polished, base white tomentose. Context pallid. Smell and taste not distinctive.

Spores  $7.0-9.5\times6.0-8.5~\mu m$ , average spore  $8.1-8.5\times6.3-6.9~\mu m$ , Q=1.1-1.3-1.5, heterodiametrical, 5-7-angled in side-view. Basidia  $20-35\times6-11~\mu m$ , 4-spored, clamped. Lamella edge fertile. Cystidia absent. Pileipellis a cutis with transitions to a trichoderm, made up of cylindrical to inflated elements, up to  $20~\mu m$  wide. Pigment abundant, brown, intracellular in pileipellis. Clamps abundant.

Habitat. In meadow on calcareous soil.

Collections examined. Estonia, Saaremaa, Viidu, 11 Aug. 1985, Vello Liiv 171 (holotype, L; isotype, TAM); ditto, 21 Aug. 1984, Vello Liiv 135; ditto, 26 July 1990, Vello Liiv 259 (all in L).

Entoloma politoflavipes belongs to section Griseorubida, subsect. Parvisporae on account of it Leptonioid habit, clamped basidia, trichodermal pileipellis and small spores. It comes close to E. farinasprellum, which has dark grey-brown fruit-bodies and a farinaceous smell. Entoloma olivaceotinctum and E. weholtii differ in colour and by having a sterile lamella edge with well-differentiated cheilocystidia.

### 9. Entoloma mutabilipes Noordel. & Liiv, spec. nov.

Pileus 10-20 mm latus, convexus demum plano-convexus, umbilicatus, hygrophanus, translucido-striatus, pallide roseo-alutaceus, centro obscuriore, expallens, glabrus centro squamulosus; lamellae quasi liberae, ventricosae, albidae demum roseae; stipes  $30-45\times1-2$  mm, cylindraceus, azureus demum pallide griseus, glabrus, politus. Odore saporeque nullis. Sporae  $9.0-12.0\times7.5-9.5$  µm, Q=1.1-1.3-1.5, 5-8 angulatae; basidia  $23-40\times7.5-12$  µm, 4-sporigera, efibulata; acies lamellarum sterilis vel heterogeneis; cheilocystidia  $24-70\times6-15$  µm, cylindracea vel clavata. Pileipellis cutis vel trichoderma elementis cylindraceis vel clavata, ad 20 µm lata pigmentis intracellulosis; fibulae desunt. Holotypus:  $Vello\ Liiv\ 300.\ 26-1X-1990$ , Viidu, Saaremaa, Estonia (L; isotypus in TAM).

Pileus 10-20 mm broad, convex to plano-convex, umbilicate, with deflexed or straight, crenate margin, hygrophanous, when moist translucently striate up to three-quart of the radius, pale beige with slight pink tinge with darker, grey-brown centre (calotte), pallescent to pale grey with darker centre, almost glabrous at margin, at centre minutely squamulose. Lamellae almost free, ventricose, white then almost purely pink with concolorous, entire edge. Stipe  $30-45\times 1-2$  mm, cylindrical, sky-blue when young, soon becoming pale grey, smooth, polished, base white tomentose. Context pallid. Smell and taste indistinct.

Spores  $9.0-12.0\times7.5-9.5~\mu m$ , average  $10.6\times7.9~\mu m$ , Q=1.1-1.3-1.5, heterodiametrical, 5-8-angled in side-view. Basidia  $23-40\times7.5-12~\mu m$ , 4-spored, clampless. Lamella edge usually entirely sterile, sometimes heterogeneous, with dense clusters of cystidia mixed among fertile basidia. Cheilocystidia  $24-70\times6-15~\mu m$ , cylindrical to clavate. Hymenophoral trama regular, made up of cylindrical to inflated hyphae, elements  $110-300\times4-20~\mu m$ . Pileipellis a cutis of cylindrical,  $4-15~\mu m$  wide hyphae with scattered trichodermal tufts of ascending, clavate terminal elements,  $20-70\times12-22~\mu m$ . Pigment brownish, intracellular in pileipellis. Brilliant granules and oily guttules very abundant in trama. Clamps absent.

Habitat. In large group in grassy spot in deciduous forest.

Collection examined. Estonia, Saaremaa, Viidu, 26 Sept. 1990, Vello Liiv 300 (holotype, L; isotype, TAM).

Entoloma mutabilipes belongs to stirps Asprellum on account of the strongly striate subglabrous pileus and blue-grey, polished stipe. It differs from all known taxa in this group in the tender pinkish yellow-grey colour of the pileus, in combination with a sterile lamella edge.

### 10. Entoloma roseotinctum Noordel. & Liiv, spec. nov.

Pileus 15–35 mm latus, convexus, umbilicatus, haud hygrophanus, haud translucido-striatus, griseoroseus, radialiter fibrillosus vel squamulosus; lamellae liberae, ventricosae, albae demum roseae; stipes  $25-50\times2-3$  mm, cylindraceus, griseus, glaber, politus. Odore saporeque nullis; sporae  $9-11\times6-7.7$  µm, 5–7-angulatae; basidia 4-sporigera, efibulata; acies lamellarum sterilis; cheilocystidia  $25-60\times8-13$  µm, clavata vel lageniformia; pileipellis trichoderma vel hymeniderma elementis clavatis ad 30 µm latis pigment intracellulosis; fibulae desunt. Holotypus: *Vello Liiv 181*, 15-VIII-1985, Viidu, Saaremaa, Estonia (L; isotypus in TAM).

Pileus 15-35 mm broad, convex, umbilicate, with deflexed margin, not hygrophanous, not translucently striate, tender greyish pink, radially fibrillose, finely grooved in marginal zone. Lamellae free, ventricose, white then purely pink with concolorous, entire edge. Stipe  $25-50\times2-3$  mm, cylindrical with distinctly broadened, almost bulbous base, grey, glabrous, polished, base white tomentose. Context pallid. Smell and taste indistinct.

Spores  $9.0-11.0\times6.0-7.7~\mu m$ , average spore  $9.9\times7.1~\mu m$ , Q=1.2-1.4-1.6, heterodiametrical, 5-7-angled in side-view. Basidia 4-spored, clampless. Cheilocystidia  $25-60\times8-13~\mu m$ , clavate to lageniform, in dense clusters. Pileipellis a trichoderm at margin, a hymeniderm at centre, made up of (broadly) clavate terminal elements,  $30-70\times12-20~\mu m$ . Pigment intracellular in pileipellis. Clamps absent.

Habitat. In groups in forest.

Collections examined. Estonia, Saaremaa, Viidu, nature reserve, 15 Aug. 1985, Vello Liiv 181 (holotype, L; isotype, TAM).

Entoloma roseotinctum is distinctive on account of its greyish-pinkish pileus, grey stipe, and sterile lamella edge.

#### REFERENCE

Noordeloos, M.E. 1992. Entoloma. In: Candusso (ed.), Fungi Europei, vol. 5. Saronno (in print).

#### PERSOONIA

Published by Rijksherbarium / Hortus Botanicus, Leiden Volume 15, Part 1, pp. 33-53 (1992)

### SOME MYCENEAE OF THE HIMALAYAN FOOTHILLS

#### R.A. MAAS GEESTERANUS

Rijksherbarium / Hortus Botanicus, P.O. Box 9514, 2300 RA Leiden, The Netherlands

Among the species of Mycena and related genera of the Himalayan foothills described in this paper, the following are proposed as new: Mycena abietina, M. bathyrrhiza, M. cinnabarina, M. coalita, M. gentilis, Hydropus eburneus, and Xeromphalina aspera.

The Himalayan chain stretches across the northern part of the Indian subcontinent from the Hindu Kush in Afghanistan to the west to the Khasya Hills in Assam to the east. The central and more readily accessible parts of this chain – in Kashmir, India, Nepal, Sikkim – have frequently been visited by botanical expeditions, resulting, among other things, in a gradual increase in the number of mycological publications. It is perhaps not without reason, however, that most papers are silent about the Mycenas. The very size of most of these fungi may have convinced collectors that such denizens (if noticed at all) were not worth the trouble.

Several of the species described in the present paper were collected in 1964 by my colleague Dr. C. Bas in India in the states of the Punjab, Uttar Pradesh, and Himachal Pradesh. It may be noted that, surprisingly, none of the species collected by Hooker and described by Berkeley from Sikkim and the Khasya Hills (1850: 78–81 and 1852: 101–103) and recognizable as belonging to the genus *Mycena* have been found in the region searched by Dr. Bas. One possible explanation is that part of Hooker's collections came from much farther to the east and from higher altitudes, and another is that a number were collected much earlier in the season–May and June. The collections of Dr. Bas were made in August and September and, in view of the general experience in Europe, these months may not have been particularly favourable for Mycenas. Given the enormous expanse of the Himalayas, however, and the great range of altitudinal and vegetational differences, it does not seem unreasonable to expect further discoveries. The present paper is hopefully meant to stir the curiosity.

Grateful thanks are due to the authorities of the herbaria at Edinburgh (E) and Kew (K) for the loan of material.

#### KEY TO THE SPECIES TREATED

- 1. Spores amyloid.
  - 2. Hyphae of the pileipellis diverticulate, excrescences tending to form dense masses.
    - 3. Lamellae tender, c. 20 reaching the stipe.
      - 4. Lamellae broadly adnate, decurrent with a short tooth. Pleurocystidia present:
        - 3. Mycena abietina (sect. Fragilipedes)
      - 4. Lamellae narrowly adnate. Pleurocystidia absent:
        - 4. Mycena bathyrrhiza (sect. Fragilipedes)

- 3. Lamellae tough, 24-35 reaching the stipe. Pleurocystidia absent:
  - 1. Mycena galericulata (sect. Mycena)
- 2. Hyphae of the pileipellis smooth or only sparsely covered with excrescences.
  - 5. Cheilocystidia smooth.
    - Spores up to 5 μm broad.
      - 7. Lamellar edge concolorous with the sides or paler.
        - 8. Lamellar edge concave. Spores 2.2-2.7 μm broad:

11. Xeromphalina aspera

- 8. Lamellar edge convex. Spores more than 3.5 μm broad:
  - 7. Mycena pura (sect. Calodontes, subsect. Purae)
- 7. Lamellar edge darker than the sides, dark red:
  - 6. Mycena cinnabarina (sect. Calodontes, subsect. Marginatae)
- Cheilocystidia apically covered with very coarse excrescences.
  - 9. Lamellae toughish. Hyphae of the pileipellis embedded in gelatinous matter:
    - 2. Mycena tintinnabulum (sect. Mycena)
  - 9. Lamellae tender. Hyphae of the pileipellis not gelatinized:
    - 5. Mycena coalita (sect. Fragilipedes)

- Spores inamyloid.
  - Cheilocystidia apically and ventrally almost equally broad. Pleurocystidia present:
     Mycena olida (sect. Hiemales, subsect. Hiemales)
  - 10. Cheilocystidia apically much narrower than ventrally. Pleurocystidia absent:
    - 8. Mycena gentilis (sect. Calodontes, subsect. Violacellae)

### MYCENA (Pers.) Roussel

Agaricus [sect.] Mycena Pers., Tent. Fung. Suppl. (1797) 69. — Agaricus sect. Mycena Pers., Syn. meth. Fung. (1801) XVI, 375. — Mycena (Pers.) Roussel, Flore Calvados, 2nd ed. (1806) 64. — Type species: Agaricus galericulatus Scop.

# Mycena sect. Mycena

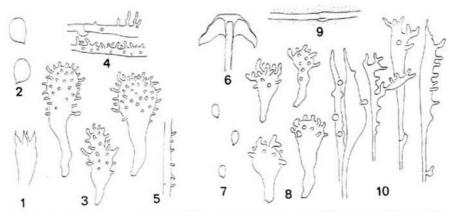
Agaricus trib. Propriae Fr., Obs. mycol. 2 (1818) 155. — Lectotype: Agaricus galericulatus Scop. For further synonymy, see Maas Geesteranus (1985: 339).

# 1. Mycena galericulata (Scop.: Fr.) S.F. Gray — Figs. 1-5

Agaricus galericulatus Scop., Flora carniol., ed. 2, 2 (1772) 455; Fr., Syst. mycol. 1 (1821) 143. — Mycena galericulata (Scop.: Fr.) S.F. Gray, Nat. Arrang. Br. Pl. 1 (1821) 619. — Neotype: 'Fungi of Yugoslavia / Mycena galericulata' (No. 982.217-639; L).

For further synonymy, see Maas Geesteranus (1985: 346).

Basidiomata subfasciculate. Pileus 12-35 mm across, conical to campanulate, with little pronounced, rounded umbo, smooth to shallowly sulcate, appearing glabrous, pale brownish beige, darker at the centre, becoming darker flesh-coloured brown with age, and the surface becoming slightly fissured. Flesh thin except at the centre of the pileus,



Figs. 1–5. Mycena galericulata (Bas 4142; L). 1. Basidium; 2. spores; 3. cheilocystidia; 4. hyphae of the pilcipellis; 5. hypha of the cortical layer of the stipe. — Figs. 6–10. Mycena tintinnabulum (Bas 4257; L). 6. Section of pileus (dried); 7. spores; 8. cheilocystidia; 9. hypha of the pilcipellis; 10. terminal cells of hyphae of the cortical layer of the stipe. (Fig. 6, × 2; all others, × 700.)

pale beige, in age flushed with pink. Odour raphanoid when crushed. Taste raphanoid. Lamellae 24-35 reaching the stipe, elastic-tough, ascending, up to 4.5 mm broad, ventricose, adnate, decurrent with a short tooth, smooth to veined, very pale beige, becoming flushed with a reddish tint, edge convex, concolorous. Stipe  $35-90 \times 1-3$  mm, hollow, tough, equal, terete to somewhat compressed, straight or slightly flexuous, smooth, finely pruinose at the apex, glabrous farther below, shiny, beige to more brownish or with a pinkish shade, the base somewhat rooting, covered with whitish fibrils.

Basidia (none seen fully mature)  $24-27 \times 10-11~\mu m$ , clavate, 4-spored, clamped, with plump sterigmata  $7-8~\mu m$  long. Spores (not quite mature)  $7.2-9.0 \times 5.8-6.3~\mu m$ , broadly pip-shaped, smooth, amyloid. Cheilocystidia  $40-45 \times 9-14.5~\mu m$ , forming a sterile band, clavate, clamped, covered with not very numerous, simple, mostly curved excrescences  $2-3.5 \times 1-2~\mu m$ . Pleurocystidia absent. Lamellar trama weakly brownish vinescent in Melzer's reagent. Hyphae of the pileipellis  $2-4.5~\mu m$  wide, clamped, covered with simple to furcate, cylindrical excrescences  $2-7 \times 1-2~\mu m$ . Hyphae of the cortical layer of the stipe  $1.5-2.5~\mu m$ , wide, clamped, sparsely covered with simple, cylindrical excrescences  $1.8 \times 1~\mu m$ .

Material examined (all collections 4-spored). PAKISTAN: West Pakistan, Patriata, Murree Hills, 20 Aug. 1953, Sultan Ahmad 12069, on rotten stump of Pinus excelsa (K); Murree Hills, Khanspur, 20 Sept. 1975, Shi Mycol. Herb. 2142, on logs (K); same locality, same date, Shi Mycol. Herb. 2148, on the ground (K).

INDIA: Himachal Pradesh, Narkanda, 11 Aug. 1964, C. Bas 4142, on decaying log in forest of Abies pindrow and Picea smithiana, c. 2750 m alt. (No. 964.289-260; L).

The macroscopic description of the species is adapted from the notes accompanying collection *Bas 4142*, complemented by my own observations on the dried material. The microscopic details are based on reexamination of this collection.

Berkeley (1852: 101) recorded Agaricus galericulatus from Sikkim collected by [J.D.] Hooker in 1849 (Ser. 2, No. 6). I have studied this material (preserved at K) which consists of four separate basidiomes and a fasciculate group of six specimens. This fasciculate habit is unusual in Mycena galericulata. The hyphae of both the pileipellis and the cortical layer of the stipe appear to be smooth. These two last named characters combined with the tender consistency of the lamellar trama show that Hooker's No. 6 is not Mycena galericulata and very probably not even a member of section Mycena. The poor quality of the material, of which neither basidia nor cystidia could be made out, defies identification.

### 2. Mycena tintinnabulum (Fr.) Quél. - Figs. 6-10

Agaricus tintinnabulum Fr., Epicr. Syst. mycol. (1838) 107. — Mycena tintinnabulum (Fr.) Quél., Mém. Soc. Emul. Montbél. II 5 (1872) 105. — Type locality: Sweden.

For further synonymy, see Maas Geesteranus (1985: 364).

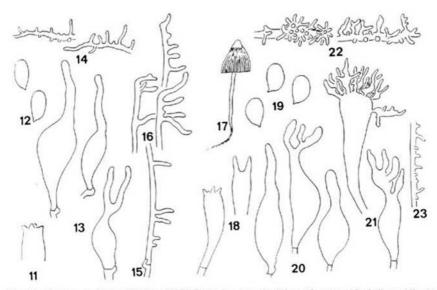
Basidiomata fasciculate. Pileus 6-12 mm across, parabolical, conical or convex, somewhat umbonate, centrally flattened or slightly depressed, smooth, translucent-striate, glabrous, fairly dark grey-brown to pale beige. Flesh thin, except at the centre of the pileus, more or less concolorous with the pileus. Odour and taste indistinctive. Lamellae 21 reaching the stipe, fairly tough, ascending, up to 2.5 mm broad, broadly adnate, decurrent with a short tooth, smooth to ribbed, dorsally intervenose, pale grey-brown at the base, whitish towards the edge, becoming flushed with pink with age, the edge shallowly convex, whitish. Stipe  $15-25\times0.6-1.8$  mm, hollow, equal or somewhat narrowed below, curved, terete, smooth, pruinose above, glabrous farther down, whitish at the apex, pale yellowish sepia brown below, the base densely covered with long, rather coarse, whitish fibrils.

Basidia (none seen fully mature) c.  $17 \times 4.5 \,\mu\text{m}$ , clavate, clamped, 4-spored, with sterigmata c.  $2.7 \,\mu\text{m}$  long. Spores  $4.7-5.5 \times 2.8-3.1 \,\mu\text{m}$ , pip-shaped, smooth, amyloid. Cheilocystidia  $21.5-28 \times 7-11 \,\mu\text{m}$ , locally forming a short sterile band, clavate, covered with fairly few, unevenly spaced, coarse, simple to furcate, straight to curved excrescences  $2.5-8 \times 1.8-2.5 \,\mu\text{m}$ . Pleurocystidia absent. Lamellar trama weakly brownish vinescent in Melzer's reagent. Hyphae of the pileipellis  $2.5-3.5 \,\mu\text{m}$  wide, clamped, embedded in gelatinous matter, smooth. Hyphae of the cortical layer of the stipe  $1.3-2.5 \,\mu\text{m}$  wide, clamped, easily separable from each other (indicating the presence of gelatinous matter), smooth or with a few scattered, coarse excrescences, the terminal cells  $3.5-13.5 \,\mu\text{m}$  wide, fusiform to clavate, simple to furcate, diverticulate, excrescences  $2.5-6.5 \times 1-3.5 \,\mu\text{m}$ .

Material examined. INDIA: Punjab, Kulu valley, Manali, 26 Aug. 1964, C. Bas 4257, on decaying stump in Cedrus-Picea forest, c. 2200 m alt. (No. 969.134-074b; L).

The macroscopic description of the species is adapted from the collector's notes, complemented by my own observations on the dried material. The microscopic details are based on reexamination of the collection cited above.

European specimens of Mycena tintinnabulum are noted for the thick pellicle covering the pileus, rendering the latter somewhat viscid when wet. This gelatinous layer was not observed in the fresh material, but is easily seen under the microscope. Another difference is that European tintinnabulum is known to grow on wood of deciduous trees, whereas the Indian collection was found in a coniferous forest.



Figs. 11–16. Mycena abietina (Watling 13081; E). 11. Immature basidium; 12. spores; 13. cheilocystidia; 14. hyphae of the pileipellis; 15. hypha of the cortical layer of the stipe; 16. terminal cells. — Figs. 17–23. Mycena bathyrrhiza (Abraham K 1503; K). 17. Habit sketch (by the collector; c. × 0.4); 18. basidia; 19. spores; 20. cheilocystidia (from middle of lamella); 21. cheilocystidia (from near the pileus margin); 22. hyphae of the pileipellis; 23. hypha of the cortical layer of the stipe. (All figures, except Fig. 17, × 700.)

## Mycena sect. Fragilipedes (Fr.) Quél.

Agaricus [sect.] Fragilipedes Fr., Epicr. Syst. mycol. (1838) 108. — Mycena [sect.] Fragilipedes (Fr.) Quél., Mém. Soc. Emul. Montbél. II 5 (1872) 105. — Lectotype: Agaricus alcalinus Fr. sensu Kühn., 1938. For further synonymy, see Maas Geesteranus (1988a: 43).

## 3. Mycena abietina Maas G., spec. nov.1 — Figs. 11-16

Basidiomata sparsa. Pileus 8-12 mm latus, campanulatus, umbonatus, pruinosus, sulcatus, siccus umbrinus, centro obscurior. Caro tenuis, pileo concolor, odore saporeque ignotis. Lamellae c. 21 stipitem attingentes, molles, adscendentes, c. 1 mm latae, late adnatae, dente decurrentes, albidae, margine convexo, concolore. Stipes c.  $35 \times 1.5$  mm, cavus, aequalis, cylindraceus, minute puberulus, pileo concolor.

Basidia c.  $30 \times 9 \,\mu\text{m}$ , clavata, 4-sporigera, fibulata. Sporae  $9.8-11.6 \times 5.4-6.4 \,\mu\text{m}$ , inaequilateraliter ellipsoideae, leves, amyloideae. Cheilocystidia  $45-63 \times 8-18 \times 3.5-4.5 \,\mu\text{m}$ , lageniformia, fibulata, levia. Pleurocystidia similia. Hyphae pileipellis  $2-2.7 \,\mu\text{m}$  latae, fibulatae, surculis  $2.5-9 \times 1.3-1.8 \,\mu\text{m}$  praeditae, haud gelatinosae. Hyphae stipitis corticales  $1.3-3.5 \,\mu\text{m}$  latae, fibulatae, surculis sparsis  $2.5-7 \times 1.8-2.5 \,\mu\text{m}$  instructae, haud gelatinosae, cellulae terminales c.  $45 \times 2.5-3.5 \,\mu\text{m}$ , surculis  $4.5-20 \times 2-3.5 \,\mu\text{m}$  munitae.

Ad truncum coniferis muscosum.

Holotypus: 'Mycena cf. atrocyanea', R. Watling 13081 (E).

<sup>1)</sup> Etymology: abietinus, applied to cryptogams which grow on firs (or conifers in general).

Basidiomata scattered. Pileus 8-12 mm across, campanulate, with a small umbo, pruinose, sulcate, dark grey-brown, blackish at the centre. Flesh thin, concolorous with the pileus. Odour and taste unknown. Lamellae c. 21 reaching the stipe, tender, ascending, c. 1 mm broad, broadly adnate, decurrent with a short tooth, whitish, with convex, concolorous edge. Stipe c.  $35 \times 1.5$  mm, hollow, equal, terete, minutely and sparsely puberulous, concolorous with the pileus.

Basidia c.  $30 \times 9~\mu m$ , clavate, a few seen with four incipient sterigmata, clamped. Spores  $9.8-11.6 \times 5.4-6.4~\mu m$ , pip-shaped, smooth, amyloid. Cheilocystidia  $45-63 \times 8-18 \times 3.5-4.5~\mu m$ , forming a sterile band (lamellar edge homogeneous), lageniform, stalked or not, clamped, smooth, at times with a furcate neck. Pleurocystidia similar, infrequent. Lamellar trama brownish vinescent in Melzer's reagent. Hyphae of the pileipellis  $2-2.7~\mu m$  wide, clamped, not gelatinized, at first covered with widely spaced, simple, straight to curved or flexuous, cylindrical excrescences  $2.5-9 \times 1.3-1.8~\mu m$ , but finally very densely diverticulate. Hyphae of the cortical layer of the stipe  $1.3-3.5~\mu m$  wide, clamped, not gelatinized, sparsely covered with simple, cylindrical excrescences  $2.5-7 \times 1.8-2.5~\mu m$ , the terminal cells c.  $45 \times 2.5-3.5~\mu m$ , cylindrical, covered with fairly few, widely spaced, somewhat curved or flexuous excrescences  $4.5-20 \times 2-3.5~\mu m$ .

On moss-covered coniferous log.

Holotype: 'Flora of Kashmir-India / Mycena cf. atrocyanea / Gulmarg / 28 Sept. 1978 / Roy Watling 13081 / 6000 ft' (E).

Both the macroscopic description and the microscopic details are based on reexamination of the dried type material. The species is a member of section *Fragilipedes*.

Watling (apud Watling & Gregory, 1980: 556) doubtfully identified his collection with Mycena atrocyanea (Fr.) Gillet, and from his letter (June 1991) it becomes clear that M. atrocyanea sensu A.H. Smith was meant. The latter proved to be a new species and was subsequently described as Mycena coracina (Maas Geesteranus, 1988a: 69). Although no information is available on the colours of M. abietina in the fresh condition, it is not difficult to see that this species differs from M. coracina. Its stipe does not in the least give the impression of having been 'rigid and cartilaginous' as was stated for M. atrocyanea sensu Smith (1947: 255), but it may have been firm when fresh. In M. abietina, the spores (5.4-6.4 µm broad) are narrower than those of M. coracina (6.3-7.2 µm), the cheilocystidia are longer (45-63 µm) than those of M. coracina (27-45 um), while the terminal cells of the hyphae of the cortical layer of the stipe of M. abietina with their long, pennant-like excrescences look very different indeed. If it is assumed that the stipe of Watling's material was fragile when fresh, three further species should be considered: Mycena abramsii (Murrill) Murrill, M. alnicola A.H. Smith, and M. subcana A.H. Smith. Careful perusal of the pertinent descriptions, however, show all three to be different from M. abietina.

## 4. Mycena bathyrrhiza Maas G., spec. nov.2 — Figs. 17-23

Basidiomata congregata. Pileus > 20 mm latus, conico-campanulatus, sulcatus, pruinosus, glabrescens, griseus (siccatus fuscus), margine concolor. Caro tenuis, pallida, odore saporeque ignotis. Lamellae c. 20 stipitem attingentes, molles, adscendentes, usque ad 1.5 mm latae, anguste adnatae, albae, basi gri-

Etymology: βαθύσ, deep; ὑιζα, root.

seae, margine albae. Stipes 50(-80) × 5 mm, cavus, fragilis, aequalis, cylindraceus, apice minute pruinosus, deorsum glaber, levis, murinus, basi longe radicatus, fibrillis albis munitus.

Basidia (immatura)  $30-36\times7-9~\mu m$ , clavata, 2-(3-)sporigera, efibulata, sterigmatibus  $6.5-7~\mu m$  longis praedita. Sporae  $9.0-11.6\times5.8-7.2~\mu m$ , inaequilateraliter ellipsoideae, leves, amyloideae. Cheilocystidia  $27-60\times10-16\times5.5-8~\mu m$ , lageniformia, subclavata, subfusiformia, efibulata, levia vel apice furcata, pilei marginem versus surculis varieformibus instructa. Pleurocystidia nulla. Trama lamellarum iodi ope brunneo-vinescens. Hyphae pileipellis  $2.7-4.5~\mu m$  latae, efibulatae, surculis  $1.8-8\times1.5-1.8~\mu m$ , simplicibus vel ramosis praeditae. Hyphae stipitis corticales  $1.8-3.5~\mu m$  latae, efibulatae, surculis  $2.5-3.5\times1.5-1.8~\mu m$  instructae.

Ad arborem putridum.

Holotypus: S.P. Abraham K 1503 (K).

Basidiomata in groups. Pileus > 20 mm across, conico-campanulate, sulcate, pruinose, glabrescent, greyish (fairly dark fuscous in the dried material), concolorous at the margin. Flesh thin, pallid. Odour and taste not recorded. Lamellae c. 20 reaching the stipe, tender, ascending, up to 1.5 mm broad, narrowly adnate ('adnexed' according to the collector), white, more greyish at the base, edge straight to shallowly convex, white. Stipe 50(-80) × 5 mm, hollow, fragile, equal, terete, apically minutely pruinose, glabrous farther below, smooth, mouse-greyish, the base extending into a long root, covered with white fibrils.

Basidia (immature)  $30-36\times7-9~\mu m$ , fairly slender-clavate, 2-spored, occasionally 3-spored, clampless, with sterigmata  $6.5-7~\mu m$  long. Spores  $9.0-11.6\times5.8-7.2~\mu m$ , broadly pip-shaped, smooth, amyloid. Cheilocystidia  $27-60\times10-16\times5.5-8~\mu m$ , forming a sterile band (lamellar edge homogeneous), lageniform, more infrequently subclavate or subfusiform, clampless, smooth or apically furcate (in the middle of the lamella) to apically more or less branched or covered with coarse, variously shaped and branched, excrescences  $6-15\times2-5~\mu m$  (near the margin of the pileus). Pleurocystidia absent. Lamellar trama brownish vinescent in Melzer's reagent. Hyphae of the pileipellis  $2.7-4.5~\mu m$  wide, clampless, covered with (somewhat gelatinized?) fairly coarse, simple to furcate or branched excrescences  $1.8-8\times1.5-1.8~\mu m$ . Hyphae of the cortical layer of the stipe  $1.8-3.5~\mu m$  wide, clampless, covered with simple, straight to somewhat curved excrescences  $2.5-3.5\times1.5-1.8~\mu m$ .

On decayed stumps.

Holotype: '[as Mycena alcalina] India, Jammu & Kashmir, Pahalgam / 22 June 1987 / S.P. Abraham K 1503' (K).

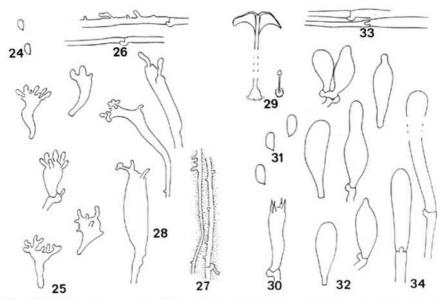
The macroscopic description of the species is adapted from the collector's notes, complemented by my own observations on the dried material. The microscopic details are based on reexamination of the type.

Mycena bathyrrhiza is a member of section Fragilipedes, distinct from any species thus far known in the Northern Hemisphere, and equally different from any 'Sikkim Himalaya' Mycenas described by Berkeley (1850, 1852).

## 5. Mycena coalita Maas G., spec. nov. 3 - Figs. 24-28

Basidiomata fasciculata. Pileus 11-18 mm latus, plane conicus, subumbonatus, haud lubricus, levis, translucente striatus, glaber, pallide argillaceus, marginem versus pallidior. Caro tenuis, albida, odore chlorino, sapore haud distincto. Lamellae 22-27 stipitem attingentes, molles, adscendentes, usque ad

<sup>3)</sup> Etymology: coalitus, joined together.



Figs. 24–28. Mycena coalita (Bas 4369; L). 24. Spores; 25. cheilocystidia; 26. hyphae of the pileipellis; 27. hyphae of the cortical layer of the stipe; 28. terminal cells. — Figs. 29–34. Mycena cinnabarina (Bas 4361; L). 29. Habitus of young specimen and section of old specimen (dried); 30. basidium; 31. spores; 32. cheilocystidia; 33. hyphae of the pileipellis; 34. terminal cells of hyphae of the cortical layer of the stipe. (Fig. 29, × 2; all others, × 700.)

2 mm latae, late adnatae, dente decurrentes, pallide cremeo-argillaceae rosco-tinctae. Stipes  $50-75\times0.8-1.5$  mm, cavus, fragilis, aequalis, cylindraceus, apice floccoso-pruinosus, deorsum glaber, levis, pallide cremeus brunneolotinctus, basi radicatus, fibrillis pallidis dense instructus.

Basidia (immatura) c.  $18 \times 5.5 \,\mu\text{m}$ , clavata, 4-sporigera, fibulata. Sporae  $4.9-5.4 \times 2.4-2.8 \,\mu\text{m}$ , inaequilateraliter ellipsoideae, leves, amyloideae. Cheilocystidia  $14.5-22.5 \times 6.5-10 \,\mu\text{m}$ , clavata, fibulata, surculis crassis  $3.5-8 \times 1.5-3 \,\mu\text{m}$  munita. Pleurocystidia sparsa, similia. Trama lamellarum iodi ope plus minusve rubro-brunnea. Hyphae pileipellis  $2.5-4.5 \,\mu\text{m}$  latae, fibulatae, leves vel surculis sparsis praeditae, haud gelatinosae. Hyphae stipitis corticales  $1.5-2.5 \,\mu\text{m}$  latae, fibulatae, maxima ex parte leves, materiam gelatinosam immersae, cellulae terminales  $5.5-10.5 \,\mu\text{m}$  latae, apicibus diverticulatis.

In nemore frondoso.

Holotypus: C. Bas 4369 (No. 964.289-279; L).

Basidiomata fasciculate. Pileus 11-18 mm across, shallowly conical, with little pronounced or centrally somewhat depressed umbo, not lubricous, smooth, translucent-striate, glabrous, pale beige to beige-brown at the centre, very pale beige at the margin. Flesh thin, watery whitish. Odour chlorine-like, more pronounced when crushed. Taste indistinct. Lamellae 22-27 reaching the stipe, tender, ascending, becoming almost horizontal, up to 2 mm broad, broadly adnate, decurrent with a tooth, very pale creamy beige with some pinkish tint, the edge shallowly convex to straight, concolorous. Stipe  $50-75\times0.8-1.5$  mm, hollow, fragile, equal, terete, apically sparsely floccose-pruinose, glabrous and polished farther below, watery pale cream with some brownish tint, in the

end becoming pale brownish, the base rooting, dull brown, densely covered with long, coarse, whitish fibrils.

Basidia (none seen mature) c.  $18 \times 5.5 \,\mu m$ , clavate, with 4 incipient sterigmata, clamped. Spores  $4.9-5.4 \times 2.4-2.8 \,\mu m$ , pip-shaped, smooth, amyloid. Cheilocystidia  $14.5-22.5 \times 6.5-10 \,\mu m$ , forming a sterile band (lamellar edge homogeneous), clavate to more or less irregularly shaped, clamped, covered with fairly few, unevenly spaced, coarse, simple to furcate, cylindrical to more or less inflated excrescences  $3.5-8 \times 1.5-3 \,\mu m$ . Pleurocystidia scarce, similar. Lamellar trama staining somewhat reddish-brownish in Melzer's reagent. Hyphae of the pileipellis  $2.5-4.5 \,\mu m$  wide, clamped, smooth or sparsely covered with cylindrical excrescences  $2-6.5 \times 1-2 \,\mu m$ , not gelatinized. Hyphae of the cortical layer of the stipe  $1.5-2.5 \,\mu m$  wide, clamped, somewhat embedded in gelatinous matter, smooth for the greater part, terminally with scattered warts or short cylindrical excrescences  $1-2.5 \times 1-1.5 \,\mu m$ , terminal cells infrequent,  $5.5-10.5 \,\mu m$  wide, cylindrical to more or less inflated, apically with variously shaped, simple to branched excrescences.

In forest of Ouercus incana-Rhododendron arboreum.

Holotype: 'Fungi of India / Mycena coalita Maas G. / Uttar Pradesh, Mussoorie, near Charlesville / 13 Sept. 1964 / C. Bas 4369 / c. 2000 m alt., growing from under a rock' (No. 964.289-279; L).

The macroscopic description of this member of section *Fragilipedes* is adapted from the collector's notes, complemented by my own observations on the dried material. The microscopic details are based on reexamination of the type.

The slightly pinkish-tinted lamellae and the short, coarsely diverticulate cheilocystidia of the present species may remind one of *Mycena pseudo-inclinata* A.H. Smith, a species of North America (Maas Geesteranus, 1988b: 155). This species, however, has a differently coloured pileus, a subfarinaceous odour, a dry stipe, and bigger spores.

Berkeley (1852: 101) described an Agaricus colligatus from Sikkim which, like Mycena coalita, is characterized by caespitose habit, pale pileus, and narrow, nearly white lamellae. But a glance at the cheilocystidia of A. colligatus (Maas Geesteranus, 1982: 528) and the fact that its lamellae are truly arcuate quickly learn that this species must be different from Mycena coalita.

## Mycena sect. Hygrocyboideae (Fr.) Sing.

Agaricus [sect.] Hygrocyboideae Fr., Syst. mycol. 1 (1821) 155. — Mycena sect. Hygrocyboideae (Fr.) Sing., Beih. Sydowia 7 (1973) 49. — Lectotype: Mycena epipterygia (Scop.: Fr.) S.F. Gray. For further synonymy, see Maas Geesteranus (1989a: 89).

Berkeley (1852: 103) recorded *Agaricus epipterygius* from Sikkim and cited Hooker's numbers 9 and 17. In both collections the tissues are badly collapsed, they differ from each other and neither represents *Mycena epipterygia*.

## Mycena sect. Calodontes (Fr. ex Berk.) Quél.

Agaricus subtrib. Calodontes Fr., Syst. mycol. 1 (1821) 111. — Agaricus [sect.] Calodontes Fr. ex Berk., J.E. Smith, Engl. Flora 5 (2) (1836) 43. — Mycena [sect.] Calodontes (Fr. ex Berk.) Quél., Mém. Soc. Emul. Montbél. II 5 (1872) 102. — Lectotype: Agaricus pelianthinus Fr.

For further synonymy, see Maas Geesteranus (1989b: 480).

## Mycena sect. Calodontes subsect. Marginatae J.E. Lange

Mycena sect. Ciliatae [subsect.] Marginatae J.E. Lange, Dansk bot. Ark. 1 (5) (1914) 13, 18. — Lectotype: Mycena pelianthina (Fr.) Quél.

For further synonymy, see Maas Geesteranus (1989b: 481).

## 6. Mycena cinnabarina Maas G., spec. nov.4 — Figs. 29-34

Basidiomata sparsa. Pileus 5–12 mm latus, campanulatus, convexus vel plano-convexus, centro applanatus vel depressus, siccus vel sublubricus, initio levis, deinde sulcatus, translucente striatus, glaber, hygrophanus, cinnabarinus, centro obscurior, atratus, siccus pallidior, centro ochraceo-roseus. Caro tenuis, pileo concolor, odore saporeque raphanoideis. Lamellae 15–21 stipitem attingentes, molles, adscendentes, usque ad 1.5 mm latae, late adnatae, decurrentes, roseae, margine concavo, cinnabarino. Stipes 13–58 × 0.2–0.7 mm, fistulosus, fragilis, aequalis, cylindraceus, apice minute sparseque puberulus, deorsum glaber, levis, lubricus, pileo concolor, basi bulbosus roseo-tomentosus.

Basidia  $22.5-24\times6.5-7~\mu m$ , clavata, 4-sporigera, fibulata, sterigmatibus  $5.5~\mu m$  longis praedita. Sporae  $6.7-8.1\times3.6-4.2~\mu m$ , inaequilateraliter ellipsoideae, leves, amyloideae. Cheilocystidia  $22.5-35\times6.5-10~\mu m$ , clavata, fusiformia, lageniformia, fibulata, pigmento rubro repleta, levia. Pleurocystidia similia. Hyphae pileipellis  $2.5-4.5~\mu m$  latae, fibulatae, leves. Hyphae stipitis corticales  $2.5-3.5~\mu m$  latae, fibulatae, leves, cellulae terminales  $30-80\times6.5-13.5~\mu m$ , clavatae, subcylindraceae, fibulatae, leves.

Ad Quercus incanae folia dejecta. Holotypus: C. Bas 4361 (No. 964.298-385; L).

Basidiomata scattered. Pileus 5-12 mm across, at first campanulate, then convex to plano-convex, at the centre becoming flattened or depressed, dry or somewhat lubricous, smooth when young, later sulcate, glabrous, translucent-striate, hygrophanous, fresh deep cinnabar red, darker at the centre to almost blackish, on drying becoming much paler, with ochraceous-pink centre. Flesh thin, concolorous with the pileus. Odour weak but raphanoid when crushed. Taste raphanoid. Lamellae 15-21 reaching the stipe, tender, ascending, up to 1.5 mm broad, broadly adnate and decurrent, smooth, occasionally intervenose, reddish pink, the edge concave, dark cinnabar red. Stipe  $13-58\times0.2-0.7$  mm, hollow, fragile, equal, terete, more or less curved, smooth, apically minutely and sparsely puberulous, glabrous farther below, lubricous, concolorous with the pileus, the base bulbous and covered with pinkish tomentum

Basidia  $22.5-24\times6.5-7$  µm, clavate, 4-spored, clamped, with sterigmata 5.5 µm long. Spores  $6.7-8.1\times3.6-4.2$  µm, pip-shaped, smooth, amyloid. Cheilocystidia  $22.5-35\times6.5-10$  µm, forming a sterile band (lamellar edge homogeneous), clavate, fusiform, lageniform, clamped, with red contents, smooth. Pleurocystidia similar. Hyphae of the pileipellis 2.5-4.5 µm wide, clamped, with red contents, smooth. Hyphae of the cortical layer of the stipe 2.5-3.5 µm wide, clamped, with red contents, smooth, the terminal cells  $30-80\times6.5-13.5$  µm, clavate to subcylindrical, clamped, with red contents (?), smooth.

Growing on fallen leaves of Quercus incana.

Holotype: 'Fungi of India / Mycena cinnabarina Maas G. / Uttar Pradesh, Mussoorie near Charlesville / 13 Sept. 1964 / C. Bas 4361 / c. 2000 m alt.' (No. 964, 298-385; L).

Etymology: cinnabarinus, vermilion.

The macroscopic description of the species is adapted from the collector's notes, complemented by my own observations on the dried material. The microscopic details are based on reexamination of the type.

All essential characters identify Mycena cinnabarina as a species of section Calodontes (Fr. ex Berk.) Quél. subsect. Marginatae J.E. Lange, but two features separate it from other members of the subsection. One of the differences is that the base of the stipe of M. cinnabarina is swollen to form a conspicuous bulb. The other, even more striking, difference is in the presence of copious red pigment inside all surface hyphae of pileus and stipe. Probably some of this matter is also excreted on the outside of the hyphal walls which may explain the lubricous feel of pileus and stipe noted by the collector. Under the microscope, however, no gelatinization of the hyphal walls was observed. The red contents of the surface hyphae, dissolving in a dark violet cloud in diluted KOH, is so concentrated as to render it difficult to check whether or not this matter also occurs in the hyphae of the tramal tissues. It is equally difficult to observe whether the lamellar trama stains a purplish colour in Melzer's reagent.

Red is not a colour commonly seen in Mycena, so any red species lacking important details in its original description will require scrutiny. Some examples follow here.

From Darjeeling in India and at an elevation only a little higher than that of Mussoorie, Berkeley (1850: 79) described Agaricus (Mycena) rubiaetinctus, a fungus almost as red as Mycena cinnabarina, and thus requiring a closer look. It can be told from M. cinnabarina by its lamellae (which are free), its apparent lack of pleurocystidia (Maas Geesteranus, 1982: 535), its longer cheilocystidia ( $55-65 \mu m$ ), and its different habitat (on trunks of trees).

An orange to red species of Mycena is known from Java as Mycena subacicula P. Henn. (Hennings, 1900: 157). It is easily distinguishable from M. cinnabarina on account of its small number of lamellae (eight reaching the stipe) and its subglobose spores (stated to measure  $3.5 \mu m$ ).

Mycena fuyoensis Imai (Imai, 1941: 448) attracts the attention on account of its red pileus, but the species probably is a member of section Adonideae (Maas Geesteranus, 1991b; 401) rather than subsection Marginatae.

Mycena aurantiorubra Métrod (1949: 88) described from Madagascar can be told from M. cinnabarina by its narrow lamellae with apparently concolorous edges, thick-walled cheilocystidia, and diverticulate hyphae of the pileipellis.

Mycena praeclara Horak (1978: 23), although not considered a member of Mycena as I see this genus, may be briefly mentioned on account of its orange-red pileus. Its hymeniform pileipellis separates it at once from M. cinnabarina.

Stevenson (1964: 48) described a *Mycena minirubra* from New Zealand which, to judge from the somewhat meagre data supplied, could well be a member of subsection *Marginatae*, too. It differs from *M. cinnabarina* in having a much smaller pileus (0.5–1 mm across; *cinnabarina*: 5–12 mm), apricot lamellae (*cinnabarina*: reddish pink), a much shorter stipe (3–5 mm; *cinnabarina*: 13–58 mm), and a white basal disc (*cinnabarina*: basal part of the stipe bulbous and covered with pinkish tomentum).

Yet another red species from New Zealand described by the same author (Stevenson, 1964: 54) is *Mycena miniata* (a later homonym, not *M. miniata* Petch, 1917) which can be separated from *M. cinnabarina* by its non-decurrent lamellae, lack of a bulbous swelling at the base of the stipe, more broadly pip-shaped spores ( $8 \times 6 \mu m$ ), and awl-shaped cheilocystidia.

### Mycena sect. Calodontes subsect. Purae (Konr. & Maubl.) Maas G.

Mycena sect. Purae Konr. & Maubl., Ic. sel. Fung. 6 (1934) 269. — Mycena sect. Calodontes subsect. Purae (Konr. & Maubl.) Maas G., Persoonia 11 (1980) 112. — Lectotype: Mycena pura (Pers.: Fr.) Kummer.

For further synonymy, see Maas Geesteranus (1989b: 488).

Subsection *Purae* is a group of species characterized by pronounced colours ranging from pink and purple to violet and blue, but it should not be taken for granted that every pink-capped fungus belongs to the *Purae*. *Agaricus bicrenatus* is a case in point. Berkeley (1850: 79) described this species as "of a delicate pinkish white, with deeper shades on the apex of the pileus." He terminated by stating that "The colours are nearly those of *A. purus*." Manjula (1983: 86, 87) who has studied the type concluded that *Agaricus bicrenatus* "belongs to the section *Purae* Konrad and Maubl. and is close to *M. pura*...," although he had to admit that "the microstructures could not be observed." I adhere to my former view (Maas Geesteranus, 1982: 529) that the taxonomic identity of Berkeley's species is unknown.

### Mycena pura (Pers.: Fr.) Kummer f. pura — Figs. 35-38

Agaricus purus Pers., Neues Mag. Bot. (1794) 101; Fr., Syst. mycol. 1 (1821) 151. — Mycena pura (Pers.: Fr.) Kummer, Führ. Pilzk. (1871) 107, 110. — Type locality: Germany. For further synonymy, see Maas Geesteranus (1989b: 494).

Basidiomata scattered. Pileus 15-22 mm across, shallowly conical to planoconvex, sometimes with small umbo, smooth, translucent-striate, glabrous, not lubricous, very pale lilaceous pink to pale violet-grey, paler towards the margin. Flesh thin, concolorous with the pileus. Odour and taste strongly raphanoid when crushed. Lamellae 22-26 reaching the stipe, tender, at first ascending, becoming more horizontal with age, up to almost 3 mm broad, thin, adnate or somewhat decurrent with a short tooth, smooth, dorsally intervenose, very pale lilaceous pink to somewhat greyed lilaceous cream, the edge shallowly convex, concolorous. Stipe  $45-75 \times 1.5-2.2$  mm, hollow, equal for the greater part, broadened below, straight, curved near the base, terete, smooth, pruinose above, glabrous farther down, pale grey violaceous pink to very pale lilaceous pink, the base covered with few, coarse fibrils.

Basidia (none seen mature)  $20-22.5 \times 5.5-6.5 \,\mu\text{m}$ , clavate, 4-spored, clamped, with sterigmata c. 3.5  $\mu$ m long. Spores  $7.6-9.0 \times 3.6-4.5 \,\mu\text{m}$ , pip-shaped, smooth, amyloid. Cheilocystidia  $40-63 \times 9-20 \,\mu\text{m}$ , forming a sterile band (lamellar edge homogeneous), fusiform, subclavate, short- to long-stalked, clamped, smooth, apically broadly rounded. Pleurocystidia fairly numerous, similar. Lamellar trama brownish vinescent in Melzer's reagent. Hyphae of the pileipellis  $3.5-10 \,\mu\text{m}$  wide, clamped, smooth, not gelatinized. Hyphae of the cortical layer of the stipe  $1.8-2.7 \,\mu\text{m}$  wide, clamped, smooth, terminal cells (caulocystidia)  $8-12.5 \,\mu\text{m}$  wide, fusiform, smooth.

Material examined. INDIA: Uttar Pradesh, Mussoorie, Oakville, 16 Sept. 1964, C. Bas 4404, in forest of Quercus incana and Rhododendron arborea, with scattered Cedrus deodara, c. 2300 m alt. (No. 965.11-092; L).

The macroscopic description of the species is adapted from the notes accompanying collection *Bas 4404*. The microscopic details are based on reexamination of this dried material.

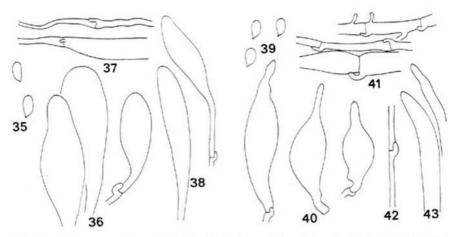


Fig. 35–38. Mycena pura f. pura (Bas 4404; L). 35. Spores; 36. cheilocystidia; 37. hyphae of the pileipellis; 38. terminal cells of hyphae of the cortical layer of the stipe. — Figs. 39–43. Mycena gentilis (Bas 4288; L). 39. Spores; 40. cheilocystidia; 41. hyphae of the pileipellis, overlying a hypha of the hypoderm; 42. hypha of the cortical layer of the stipe; 43. terminal cells. (All figures, × 700.)

Hooker's collection [Ser. 2] No. 28 from Sikkim which Berkeley (1852: 101) considered to represent Agaricus purus is not this species. None of the few spores observed prove to be amyloid, which excludes the species of subsection Purae. The hyphae of the cortical layer of the stipe appear to be smooth, but it is impossible to demonstrate the presence of caulocystidia, while the lamellar trama is inamyloid. This excludes subsection Violacellae. Lacking information on the cheilocystidia, further identification is not possible.

## Mycena sect. Calodontes subsect. Violacellae Sing. ex Maas G.

Mycena stirps Violacellae Sing., Agar. mod. taxon., 3rd ed. (1975) 395. — Mycena sect. Calodontes subsect. Violacellae Sing. ex Maas G., Persoonia 11 (1980) 112. — Lectotype: Mycena violacella (Speg.) Sing.

# 8. Mycena gentilis Maas G., spec. nov.5 — Figs. 39-43

Basidiomata sparsa. Pileus 11–20 mm latus, plano-convexus, udus sublubricus, initio levis, postea subsulcatus, translucente striatus, glaber, hygrophanus, lilacco-roseus, centro obscure vinaceo-brunneus, siccus pallidior. Caro tenuis, pileo concolor sed pallidior, odore saporeque raphanoideis. Lamellae 21–24 stipitem attingentes, molles, adscendentes, usque ad 3.5 mm latae, late adnatae, dente decurrentes, pallide lilaceo-roseae, margine convexo, pallidiore. Stipes 35–52 × 1–3 mm, cavus, fragilis, aequalis, cylindraceus, apice minute pruinosus, deorsum glaber, levis, siccus, pallide lilaceo-roseus, apice initio obscure purpureus, basi albidus vel pallide flavo-brunneus, fibrillis sparsis pallidisque instructus.

<sup>5)</sup> Etymology: gentilis, of the same tribe, alluding to the affinity with M. pearsoniana.

Basidia (immatura)  $20-22.5 \times 5.5-6.5~\mu m$ , clavata, 4-sporigera, fibulata, sterigmatibus c.  $3.5~\mu m$  longis munita. Sporae  $6.3-7.3 \times 4.0-4.7~\mu m$ , inaequilateraliter ellipsoideae, leves, inamyloideae. Cheilocystidia  $30-60 \times 10-18 \times 2-3.5~\mu m$ , fusiformia, fibulata, levia. Pleurocystidia nulla. Trama lamellarum iodi ope brunneovinescens. Hyphae pileipellis  $2.5-5.5~\mu m$  latae, fibulatae, leves vel surculis sparsis praeditae, haud gelatinosae. Hyphae stipitis corticales  $2-3.5~\mu m$  latae, fibulatae, leves, cellulae terminales  $3.5-9~\mu m$  latae, cylindraceae.

In Pineto.

Holotypus: C. Bas 4288 (No. 964.264-087; L).

Basidiomata scattered. Pileus 11-20 mm across, plano-convex, with age centrally somewhat depressed, somewhat lubricous when moist, at first smooth, later slightly sulcate, translucent-striate, glabrous, hygrophanous, vinaceous lilac-pink, at the centre dark vinaceous brown, pallescent with age, with the centre more ochraceous brownish. Flesh thin, paler concolorous with the pileus. Odour somewhat aromatic with raphanoid component. Taste strongly raphanoid. Lamellae 21-24 reaching the stipe, tender, ascending, becoming almost horizontal, up to 3.5 mm broad, thin, broadly adnate, decurrent with a tooth, smooth, dorsally intervenose, pale lilac-pink, the edge shallowly convex to straight, paler than the sides. Stipe  $35-52\times 1-3$  mm, hollow, fragile, equal, terete, apically delicately pruinose, glabrous farther below, smooth, dry, pale lilac-pink but apically dark purplish when young, the base whitish or pale brownish yellow, sparsely covered with pallid fibrils.

Basidia (none seen mature)  $20-22.5 \times 5.5-6.5~\mu m$ , clavate, 4-spored, clamped, with sterigmata c. 3.5  $\mu m$  long. Spores  $6.3-7.3 \times 4.0-4.7~\mu m$ , pip-shaped, smooth, non-amyloid. Cheilocystidia  $30-60 \times 10-18 \times 2-3.5~\mu m$ , fusiform with much narrowed apices, clamped, smooth. Pleurocystidia absent. Lamellar trama brownish vinescent in Melzer's reagent. Hyphae of the pileipellis  $2.5-5.5~\mu m$  wide, clamped, smooth for the greater part but with occasional cylindrical excrescences  $1-2~\mu m$  wide, neither gelatinized nor overlying gelatinous matter covering the hypoderm. Hyphae of the cortical layer of the stipe  $2-3.5~\mu m$  wide, clamped, smooth, the terminal cells  $3.5-9~\mu m$  wide, cylindrical.

Found in a plantation of Pinus roxburghii.

Holotype: 'Fungi of India / Mycena gentilis Maas G. / Uttar Pradesh, Dehra Dun, New Forest / 5 Sept. 1964 / C. Bas 4288 / c. 600 m alt.' (No. 964.264-087; L).

The macroscopic description of the species is adapted from the collector's notes, complemented by my own observations on the dried material. The microscopic details are based on reexamination of the type.

The specific epithet has been chosen in view of the resemblance to  $Mycena\ pearsonia-$ na, with which the specimens were doubtfully identified in the field. However,  $Mycena\ gentilis$  differs from M. pearsoniana in (1) the shorter spores (7.2–9.0  $\mu$ m in European material of M. pearsoniana), (2) the conspicuously narrowed apices of the cheilocystidia, and (3) the absence of gelatinous matter covering the hypoderm. The pallid base of the stipe of M. gentilis may prove to be yet another differential character.

Mycena auroricolor (Berk. & Br.) Petch as redescribed by Pegler (1986: 198) has the pale pink colours in common with M. gentilis but differs in being a member of subsection Purae (Konr. & Maubl.) Maas G., characterized among other features by amyloid spores.

### Mycena sect. Hiemales Konr. & Maubl.

Mycena [sect.] Hiemales Konr. & Maubl., Ic. sel. Fung. 6 (1934) 274. — Lectotype: Marasmiellus hiemalis (Osb. apud Retz.) Sing.

For further synonymy, see Maas Geesteranus (1991a: 81).

## Mycena sect. Hiemales subsect. Hiemales Maas G.

Mycena sect. Hiemales subsect. Hiemales Maas G., Persoonia 11 (1980) 114. — Type species: Mycena hiemalis (Osb. apud Retz.) Quél.

For further synonymy, see Maas Geesteranus (1991a: 82).

## 9. Mycena olida Bres. - Figs. 44-49

Mycena olida Bres., Fungi trid. 1 (1887) 73, pl. 79 fig. 1.; Icon. mycol. 5 (1928) pl. 240 fig. 1. — Marasmiellus olidus (Bres.) Sing., Lilloa 22 (1951 ['1949']) 302. — Holotype in S.

For further synonymy, see Maas Geesteranus (1991a: 86).

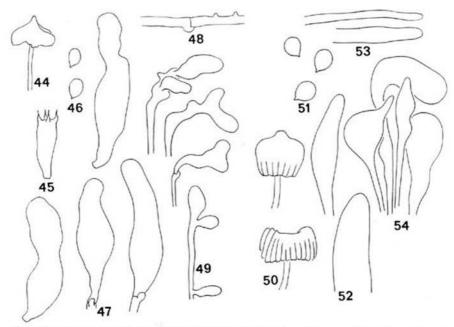
Basidiomata scattered. Pileus 3.5-8 mm across, parabolical to campanulate, with small, acute umbo, not sulcate, translucent-striate, glabrous, not lubricous, ivory white to somewhat brownish, with concolorous margin. Flesh thin, watery concolorous. Odour and taste indistinctive. Lamellae 14-16 reaching the stipe, tender, ascending, up to 2 mm broad, thin, narrowly adnate, smooth, white, the edge convex, concolorous. Stipe  $22-32\times0.4-0.6$  mm, hollow, equal, straight, curved below, terete, smooth, pruinose to minutely puberulous all over, watery white to cream, the base covered with long, coarse, white fibrils.

Basidia (few seen mature) c.  $22.5 \times 7-8 \,\mu\text{m}$ , clavate, 4-spored, clamped, with sterigmata  $4.5-5.5 \,\mu\text{m}$  long. Spores  $6.3-7.2 \times 4.9-6.0 \,\mu\text{m}$ , fairly broadly pip-shaped, smooth, inamyloid. Cheilocystidia  $46-52 \times 8-15 \times 5.5-11.5 \,\mu\text{m}$ , occurring mixed with the basidia (lamellar edge heterogeneous), fusiform, irregularly subcylindrical, clamped, smooth, apically broadly rounded. Pleurocystidia similar. Lamellar trama not vinescent in Melzer's reagent. Hyphae of the pileipellis  $2.7-5 \,\mu\text{m}$  wide, clamped, smooth except for a few isolated wart-like excrescences. Hyphae of the cortical layer of the stipe  $1.5-2.5 \,\mu\text{m}$  wide, clamped, smooth, terminal cells (caulocystidia)  $22-50 \times 5.5-9 \,\mu\text{m}$ , clavate, simple or lobed to somewhat branched, smooth.

Material examined. INDIA: Uttar Pradesh, between Mussoorie and Balansar, 22 Sept. 1964, C. Bas 4457, on moss-covered base of Quercus incana. c. 1800 m alt. (No. 965.11-019; L).

The macroscopic description of the species is adapted from the notes accompanying collection *Bas 4457*, complemented by my own observations on the dried specimens. The microscopic details are based on reexamination of this material.

My assumption that the four-spored form of *M. olida* would possess clamps (Maas Geesteranus, 1991a: 88) is confirmed by the present material. Mr. Th. Münzmay, a correspondent in Dormagen (Germany), asserted in a recently received letter that he found the four-spored form in his surroundings to be more common than the two-spored form.



Figs. 44–49. Mycena olida (Bas 4457). 44. Habitus; 45. basidium; 46. spores; 47. cheilocystidia; 48. hypha of the pileipellis; 49. caulocystidia. — Figs. 50–54. Hydropus eburneus (Bas 4147). 50. Habitus of young specimen (with umbo) and old one (with the centre depressed); 51. spores; 52. cheilocystidia; 53. hyphae of the pileipellis; 54. caulocystidia. (Fig. 44, × 2.5; Fig. 50, × 3; all others, × 700.)

### HYDROPUS Kühn. ex Sing.

Mycena, unnamed group Hydropus Kühn., Genre Mycena: (1938) 531 (not validly publ.). — Hydropus Kühn. ex Sing., Pap. Mich. Acad. Sci. 32 (1946) 127. — Type species: Agaricus fuliginarius Batsch.

## 10. Hydropus eburneus Maas G., spec. nov.6 - Figs. 50-54

Basidiomata sparsa. Pilcus 12-17 mm latus, obovoideus, parabolicus, e umbonato planoconvexus vel subdepressus, initio subviscidus, demum siccus, centro subsquamulosus, marginem versus fibrillosus, sulcatus, translucente striatus, subhygrophanus, udus eburneus, centro brunneus. Caro tenuis, pallida, odore saporeque tenuibus. Lamellae c. 23 stipitem attingentes, molles, adscendentes, vix 1 mm latae, liberae vel anguste adnatae, albae vel dilute brunneolae, margine convexo concolore. Stipes  $18-32\times0.7-1.5$  mm, cavus, aequalis, cylindraceus, apice minute pruinoso-flocculosus, albus vel subeburneus, basi incrassatus?

Basidia c. 25 × 7 μm, clavata, 4-sporigera, fibulata, sterigmatibus 4.5 μm longis instructa. Sporae 6.7–8.1 × 5.8–7.1 μm, globosae vel subglobosae, leves, amyloideae. Cheilocystidia 9–13.5 μm lata,

Etymology: eburneus, ivory coloured.

rara, fusiformia. Pleurocystidia nulla. Trama lamellarum iodi ope haud vinescens. Hyphae pileipellis  $3.5-6.5~\mu m$  latae, fibulatae, leves, pigmento brunneolo repletae. Hyphae stipitis corticales  $2-3.5~\mu m$  latae, fibulatae, leves, caulocystidia  $40-65\times 8-15~\mu m$ , clavata, fibulata, levia, longe pediculata.

In pratis.

Holotypus: C. Bas 4147 (No. 969.134-074a; L).

Basidiomata scattered. Pileus 12–17 mm across, obovoid to parabolical, then semi-globose with small umbo, finally planoconvex or with the centre somewhat depressd, at first somewhat viscid, then dry with slightly squamulose centre, appressed-fibrillose towards the margin, sulcate, translucent-striate, somewhat hygrophanous, more or less ivory coloured when moist, drying very pale buff, with brown centre and faintly brownish appressed fibrils. Flesh thin, pallid. Taste faint, somewhat chemical, odour faint, slightly aromatic-fungoid. Lamellae c. 23 reaching the stipe, tender, ascending, hardly 1 mm broad, fairly thick, free or narrowly adnate, white to very pale dingy buff, the edge convex, concolorous. Stipe  $18-32\times0.7-1.5$  mm, hollow, equal, terete for the greater part, apically minutely pruinose-flocculose, white to somewhat ivory coloured, with swollen base?

Basidia (none seen mature) c.  $25 \times 7~\mu m$ , clavate, 4-spored, clamped, with sterigmata 4.5  $\mu m$  long. Spores (abundant)  $6.7-8.1 \times 5.8-7.1~\mu m$ , globose to subglobose, smooth, amyloid, with pronounced apiculus. Cheilocystidia (very few seen)  $9-13.5~\mu m$  broad, fusiform, smooth, little projecting. Pleurocystidia absent. Lamellar trama not vinescent in Melzer's reagent. Pileipellis an epicutis with repent, radially aligned, smooth hyphae,  $3.5-6.5~\mu m$  wide, with brownish vacuolar contents. Pileocystidia not observed. Hyphae of the cortical layer of the stipe  $2-3.5~\mu m$  wide, clamped, smooth. Caulocystidia  $40-65 \times 8-15~\mu m$ , clavate, clamped, smooth, in part geniculate, long-stalked. Trama of the stipe sarcodimitic, containing fusiform, unbranched hyphae, at least 300  $\mu m$  long and  $20-27~\mu m$  wide, with metachromatic cell-walls.

In grass land grazed by cattle.

Holotype: 'Fungi of India / Hydropus eburneus Maas G. / Punjab, Kulu Valley / 17 Aug. 1964 / C. Bas 4147 / c. 1200 m alt. (No. 969.134-074a; L).

The macroscopic description of the species is adapted from the collector's notes, complemented by my own observations on the dried material. The microscopic details are based on reexamination of the type.

Apparent lack of pileocystidia, presence of hyphae of the pileipellis with brownish vacuolar contents, amyloid spores, and absence of pleurocystidia identify the present species as a member of *Hydropus* sect. *Floccipedes* (Kühn.) ex Sing., subsect. *Spurii* (Kühn.) ex Sing. (Singer, 1962: 66; 1982: 112). The two species of this section thus far known – *Hydropus scabripes* (Murrill) Sing. and *H. taxodii* (Murrill) Sing. (Singer, 1986: 418) – possess pip-shaped spores and are different from *H. eburneus* in having dark flesh. *Hydropus scabripes*, moreover, can be told from *H. eburneus* by its much broader lamellae (3.5–6 mm, according to Smith, 1947: 236) and abundant cheilocystidia, while *H. taxodii* appears associated with *Taxodium*.

Pegler (1986: 183) gave a redescription of *Hydropus porphyrodes* (Berk. & Br.) Pegler from Sri Lanka which is characterized by subglobose spores of the same size as those of *H. eburneus*. One of the differences between the two species is that the lamellae of *H. porphyrodes* are described as 'adnate to adnato-decurrent ... 4–5 mm broad.'

There is another species with subglobose spores described by Hennings (1901: 334) from Saharanpur (India) as *Mycena conocephala* which has some characters rather similar to those of *H. eburneus*. Although the description by Hennings is rather too short to be of much use for identification, a few features are definitely not applicable to the present species, such as caespitose habitus, acute papilla crowning the conical and smooth pileus, and (apparently normally) adnate lamellae.

#### XEROMPHALINA Kühn. & Maire

Xeromphalina Kühn. & Maire, Bull. Soc. mycol. Fr. 50 (1934) 18 (footnote). — Type species: Omphalina campanella (Batsch: Fr.) Kummer (Donk, 1962: 300).

## 11. Xeromphalina aspera Maas G., spec. nov.7 — Figs. 55-61

Basidiomata fasciculata. Pileus (siccatus) 7–16 mm latus, campanulatus, tenuiter sulcatus, glaber, rubrobrunneus. Caro tenuis, supra pileo concolor, infra pallida, odore saporeque ignotis. Lamellae c. 21-25 stipitem attingentes, lentae, decurrentes, interdum furcatae, intervenosae, c. 1 mm latae, pileo pallidiores. Stipes  $30-50\times 1-2$  mm, fistulosus, lentus, aequalis pro maxima parte, cylindraceus, basi incrassatus usque ad 4 mm, omnino tomentosus, basi hirsutus, pileo pallidior potius flavobrunneus.

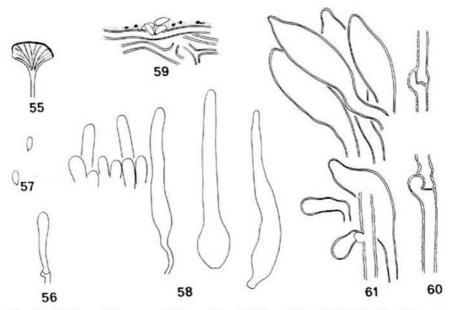
Basidia (immatura) c.  $23.5 \times 5.5 \,\mu m$ , clavata, 4-sporigera, fibulata, sterigmatibus c.  $3.6 \,\mu m$  longis instructa. Sporae  $4.9-5.6 \times 2.2-2.7 \,\mu m$ , inaequilateraliter ellipsoideae, leves, amyloideae. Cheilocystidia  $65-80 \times 7-11.5 \times 3.5-5.5 \,\mu m$ , sparsa, lageniformia vel subcylindracea, fibulata, levia. Pleurocystidia similia. Trama lamellarum iodi ope haud vinescens. Hyphae pileipellis  $1.8-4.5 \,\mu m$  latae, fibulatae, parietibus crassis instructae, leves sed materia gelatinosa sordibus sporisque obtectae, pileocystidiis destitutae. Hyphae stipitis corticales  $3.5-6.5 \,\mu m$  latae, fibulatae, parietibus crassis asperisque instructae, caulocystidia  $13.5-63 \times 7-12.5 \,\mu m$ , clavata vel fusiformia, fibulata.

Holotypus: Stainton, Sykes & Williams 7881 ('Mycena sanguinolenta'; K).

Basidiomata fasciculate. Pileus (dried) 7–16 mm across, campanulate, shallowly sulcate, glabrous, reddish brown. Flesh thin, rather tough, concolorous with the pileus under the surface, pallid farther below. Odour and taste unknown. Lamellae c. 21-25 reaching the stipe, fairly tough, decurrent, occasionally furcate, intervenose, c. 1 mm broad, paler than the pileus, lamellar edge arcuate, concolorous with the sides. Stipe  $30-50\times1-2$  mm, fistulose, tough, equal for the greater part, terete, gradually broadened towards the base up to 4 mm, matted tomentose throughout but hirsute at the base, paler than the pileus and rather more yellowish brown.

Basidia (none seen mature, springing from a dense and broad zone of intricately interconnected hyphae which may be the cause of the toughness of the lamellae) c.  $23.5 \times 5.5$   $\mu m$ , slender-clavate, 4-spored, clamped, with sterigmata c. 3.6  $\mu m$  long. Spores  $4.9-5.6 \times 2.2-2.7$   $\mu m$ , narrowly pip-shaped, almost cylindrical, smooth, amyloid. Cheilocystidia  $65-80 \times 7-11.5 \times 3.5-5.5$   $\mu m$ , not numerous, occurring mixed with the basidia, lageniform or subcylindrical, clamped, deep-seated, little projecting, smooth, thin-walled. Pleurocystidia similar. Lamellar trama not vinescent in Melzer's reagent. Hyphae of the pileipellis 1.8-4.5  $\mu m$  wide, clamped, thick-walled, smooth but covered with gelatinous matter, dirt, and spores, pileocystidia absent. Hyphae of the cortical layer of the stipe 3.5-6.5  $\mu m$  wide, clamped, thick-walled, lacking excrescences but very rough from

<sup>7)</sup> Etymology: asper, rough, referring to the rough, pigment-encrusted outer hyphae of the stipe.



Figs. 55-61. Xeromphalina aspera (Stainton, Sykes & Williams 7881; K). 55. Underside of flattened pileus, showing decurrent lamellae; 56. immature basidium; 57. spores; 58. cheilocystidia; 59. hyphae of the pileipellis; 60. hyphae of the cortical layer of the stipe; 61. caulocystidia. (Fig. 55, × 1; all others, × 700.)

pigment-incrustation, caulocystidia  $13.5-63 \times 7-12.5 \,\mu\text{m}$ , clavate to fusiform, clamped, fairly thick-walled, occurring mainly in the middle of the stipe and not infrequently in fascicles.

Trama of both pileus and stipe staining dark red-brown in KOH.

Found growing on a fallen tree in a wood.

Holotype: 'Flora of Nepal / [in pencil] Mycena sanguinolenta / Lete (S of Tukucha) Kali Gandaki / 8000 ft / 16.9.1954 / colour reddish-brown / Stainton, Sykes & Williams 7881' (K).

The description of the species is based entirely on my observations on the dried material. The only information on the colour given by the collectors is 'reddish-brown.'

Following Miller's subdivision of the genus (1968: 159), the present species belongs to section *Xeromphalina*, but it is difficult to make a choice between his subsections. Considering that the trama of the pileus stains a dark red-brown in KOH and the caulocystidia are fairly thick-walled and sometimes occur in fascicles, one would judge *X. aspera* to be a member of subsection *Mutabiles* A.H. Smith (later raised to section by Redhead, 1988: 480). However, the habit of the basidiomes in this subsection is solitary or gregarious to subcaespitose, and the caulocystidia are either narrow and hypha-like or irregular and contorted to branched. Fusiform caulocystidia similar to those of *X. aspera* are known to

occur in subsection Xeromphalina, but here these structures are thin-walled, while the pileus trama stains yellow in KOH.

Horak (1980: 104) described as a new species *Xeromphalina disseminata* from Sikkim which in view of its overall colouring could be mistaken for *X. aspera*. However, *X. disseminata* is stated to have a pileus 3–7 mm acrross (7–16 mm in dried condition in *X. aspera*), 8–14 lamellae (21–25 in *X. aspera*), a stipe which is pruinose at the apex (matted tomentose throughout in *X. aspera*), and rather larger spores,  $5-6.5 \times 3 \mu m$  (4.9–5.6 × 2.2–2.7 in *X. aspera*).

As is apparent from the title of his paper referred to above, Redhead placed the genus Xeromphalina in the family Xerulaceae. Having no intention just now to follow a path into what is largely terra incognita to me, I prefer to adhere to Singer's view (1986: 424) who regards the genus as a member of Fayod's tribus Myceneae.

#### REFERENCES

Berkeley, M.J. 1850. Decades of fungi (Decades XXV to XXX). Sikkim Himalaya fungi, collected by Dr. J.D. Hooker, Hooker's J. Bot. 2: 76–88.

Berkeley, M.J. 1852. Decades of fungi (Decades XXXVII, XXXVIII). Sikkim and Khassya fungi, collected by Dr. J. D. Hooker. Hooker's J. Bot. 4: 97–107.

Donk, M. A. 1962. The generic names proposed for Agaricaceae. Beih. Nova Hedwigia 5.

Hennings, P. 1900. Fungi monsunenses. O. Warburg, Monsunia 1.

Hennings, P. 1901. Fungi Indiae orientalis II cl. W. Gollan a. 1900 collecti. Hedwigia 40: 323-342.

Horak, E. 1978. Mycena rorida (Fr.) Quél. and related species from the Southern Hemisphere. Rep. Swiss bot. Soc. 88: 20-29.

Horak, E. 1980. Indian Boletales and Agaricales. Revisions and new taxa. Sydowia 33: 88-110.

Imai, S. 1941. Studia Agaricacearum japonicarum. II. Bot. Mag. Tokyo 55: 444–451.

Maas Geesteranus, R.A. 1982. Studies in Mycenas 72. Berkeley's fungi referred to Mycena-2. Proc. K. Ned. Akad. Wet. (C) 85: 527-539.

Maas Geesteranus, R.A. 1985. Conspectus of the Mycenas of the Northern Hemisphere-4. Section Mycena. Proc. K. Ned. Akad. Wet. (C) 88: 339-369.

Maas Geesteranus, R.A. 1988a. Conspectus of the Mycenas of the Northern Hemisphere-9. Section Fragilipedes, species A-G. Proc. K. Ned. Akad. Wet. (C) 91: 43-83.

Maas Geesteranus, R.A. 1988b. Conspectus of the Mycenas of the Northern Hemisphere-9. Section Fragilipedes, species J-R. Proc. K. Ned. Akad. Wet. (C) 91: 129-159.

Maas Geesteranus, R. A. 1989a. Conspectus of the Mycenas of the Northern Hemisphere-11. Section Hygrocyboideae. Proc. K. Ned. Akad. Wet. (C) 92: 89-108.

Maas Geesteranus, R.A. 1989b. Conspectus of the Mycenas of the Northern Hemisphere-13. Sections Calamophilae and Calodontes. Proc. K. Ned. Akad. Wet. (C) 92: 477-504.

Maas Geesteranus, R.A. 1991a. Conspectus of the Mycenas of the Northern Hemisphere-15. Sections Hiemales and Exornatae. Proc. K. Ned. Akad. Wet. 94: 81-102.

Maas Geesteranus, R.A. 1991b. Studies in Mycenas. Additions and Corrections, Part 1. Proc. K. Ned. Akad. Wet. 94: 377-403.

Manjula, B. 1983. A revised list of the agaricoid and boletoid basidiomycetes from India and Nepal. Proc. Indian Acad. Sci. (Plant Sci.) 92: 81–213.

Métrod, G. 1949. Les Mycènes de Madagascar (Mycena, Corrugaria, Pterospora). Prodr. flore mycol. Madagascar ... etc. 3.

Miller Jr., O.K. 1968. A revision of the genus Xeromphalina. Mycologia 60: 156-188.

Pegler, D.N. 1986. Agaric flora of Sri Lanka. Kew Bull. Addit. Ser. 12.

Redhead, S. A. 1988. Notes on the genus Xeromphalina (Agaricales, Xerulaceae) in Canada: biogeography, nomenclature, taxonomy. Can. J. Bot. 66: 479-507. Singer, R. 1962. Diagnoses fungorum novorum Agaricalium II. Sydowia 15: 45-83.
Singer, R. 1982. Hydropus (Basidiomycetes-Tricholomataceae-Myceneae). Flora neotrop. Monogr. 32.
Singer, R. 1986. The Agaricales in modern taxonomy. 4th ed. Koenigstein.
Smith, A. H. 1947. North American species of Mycena. Univ. Mich. Stud., Scient. Ser. 17.
Stevenson, G. 1964. The Agaricales of New Zealand: V. Kew Bull. 19: 1-59.
Watling, R. & N.M. Gregory. 1980. Larger fungi from Kashmir. Nova Hedwigia 32: 493-564.

#### PERSOONIA

Published by Rijksherbarium / Hortus Botanicus, Leiden Volume 15, Part 1, pp. 55-62 (1992)

## OCTOSPORA RUBENS AND OCTOSPORA RUSTICA IN THE NETHERLANDS (Pyronemataceae, Ascomycetes)

#### PETER BILLEKENS

Lodewijk van Nassaustraat 7, 5923 BC Venlo, The Netherlands

Two species of Ascomycetes, Octospora rubens and O. rustica, collected from the former Wambach clay pit in the municipality of Tegelen (prov. Limburg), are described and shown to be new to the Netherlands.

### Octospora rubens (Boud.) Moser — Figs. 1, 2

Humaria rubens Boud., Bull. Soc. mycol. Fr. 12 (1896) 13. — Humarina rubens (Boud.) Seaver, North Amer. Cup-fungi (Operc.) (1928) 127. — Humaria sanguinea Vel., Monogr. Discom. Boh. (1934) 325. — Octospora rubens (Boud.) Moser, Ascomyceten. In: H. Gams, Kl. Kryptog.-Fl. IIa (1963) 110.

Apothecia sessile, solitary or in small groups, 1-3.8 mm in diam., 0.5-1 mm high, gymnohymenial. Receptacle hemispherical or cup-shaped, then lenticular or convex, pastel red (Methuen, 7A4; see Kornerup & Wanscher, 1978), pale red (Methuen, 7A3), when juvenile with pink bloom, never orange or yellow, demarcated from the hymenium by a broad, finely serrated rim via a change in colour; the margin projecting over the hymenium, finely tomentose, never torn. Hymenium concave, flat, later either flat or convex, granular, orange red (Methuen, 8A7), tomato red (Methuen, 8C8), greyish red (Methuen, 9C5), or cardinal (red) (Methuen, 10D8), when juvenile often with a pinkish bloom, but never orange or yellow. Hypothecium 10-30 µm thick, clearly differentiated from medullary excipulum, cells sinuous, barrel-shaped, lobate, small, weakly cyanophilous, 2-8 µm broad (textura intricata), with very fine, concentrated granules of reddish intracellular pigment. Medullary excipulum 40-50 µm thick, towards the margin decreasing to 25 µm; cells small, lobate, subcylindrical, sinuous, irregular, thin-walled, weakly cyanophilous, 4-28 × 2-5 µm (textura intricata), with very fine concentrated granules of reddish intracellular pigment. Ectal excipulum easily distinguished from medullary excipulum, with very fine, diffuse granules of reddish intracellular pigment; 65-80 µm thick at base, somewhat attenuating upwards to 50-60 µm, then broadening again close to the margin up to 110-130 μm, with a wedge-shaped zone of textura intricata to textura epidermoidea; the cells are subcylindrical, sinuous, lobate, or irregular, 10-30 × 5-8 μm; at about 100 µm from the top of the margin a deep layer of irregularly arranged, angular cells, 15-45 x 5-9 µm (textura prismatica), running parallel to the outer surface of the apothecium, terminating over the entire width of the very broad margin in clavate end-cells 25-60 × 5.5-14 μm; the inner part of the margin projecting above the hymenium. Spores uniseriate, ellipsoid, hyaline, smooth, uninucleate, carminophobic, cyanophilous, mostly with a single large oil globule, sometimes with one large and several smaller ones, rarely with two large globules,  $16.4-18.5\times11.1-12.7~\mu m$ . Asci (4- to) 8-spored, operculate, cylindrical, gradually narrowing towards the apo- or pleurorhynchous base,  $214-276\times14-22~\mu m$ ; ending at different levels (range  $8-40~\mu m$ ), projecting above the paraphyses. Paraphyses straight or irregular, often branched,  $3.7-7.4~\mu m$ , at the top up to  $13.8~\mu m$ ; septate, often swollen at the septa, with homogeneous, round globules of reddish, intracellular, carotenoid pigment.

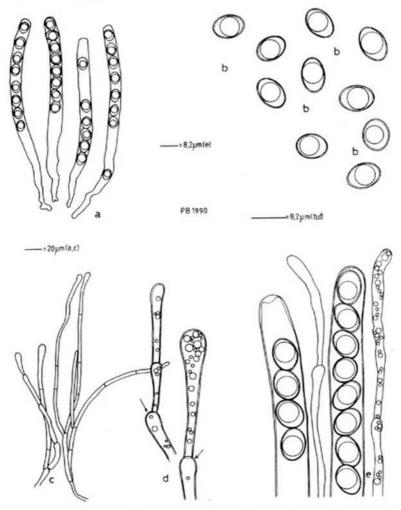


Fig. 1. Octospora rubens, a. Asci with contents; b. spores; c. paraphyses; d. upper parts of paraphyses with intracellular carotenoid pigment; e. parts of asci and paraphyses with one paraphysis showing contents.

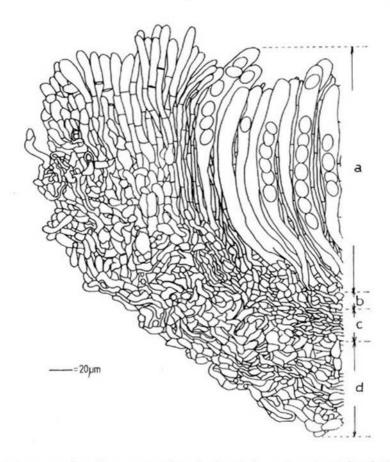


Fig. 2. Octospora rubens. Median section of margin of apothecium: a. hymenium; b. hypothecium; c. medullary excipulum; d. ectal excipulum.

Habitat and distribution. At the foot of an uncultivated slope, humus-deficient calcareous sand mixed with calcareous lime; associated with Ceratodon purpureus and Atrichum undulatum, close to Robinia pseudoacacia, Cytisus scoparius, and Quercus robur and amongst Rumex acetosella, Jasione montana, and Tussilago farfara; rather rare; fruiting throughout the year; Germany (Moser, 1963; Itzerott, 1981; Itzerott & Döbbler, 1982), United States (Seaver, 1928), Great Britain? (Dennis, 1978), France (Boudier, 1896; Grelet, 1932–1943; Caillet & Moyne, 1987), Czechoslovakia (Velenovský, 1934; Moravec, 1969) and now also the Netherlands.

Collections examined. The Netherlands: prov. Limburg, Tegelen, 17 Febr., 20 Mar., 28 Apr., 7 June and 19 Sept. 1990, P. Billekens (L and herbarium of the author).

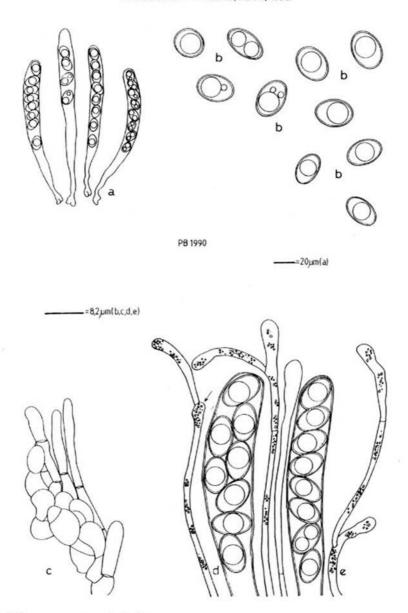


Fig. 3. Octospora rustica. a. Asci with ascospores; b. spores; c. ectal excipular cells with terminal, clavate end-cells forming the margin; d. upper halves of asci with contents; e. upper parts of paraphyses with intracellular carotenoid pigment.

The original description (Boudier, 1896) with accompanying illustrations of this species - under the name of Humaria rubens Boud. - as well as subsequent descriptions (Boudier, 1905-1910; Seaver, 1928; Grelet, 1932-1943; Velenovský, 1934 - as Humaria sanguinea Vel.; Moser, 1963; Moravec, 1969), with or without illustrations, agree well, as far as habitat, colour, size, macroscopic structure of receptacle, and most of the hymenial microscopic characters are concerned. However, some differences have to be noted. It is very puzzling that none of the above authors has stated anything about the crumbly structure of the hymenium which is caused by asci and paraphyses ending irregularly at different levels. This can even be seen with the help of a hand lens. Plate 396C of Boudier (1905-1910) shows a somewhat granular structure, but this is not mentioned in the text. In all specimens studied, this feature is clearly visible, in young as well as in fairly old fruit-bodies. Most of the above authors did not study excipular structures of Octospora rubens. Itzerott (1981) and Caillet & Moyne (1987), however, did, with Itzerott describing the ectal excipulum of O. rubens as textura globulosa or angularis and noting that it is mixed with broad hyphae. In addition, he observed a thick medullary excipulum and a hypothecium of textura intricata, whereas Caillet & Moyne classified the medullary and ectal excipula as textura intricata.

In the present study, both hypothecium and medullary excipulum were found to consist of tissues of textura intricata. The ectal excipulum can be clearly distinguished by a transition between textura intricata and textura epidermoidea, and by wider hyphae than in the medulla ( $5-8~\mu m$  vs.  $2-5~\mu m$ ). Comparison of these data with those from the literature corroborates Itzerott's experience that within a single species the structure of excipulum and hypothecium is variable; *Octospora rubens* is no exception! This comparison also shows that the Dutch material most closely resembles that studied by Caillet & Moyne (1987) from the French Jura.

## Octospora rustica (Vel.) Moravec - Figs. 3, 4

Humaria rustica Vel., Monogr. Discom. Boh. (1934) 327. — Octospora libussae Svrček & Kubička, Česká Mykol. 17 (1963) 65. — Octospora rustica (Vel.) Moravec, Česká Mykol. 23 (1969) 226.

Apothecia sessile, solitary or in groups; 0.5-2.2 mm in diam., 0.3-0.8 mm high, gymnohymenial. Receptacle invariably disc- or cup-shaped, margin smooth or very finely serrated, with outermost margin directed towards the substrate, almost smooth, very finely pruinose, occasionally with a few very obscure excipular hairs, unicolorous, not projecting over the hymenium. Hymenium smooth, invariably convex or nearly flat, pale orange (Methuen, 5A3), light orange (Methuen, 5A4), melon yellow (Methuen, 5A6), orange (Methuen, 5A7), dark orange (Methuen, 5A8), golden yellow (Methuen, 5B7), (Methuen, 11A6), or pastel pink (Methuen, 11A4). Hypothecium cells small, barrelshaped, lobate, irregularly angular,  $3-20\times2-8\,\mu\text{m}$  (textura intricata), with concentrated very small granules of orange-yellow intracellular pigment. Medullary excipulum cells inflated, lobate, regularly or irregularly rectangular,  $4-32\times2-17\,\mu\text{m}$  (textura intricata), with very small granules of concentrated orange-yellow intracellular pigment. Ectal excipulum cells isodiametrically subglobose or polygonal to irregularly angular,  $10-40\times6-30\,\mu\text{m}$  (textura globulosa to textura angularis), without intracellular and membrane pigment;  $100-175\,\mu\text{m}$  from the margin cells become abruptly rectangular,  $44-60\times9-15$ 

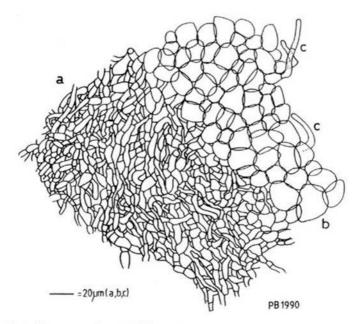


Fig. 4. Octospora rustica. a. Medullary excipulum; b. ectal excipulum; c. hyphoid hairs.

μm (textura prismatica); in the narrow margin which does not project over the hymenium these cells terminate in clavate end-cells,  $40-100\times8-17$  μm, with scattered hyphoid hairs. Hairs  $50-175\times5-8$  μm, hyaline, hyphoid, straight or sinuous, rounded at extremities; thin-walled (wall 0.4-0.8 μm), without septa or uni- to triseptate, with a 10-12.5 μm thick, bulbous base. Spores uni- or biseriate, ellipsoid, carminophobic, uninucleate, with one or occasionally two large oil globules, or with one large oil globule accompanied by one to three smaller; smooth,  $15-17.8\times7.6-11.9$  μm. Asci (4- to) 8-spored, operculate, cylindrical or occasionally clavate, non-amyloid, gradually narrowing to a pleurorhynchous base; comparatively small (compare Figs. 1a and 3a),  $144-212\times12-21$  μm. Paraphyses branching, occasionally forked; narrower below, 4.9-8.4 μm, broadening gradually towards upper ends, where they become club-shaped or irregular, 12-20 μm broad, septate, sometimes swollen at septa; varying from small to very small grains of orange-yellow intracellular carotenoid pigment, present over the entire length of the paraphyses.

Habitat and distribution. With regard to habitat, see O. rubens above. Rare; fruiting throughout the year; France (Caillet & Moyne, 1987), Germany (Itzerott, 1981), Great Britain (Dennis, 1978), Czechoslovakia (Velenovský, 1934; Svrček & Kubička, 1963; Moravec, 1969), Greenland (Dissing, 1982), and the Netherlands (this report).

Collections studied. The Netherlands: prov. Limburg, Tegelen, 28 Feb., 22 Mar., 6 May, 23 Sept. and 10 Oct. 1990, P. Billekens (L and herbarium of the author).

With the help of Itzerott's data, it was relatively easy to identify the material. The combination of characters immediately led to *O. rustica*. There is a great similarity between excipulum structure, hairs, asci, and paraphyses in the photographs (Figs. 1/10 and 18) in Itzerott's paper and my camera lucida drawings. One of Itzerott's illustrations clearly shows the broad medullary excipulum to consist of textura intricata. This is clearly demarcated from the broad textura globulosa/angularis of the ectal excipulum, to which virtually invisible hyphoid hairs are attached. Anatomically, and partly also morphologically, the above description corresponds well with Moravec's (1969) description of *Octospora libussae* Svrček & Kubička, especially his fig. 3.3 shows many striking similarities. There are no apparent differences between *O. rustica* and *O. libussae*, and I therefore agree with Caillet & Moyne (1987) in treating the latter name as a synonym of *O. rustica*. Anatomically, *O. rustica* is a very homogeneous and well-defined species.

The two species described are new records for the Netherlands. This, together with other previously described species (Billekens, 1985, 1988, 1989, 1990) justify the hope that more new records of Ascomycetes may be expected from the Dutch province of Limburg.

#### **ACKNOWLEDGEMENTS**

I wish to extend my thanks to Dr. J. van Brummelen (Leiden) for literature search and discussions, Dr. C. Bas (Leiden) for literature search, Mr. J. W. M. Jagt (Venlo) for translation into English of the manuscript, and the Sanders & Geraedts company (Swalmen) for access to the former clay pit.

#### REFERENCES

- Billekens, P. 1985. Caloscypha fulgens: een bekerzwam uniek in Nederland. Natuurhist, Maandbl. 74 (12): 231–234.
- Billekens, P. 1988. Lamprospora macracantha, een minuscule bekerzwam in de grote wereld. Natuurhist. Maandbl. 77 (6): 116–119.
- Billekens, P. 1989. Nieuwe bekerzwammen voor Nederland: Tricharina boudieri en Tricharina ochroleuca. Natuurhist. Maandbl. 78 (2): 26–30.
- Billekens, P. 1990. Nieuwe bekerzwammen voor Nederland II: Aleuria bicucullata, Cheilymenia aurea en Cheilymenia raripila. Natuurhist. Maandbl. 79 (9): 221–228.
- Boudier, E. 1896. Description de quelques nouvelles espèces de discomycètes de France. Bull. Soc. mycol. Fr. 22: 13, pls. 1, 2.
- Boudier, E. 1905-1910. Icones Mycologicae. Paris, Lausanne, reprint ed. 1981.
- Caillet, M. & G. Moyne. 1987. Contribution à l'étude du genre Octospora Hedw. ex S.F. Gray (Pezizales). Espèces à spores elliptiques ou fusiformes. Bull. Soc. mycol. Fr. 103: 200–202.
- Dennis, R.W.G. 1978. British Ascomycetes. Ed. 2. Vaduz: 57.
- Dissing, H. 1982. Operculate Discomycetes (Pezizales) from Greenland. In: G.A. Laursen & J.F. Ammirati, Arctic and Alpine Mycology. Washington D.C.
- Grelet, L.-J. 1932/43. Les Discomycètes de France d'après la classification de Boudier. Reprint ed. 1979.
  Itzerott, H. 1981. Die Gattung Octospora mit besonderer Berücksichtigung der Pfälzer Arten. Nova Hedwigia 34: 265–280.
- Itzerott, H. & P. Döbbler. 1982. Octospora meslinii und O. rubens (Pezizales), zwei weitere bryophile Gallbildner. Mitt. Bot. München 18: 201–212.
- Kornerup, A. & J.H. Wanscher. 1978. Methuen handbook of colour. Ed. 3. London.
- Moravec, J. 1969. Některé operkulátní diskomycety nalezené v okresech Mladá Boleslav a Jičín. Česká Mykol. 23: 222–235.

Moser, M. 1963. Ascomyceten. In: H. Gams, Kl. Kryptog.-Fl. Band IIa.
Seaver, F.J. 1928. The North American Cup-fungi (Operculates). New York.
Svrček, M. & J. Kubička. 1963. Druhý příspěvek k operkulátním discomycetům z okolí rybníka Dvo-fiště v jižních Čechách. Česká Mykol. 17: 61–70.
Velenovský, J. 1934. Monographia Discomycetum Bohemiae. Pragae.

#### PERSOONIA

Published by Rijksherbarium / Hortus Botanicus, Leiden Volume 15, Part 1, pp. 63-70 (1992)

#### SOME RAMULARIA-LIKE FUNGI ON MALVACEAE

#### G.S. DE HOOG

Centraalbureau voor Schimmelcultures, P.O. Box 273, 3740 AG Baarn, The Netherlands

The species Ramularia gossypii (Speg.) Cif., R. malvae Fuckel and Cladosporium anomalum Berk. & Curt. are redescribed. A new combination in Pseudocercospora is proposed for Cladosporium anomalum.

In the course of a screening of fungi related to *Hyalodendron fusiforme* Reddy & Bilgrami on *Gossypium* (de Hoog & Batenburg-van der Vegte, 1989), various Hyphomycetes growing on Malvaceae were studied, which are briefly discussed below.

#### ON GOSSYPIUM

### Ramularia gossypii (Speg.) Cif. - Fig. 1

Cercosporella gossypii Speg., An. Soc. cient. Argent. 22 (1886) 209. — Ramularia gossypii (Speg.) Cif., Atti Ist. bot. Lab. crittog. Univ. Pavia, Ser. 5, 19 (1962) 124.

Ramularia areola Atkinson, Bot. Gaz. 15 (1890) 166. — Symphiosira areola (Atkinson) Sawada, Spec. Publ. Coll. Agric., natn. Taiwan Univ. 8 (1959) 232.

Teleomorph: (?) Mycosphaerella areola Ehrlich & F.A. Wolf, Phytopathology 22 (1933) 229.

Leaf spots hypophyllous, whitish, angular, delimited by the leaf veins. Conidiophores emerging from pseudoparenchymatous tissue through the stomata in loose fascicles, (sub)hyaline, up to 70  $\mu$ m in length, sympodially proliferating, with 2–6 conspicuous scars. Stroma absent. Conidia arising in short chains, hyaline, thin-walled, punctate, fusiform, 1–2(–3)-septate, mostly 20–30 × 4  $\mu$ m.

Material examined. Atkinson Pl. Crypt. No. 1740 (type of R. areola) and Econ. Fungi No. 407, on Gossypium herbaceum leaves, Auburn, Ala., Sept. 1890 (NY); Fl. Alabama, various collections on Gossypium sp., Auburn, Ala., L.M. Underwood, Nov. 1890 (NY); ibid., F.S. Earle, Oct. 1895 and July 1896 (NY); on Gossypium sp., Starkville, Miss., S.M. Tracy, Oct. 1893 and 1895 (NY); on Gossypium sp., Puerto Rico, F.S. Earle, July 1903 (NY); Secc. Phyt. No. 522, on Gossypium hirsutum, Sergipe, Brazil, H. Borborema and H.P. Krug, Nov. 1934 (NY); on Gossypium hirsutum, Tupy, Brazil, A.S. Costa, March 1935; Bur. Pl. Indust. No. 60181, on Gossypium herbaceum, Tallassee, Ala., W.A. Orton, Sept. 1905 (NY); Herb. Agric. Exp. Stn, on Gossypium hirsutum, Gainesville, Fla., G.F. Weber, Oct. 1923 (NY).

Recent descriptions of this causal organism of grey mildew were given by Mulder & Holliday (1976), Holliday (1979) and Watkins (1984). Many authors used the name Ramularia areola Atk., but we follow Holliday (1979) and others in maintaining the older name R. gossypii for this taxon. Rathaiah (1973, 1976) described some cultural characteristics and host symptoms. The host specificity of the species was stressed by Dutta &

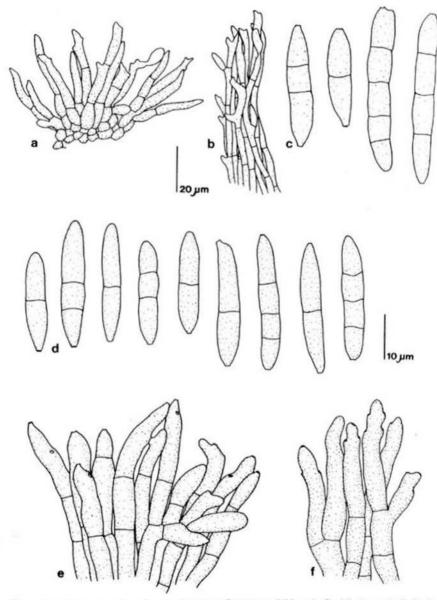


Fig. 1. Ramularia gossypti, various specimens on Gossypium (NY). a, b. Conidiophores (coll. Underwood); c, d. conidia (colls Earle and Atkinson); e, f. conidiophores (colls Atkinson and Earle).

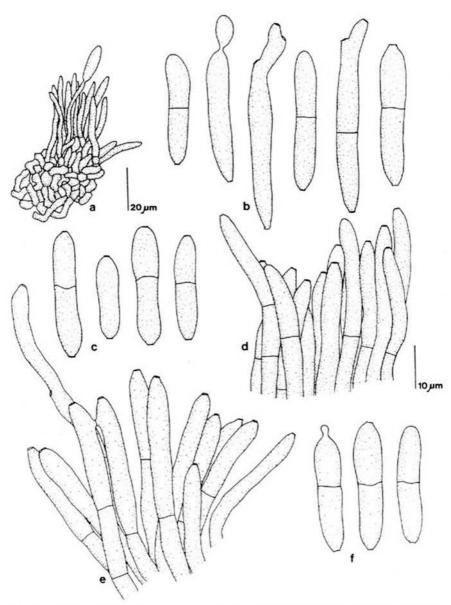


Fig. 2. Ramularia malvae, various specimens on Malva. a. Conidiophores (coll. Roivanen); b, c, f. conidia (colls Kabát and Bubák, Vestergren, Keith); d, e. conidiophores (colls Kabát and Bubák, Keith).

Jha (1979). Ehrlich & Wolf (1933) described a teleomorph in Mycosphaerella, but were unable to demonstrate the connection with Ramularia gossypii unambiguously.

Morphologically the species is closely similar to Ramularia sidae Olive (1948), described from leaves of Sida sp. (Malvaceae), but R. gossypii has slightly broader conidia.

Cercospora gossypina Cooke, the anamorph of Mycosphaerella gossypina (Cooke) Atk., was accepted in Cercospora by Hsieh & Goh (1990) on the basis of non-catenate conidia with dark scars. Cercospora gossypii Lall et al. (1962) and C. Ihuillieri Montégut (1967), both pathogenic on Gossypium leaves, are further species to be maintained in Cercospora s.str. because of their dark, prominent scars and cylindrical, multiseptate conidia. Whether or not these two species are identical remains to be established.

No authentic material was available at the herbarium KW of Cladosporium gossypii-cola Pidoplichko & Denyak and its variety minor Denyak (Pidoplichko, 1953). A secondary strain in the CBS collection, originating from Gossypium seed in Israel, CBS 320.87 (= ATCC 38026 = IMI 1266640) is indistinguishable from Cladosporium tenuissimum Cooke. No material of Cladosporium gossypii Jaczewski was available at B, BM, BR, C, CMI, E, K, KW, LE, MANCH, PAD, S or W. Cladosporium oligocarpum Corda var. malvacearum Berk. was not available at K. Because of the meagre descriptions, the four taxa are regarded to be of doubtful identity.

#### ON MALVA

## Ramularia malvae Fuckel - Fig. 2

Ramularia malvae Fuckel, Jb. nassau. Ver. Naturk. 23 (1870) 360.

Ramularia malvae Fuckel f. malvae-alceae Roum., F. sel. Gall. exsicc. (1890) No. 5085.

Ramularia malvae Fuckel var. malvae-moschatae Sacc., Syll. Fung. 4 (1886) 294. — Ramularia malvae-moschatae (Sacc.) Vestergren, Microm. rar. sel. Scand. (1902) No. 474.

Spots on living leaves hypophyllous, angular, delimited by the veins, c. 5 mm diam., pale, with regularly spaced, pale brown sporodochia. Stroma c. 80  $\mu m$  wide. Conidiophores loosely aggregated, emerging from pseudoparenchymatous subicula which develop in deteriorated leaf tissue, subhyaline, up to 100  $\mu m$  in length, mostly with a single, flat, dark apical scar. Conidia hyaline, cylindrical, 0–1-septate, mostly  $20-40\times4-6$   $\mu m$ , arising in short chains.

Material examined. Rouméguere, Fungi sel. exsicc. No. 5085, authentic for R. malvae f. malvae-alceae, on Malva alcea leaves, Forêt de Charny, France, F. Fautrey, Aug. 1890 (NY); Vestergren, Microm. rar. sel. Scand. No. 474, on Malva alcea leaf, Gotland, Sweden, T. Vestergren, 1895 and 1901 (K); on Malva moschata, Forres, Scotland, Rev. J. Keith (K); Kabát and Bubák, Fungi imperf. exsicc. No. 437, on leaves of Malva moschata, Wiborg, Jutland, Denmark, Sept. 1904 (K); on Malva moschata, Botanical Garden, Västerfjäll, Sweden, L. and H. Roivainen, June 1961 (NY).

In contrast to Ramularia gossypii, where the conidiophores protrude through the stomata, R. malvae causes small necrotic patches on otherwise healthy leaves, and develops with stromata in strongly damaged leaf tissue. The conidiophores have a single, conspicuous apical scar and the conidia are mostly broadly rounded at both ends, rather than acuminate as in R. gossypii.

The type of the R. malvae was not available at herb. B, and neither that of var. malvaemoschatae at herb. PAD. However, the species is sufficiently characteristic and typical for

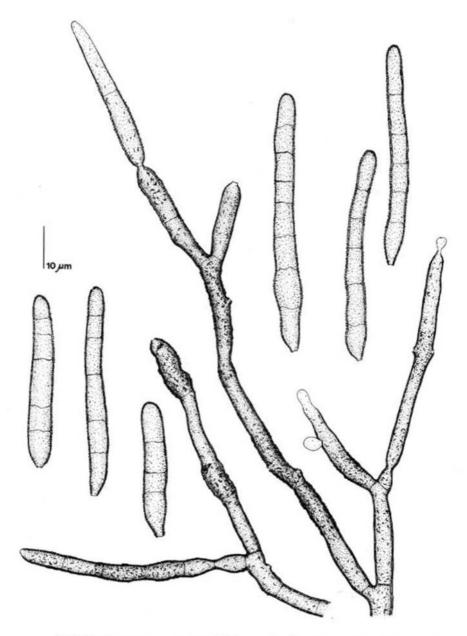


Fig. 3. Pseudocercospora anomala, conidiophores and conidia of type specimen (K).

Malva to be accepted as a valid taxon. Other Ramularia species described on Malvaceae, viz. R. malvastri Linder, R. sidae Olive and R. sidarum Petr. & Cif. all have superficial, repent hyphae with lateral conidiogenous extensions and are therefore currently classified in Mycovellosiella (Deighton, 1976; Braun, 1990).

### Pseudocercospora anomala (Berk. & Curt.) de Hoog, comb. nov. — Fig. 3

Cladosporium anomalum Berk. & Curt., Cub. Fungi No. 639; Sacc., Syll. Fung. 4 (1886) 363 (basionym) [non Ramularia anomala Peck, Bull. N.Y. St. Mus. 167 (1913) 47].

Spots on living leaves hypophyllous, irregular, vaguely delimited; colonies forming a brownish felt between abaxial hairs. Mycelium irregularly branched; conidiophores not or slightly differentiated, terminal or intercalary. Stroma absent. Conidiogenous cells terminal becoming intercalary, sympodial, bearing up to 3 conidia; fertile portion of conidiophore becoming darker and rough-walled; scars unpigmented. Conidia concolorous, with verrucose and rather firm walls, with 3–7 septa, up to 60 µm long, with unpigmented scars.

Material examined. Type of Cladosporium anomalum, on leaves of Malva sp., Cuba, C. Wright (K).

The species is characterized by diffusely branched hyphae with integrated, intercalary conidiogenous cells. During conidiogenesis, growth of the supporting cell seems to be arrested, thicker walls and encrusted extracellular pigment being produced. Subsequently the conidiophore proliferates again to give rise to a smooth-walled, less pigmented intermission.

No Pseudocercospora species have as yet been described on Malva (Hsieh & Goh, 1990; Guo & Liu, 1989). Pseudocercospora azanzae (Yadav) Deighton on Azanza is closely similar, but has smooth-walled conidia with more or less acute ends (Yadav, 1963). P. anomala is similar of Parastenella magnoliae (Weedon) David (Morgan-Jones, 1980; David, 1991). However, its cell walls are coarsely ornamented and conidia arise in small groups from somewhat protuding scars.

The Cercospora (s.l.) species on Malva mentioned by Chupp (1953), viz. C. malvarum Sacc., C. malvastri Mendoza, C. malvicola Ellis & Martin (= C. polymorpha Bubák) and C. sphaeralceicola (Speg.) Chupp, all have (sub)hyaline, acicular, often curved conidia and relatively dark conidiophores, and thus should be maintained in Cercospora s. str. Cercospora malvacearum Chiddarwar (1959, Bagyanarayana et al., 1991) has conidiophores with dark scars and very long, tapering, multiseptate conidia with truncate, dark bases and is therefore also maintained in Cercospora s. str.

Deighton (1976) preliminarily maintained a number of closely related *Pseudocercospora* species on *Hibiscus* as a complex around *P. abelmoschi* (Ell. & Ev.) Deighton, *P. anomala* differs from *P. abelmoschi* (= *Cercospora hibisci* Tracy & Earle; Kirk, 1980) in having procumbent or suberect hyphae, producing conidia from lateral extensions of intercalary cells. *Pseudocercospora hibisci-cannabini* (Sawada) Deighton has densely fasciculate conidiophores arising from stromata (Hsieh & Goh, 1990). *Pseudocercospora hibisci-mutabilis* (Sun) Yen differs mainly from *P. anomala* in having shorter conidiophores. *Pseudocercospora hibiscina* (Ell. & Ev.) Guo & Liu belongs to the same morphological group of species with loose mycelial wefts on abaxial leaf surfaces; its conidiophores are dark brown. *Cercospora abelmoschi-cannabini* Sawada, also causing leaf spots

on *Hibiscus*, is different in having fasciculate conidiophores and long, rostrate conidia (Prasad et al., 1960). *Pseudocercospora kydiae* Singh & Kamal (1986a) and *Stenella kydiae* Singh & Kamal (1986b) on leaves of *Kydia* both have relatively long, rostrate conidia.

### ACKNOWLEDGEMENTS

Dr. G. Morgan-Jones is thanked for kindly donating material of *Parastenella magnoliae* to the CBS Herbarium. The curators of NY and K are acknowledged for loaning herbarium specimens.

#### REFERENCES

- Bagyanarayana, G., P. Jagadeeswar & U. Braun. 1991. Miscellaneous notes on Indian Cercosporae. Mycotaxon 42: 319–326.
- Braun, U. 1990. Studies on Ramularia and allied genera (III). Nova Hedwigia 50: 499-521.
- Chiddarwar, P.P. 1959. Contributions to our knowledge of the Cercosporae of Bombay State. II. Indian Phytopathology 12: 111–121.
- Chupp, C. 1953. A monograph of the fungus genus Cercospora. Published by the author. Ithaca.
- Davis, J.C. 1991. Parastenella, a new generic name for Heterosporium magnoliae. Mycol. Res. 95: 123– 128.
- Deighton, F.C. 1976. Studies on Cercospora and allied genera. VI. Pseudocercospora Speg., Pantospora Cif. and Cercoseptoria Petr. Mycological Papers 140: 1–168.
- Dutta, A.K. & D.K. Jha. 1979. A note on the occurrence of areolate mildew of cotton in Bihar. Indian Phytopathology 32: 465–466.
- Ehrlich, J. & F.A. Wolf. 1933. Areolate mildew of cotton. Phytopathology 22: 229-240.
- Guo, Y.-I. & X.-j. Liu. 1989. Studies on the genus Pseudocercospora in China I, Mycosystema 2: 225–240.
- Holliday, P. 1979. Fungus diseases of tropical crops. Cambridge University Press, Cambridge.
- Hoog, G.S. de & W.H. Batenburg-van der Vegte. 1989. Retroconis, a new genus of ascomycetous Hyphomycetes. Studies in Mycology 31: 99-105.
- Hsieh, W.-H. & T.-K. Goh. 1990. Cercospora and similar fungi from Taiwan. Maw Chang Book Co., Taipei.
- Huguenin, B. 1966. Micromycètes du Pacifique Sud. (Troisième contribution). Dématiées de Nouvelle-Calédonie. Bulletin trimestrièl du Société mycologique de France 81: 686–698.
- Kirk, P. 1980. Pseudocercospora abelmoschi. CMI Descriptions of pathogenic Fungi and Bacteria 678.
- Lall, G., H.S. Gill & R.L. Munjal. 1962. Some Cercospora species from India. VI. Indian Phytopathology 14: 115–119.
- Montégut, J. 1967. Contribution à l'étude d'un complexe parasitaire s'attaquant à l'appareil aérien des cotonniers et entrainant son déssèchement. Rapport de mission à l'Office du Niger (Mali). Coton et Fibres tropicales 22: 439-453.
- Morgan-Jones, G. 1980. Notes on Hyphomycetes. XXXV. Stenellopsis gen. nov. Mycotaxon 10: 405–408
- Mulder, J.L. & P. Holliday. 1976. Ramularia gossypii. CMI Descriptions of pathogenic Fungi and Bacteria 520.
- Olive, L.S. 1948. Taxonomic notes on Louisiana fungi. I. Mycologia 40: 6-20.
- Pidoplichko, I.M. 1953. Gribnaya flora grubykh kormov. Akademia Nauk Ukrainskoy SSR, Kiev.
- Prasad, N., R.L. Mathur & J.P. Agnihotri. 1960. Cercospora abelmoschi-cannabini (Sawada) Prasad, Mathur and Agni, comb. nov. causing leaf spot disease of Ambari hemp (Hibiscus cannabinus Linn.) in Rajasthan, Science and Culture 25: 600–601.
- Rathaiah, Y. 1973. Étude de faux mildiou du cotonnier causé par Ramularia areola Atk. I. Croissance et sporulation du champignon en culture. Coton et Fibres tropicales 28: 287–292.

- Rathaiah, Y. 1976. Reaction of cotton species and cultivars to four isolates of Ramularia areola. Phyto-pathology 66: 1007–1009.
- Singh, A.K. & Kamal. 1986a. Fungi of Gorakhpur. XXXIV. Pseudocercospora. Kavaka 14: 25-29.
- Singh, A.K. & Kamal. 1986b. A new species of Stenella from India. Indian Phytopathology 39: 274–276.
- Watkins, G.M. (ed.). 1984. Compendium of cotton diseases. American Phytopathological Society, St. Paul.
- Yadav, A.S. 1963. Additions to the microfungi of Bihar. II. Cercosporeae. Indian Phytopathology 16: 167-170.

#### PERSOONIA

Published by Rijksherbarium / Hortus Botanicus, Leiden Volume 15, Part 1, pp. 71-92 (1992)

## CONTRIBUTIONS TOWARDS A MONOGRAPH OF PHOMA (COELOMYCETES) – I

1. Section Phoma: Taxa with very small conidia in vitro

J. DE GRUYTER1 & M.E. NOORDELOOS2

Eighteen taxa in section *Phoma* with conidia usually shorter than 5.5 µm are keyed out and described on account of their characteristics in vitro. Two taxa from section *Sclerophomella* are added because they have very similar characters in vitro. The following new binomials have been proposed: *Phoma dictamnicola* Boerema, de Gruyter & Noordel., nom. nov., *Phoma dorenboschii* Noordel. & de Gruyter, spec. nov., *Phoma minutispora* P.N. Mathur, nom. nov., and *Phoma opunticola* Boerema, de Gruyter & Noordel., spec. nov. Hostfungus and fungus-host indices are provided and short comments on the ecology and distribution of the taxa are given.

During the past 30 years Boerema and co-workers at the Plant Protection Service at Wageningen, the Netherlands, have been working on the taxonomy of *Phoma* in pure culture, in particular with respect to those species that are plant-inhabiting. In the course of this research several hundreds of taxa have been studied, resulting in a fairly large number of publications and a well-documented system of not yet published data. In the course of this project the characters obtained by studying living material in vitro is found to be indispensable for the differentiation and delimitation of taxa. The main reason is that many species of *Phoma* are plurivorous and in vivo most variable qua morphology of pycnidia and conidia. On account of the characteristics in vitro and in vivo the genus has been subdivided in a number of sections, see e.g. Van der Aa et al. (1990). The section *Phoma* is characterized by thin-walled glabrous ostiolate pycnidia producing in vitro and in vivo only aseptate conidia.

Boerema, de Gruyter and Noordeloos are planning a comprehensive publication on all the *Phoma* taxa concerned, with a classification, keys, descriptions, host-pathogen indices and notes (Boerema, de Gruyter & Noordeloos, in prep.). This paper is the first in a series of precursors of this big project, that has to be completed in the course of the forth-coming years.

The present paper gives a key and the diagnostic features of a selection of 20 *Phoma* species that have relatively small conidia in vitro, with a length usually not exceeding 5.5 µm. Most of them belong to section *Phoma*, but two species from another section are included because of comparable small conidia in vitro. The authors have tried as much as possible to interprete old names for the taxa concerned. In four cases new binomials had to be created. Indices on the fungus relations are added and short comments on the ecology and distribution of the taxa are also given.

<sup>1)</sup> Plant Protection Service, P.O. Box 9102, NL-6700 HC Wageningen, The Netherlands.

Rijksherbarium / Hortus Botanicus, P.O. Box 9514, NL-2300 RA Leiden, The Netherlands.

#### MATERIAL AND METHODS

All isolates studied were present in the collection of the Plant Protection Service as freeze-dried cultures. These were brought in culture again and transferred to oat-meal agar in petri-dishes. For the colony descriptions 5 mm mycelium plugs, taken from the edge of the active growing cultures, were transferred on oat-meal agar (OA), malt agar (MA), and cherry-decoction agar (CA) and placed in an incubator in complete darkness, at 22°C. After 7 days the diameter of the colonies was measured, and the morphology of the colonies, aerial mycelium and other structures were studied. The colours of aerial mycelium, colonies and reverse were described according to the colour-code of Rayner (1970). After 7 days the petri-dishes were placed in an incubator with a day-night regime of 13 hours NUV light and 11 hours darkness to stimulate the pigmentation of the colonies and the formation of pycnidia. Two weeks after incubation the colonies were described again, and the morphology of pycnidia, conidiogenous cells and conidia were studied from the OA cultures. The NaOH spot-test was done on MA by addition of a drop of 1N NaOH on the colony margin, and the colour-change was noted. Also the colour of the conidial slimemass excreted by mature pycnidia was noted. Chlamydospores, if present, were studied on OA and MA. Drawings were made with help of a drawing tube. Conidia dimensions refer to 30 measurements with oil-immersion at × 1250. Q stands for the length/width ratio of the conidia.

#### KEY TO THE PHOMA SPECIES TREATED IN THIS PAPER

1a.	Conidia very small, length in average not exceeding 3 $\mu m$
b.	Conidia in average between 3 and 5 µm long
2a.	Conidia subglobose to broadly ellipsoid, average Q < 1.5
	Conidia ellipsoid to subcylindrical, average $Q = 2-3$
	Conidia $2.0-2.8 \times 1.6-2.0 \mu\text{m}$ , subglobose to broadly ellipsoid, Q = 1.1-1.6, with
	one large guttule; colonies on MA with distinct pinkish-reddish or apricot colours;
	chlamydospores present; growth-rate slow, 20-25 mm 1. Phoma minutispora
b.	Conidia $2.4-3.2(-5.5) \times 1.8-2.4(-3.0) \mu m$ , Q = 1.3-1.8, broadly ellipsoid with
	one to three acentric guttules; colonies on MA with grey-olivaceous tinges; chlamydo-
	spores absent; growth-rate moderate to fast, 45-70 mm 2. Phoma anserina
4a.	Colonies growing relatively fast, growth-rate at least 30-40 mm; colonies distinctly
	pigmented with olivaceous-grey and citrine tinges; conidia usually eguttulate; patho-
	genic to Opuntia spp
b.	Colonies slow-growing, growth-rate about 20 mm, more or less unpigmented, but
	exudating a yellow pigment that diffundates into the agar; conidia sometimes with one
	or two, small, polar guttules
5a.	Conidia oblong in average Q < 2
b.	Conidia with average Q > 2
6a.	Growth-rate at least 35–50 mm
b.	Growth-rate slow, about 20 mm
7a.	Colonies not pigmented or with yellowish, pink or salmon colours, olivaceous
	tinges, if present, never dominant
b.	Colonies greenish-olivaceous or olivaceous-grey to olivaceous-black 9

8a.	Colonies colourless, sometimes with faint olivaceous-grey tinges and/or sectors,
	producing a yellow pigment that stains the agar, reverse honey to umber
	5. Phoma putaminum
b	Colonies peach to salmon, reverse similar, not producing a yellow pigment
0.	6. Phoma capitulum
9a	Colonies greenish-olivaceous; conidia sometimes with small guttules; pathogenic to
	Coffea arabica 7. Phoma costarricensis
h	Colonies distinctly dark olivaceous-grey to olivaceous-black; conidia with two,
	distinct, polar guttules; on dead stems of Valeriana spp 8. Phoma valerianae
10a	Colonies with distinct apricot or scarlet tinges on OA; NaOH spot-test positive
.ou.	9. Phoma multipora
b	Colonies grey-olivaceous, greenish-olivaceous or medium yellow on OA; NaOH
٠.	spot-test negative
11a	Conidia cylindrical to bacilliform, average Q > 3; growth-rate slow, up to 20 mm,
	colonies on OA chestnut to ochraceous with only weak olivaceous tinges; pycnidia
	thick-walled, consisting of about 4–8 layers of cells; conidia $3.0-3.6 \times 0.8-1.0$
	μm, average 3.2 × 0.9 μm; pathogenic to Olea europaea 19. Phoma incompta
b.	Conidia oblong to subcylindrical, average Q between 2 and 3
	Growth-rate about 20 mm; pathogenic to Apium graveolens 11. Phoma apiicola
	Growth-rate at least 40-50 mm
13a.	'Ice-fern' crystals are readily formed within two weeks on MA; conidia 3.0-5.5 ×
	$1.5-2.0(-2.5)$ µm, in average $4.2-4.4 \times 1.7-1.8$ µm, Q = $1.5-3.5$
	12. Phoma dorenboschii
b.	No 'Ice-fern' crystals formed
14a.	Chlamydospores absent
b.	Chlamydospores present
15a.	Pycnidia with short, but distinct neck, elongated in a later stage 16
b.	Pycnidia papillate, usually with a neck, not elongated in a later stage 17
16a.	Colonies with distinct bright yellow-green tinges (citrine); conidia $3.2-4.2 \times 1.6-$
	2.0 μm, Q = 1.8-2.6; pathogenic to Anigozanthus spp 3. Phoma anigozanthi
b.	Colonies with olivaceous-grey to olivaceous-black colour, conidia 3.6-5.6 × 1.6-
	2.2 μm, Q = 2.0-3.0
17a.	Colonies olivaceous-grey, olivaceous, or olivaceous-black with citrine tinges; growth-
	rate 40-50 mm; conidia usuallly eguttulate; pathogenic to Eucalyptus spp. and Eu-
	genia spp
b.	Colonies colourless to greenish-olivaceous or dull green; pycnidia with distinct osti-
	oles; more or less papillate
18a.	Conidia $3.0-5.9 \times 1.2-2.1$ µm, in average $3.7-4.4 \times 1.6-2.0$ µm, Q = $1.8-2.6$ ;
	NaOH spot-test negetive; saprophytic on many, mainly tropical plants
	16. Phoma tropica
b.	Conidia $4.0-6.0\times2.0-2.8~\mu m$ , in average $4.8\times2.3~\mu m$ , $Q=1.7-2.3$ ; NaOH spot-
	test positive; pathogenic to Hedera spp 17. Phoma hedericola
19a.	Pycnidia relatively thin-walled, consisting of about 3 layers of cells
	18. Phoma eupyrena
b.	Pycnidia thick-walled, consisting of about 7-8 layers of cells; pathogenic to Dictam-
	nus albus

#### HOST-FUNGUS INDEX

Ubiquitous species on various herbaceous and/or woody plants: Phoma anserina; P. capitulum; P. eupyrena; P. fimeti; P. minutispora; P. putaminum; P. tropica; P. viburnicola.

Isolated from soil: P. anserina; P. capitulum; P. eupyrena; P. fimeti; P. minutispora; P. multipora; P. putaminum.

Isolated from water: P. eucalyptica; P. flavigena.

# Isolated from specific plants:

Apium graveolens (Umbelliferae) P. apiica Callistephus (Compositae) P. doren Coffea arabica (Rubiaceae) P. costau Dictamnus albus (Rutaceae) P. dictan	zanthi
Coffea arabica (Rubiaceae) P. costar Dictamnus albus (Rutaceae) P. dictam	la
Dictamnus albus (Rutaceae) P. dictam	boschii
the second secon	ricensis
	nnicola
Eucalyptus spp. and Eugenia spp. (Myrtaceae) P. eucaly	ptica
Hedera spp. (Araliaceae) P. heder	icola
Olea europaea (Oleaceae) P. incom	pta
Opuntia spp. (Cactaceae) P. opunt	icola
Physostegia virginiana (Labiatae) P. doren	boschii
Valeriana spp. (Valerianaceae) P. valeri	anae

#### FUNGUS-HOST INDEX

P. anigozanthi	Anigozanthus spp.(Amaryllidaceae)
P. apiicola	Apium graveolens (Umbelliferae)
P. costarricensis	Coffea arabica (Rubiaceae)
P. dictamnicola	Dictamnus albus (Rutaceae)
P. dorenboschii	Physostegia virginiana (Labiatae);
	Callistephus (Compositae)
P. eucalyptica	Eucalyptus spp.and Eugenia spp. (Myrtaceae)
P. hedericola	Hedera spp. (Araliaceae)
P. incompta	Olea europea (Oleaceae)
P. opunticola	Opuntia spp. (Cactaceae)
P. valerianae	Valeriana spp. (Valerianaceae)

#### DESCRIPTIVE PART

#### Section Phoma

# 1. Phoma minutispora P.N. Mathur, nom. nov. - Fig. 1

Phoma oryzae Cooke & Massee, Grevillea 16 (1887) 15; not Phoma oryzae Cattanea, Arch. Bot. crittog. Pavia 2-3 (1877) 118. — Phyllosticta oryzae (Cooke & Massee) Miyake, J. Coll. Agric. imp. Univ. Tokyo 2 (4) (1910) 252.

Selected literature: Padwick (1950), Shukla et al. (1984).

Description in vitro

OA: growth-rate 19–21 mm (14 days: 33–35 mm); colony regular or somewhat irregular in shape, predominantly grey with olivaceous tinges (grey-olivaceous to olivaceousgrey) with abundant, grey, floccose aerial mycelium; reverse olivaceous-black, towards margin grey-olivaceous to vinaceous-buff, centre vinaceous-buff (rarely colony salmon with poorly developed aerial mycelium, reverse salmon).

MA: growth-rate 25-26 mm (14 days: 46-49 mm); colony slightly irregular with undulating margin, pinkish-red (flesh to peach) with darker zone near margin, with thin, felted-velvety aerial mycelium; reverse apricot with darker sectors (greyish-sepia), after three weeks the general colour is more towards grey-olivaceous with bright apricot spots and sectors.

CA: growth-rate 24–25 mm (14 days: 43 mm); colony regular or irregular in shape with undulating margin, olivaceous-grey with woolly aerial mycelium; reverse very dark olivaceous-black at centre, towards margin buff (or colony salmon with olivaceous-grey tinges towards margin, with finely floccose aerial mycelium; reverse salmon-peach with olivaceous-black centre).

Pycnidia  $100-250\times80-200~\mu m$ , globose with one or up to 4, sometimes indistinct ostioles, without neck; olivaceous-black, abundantly formed within three weeks at centre and in concentric zones, both on and in the agar and in aerial mycelium, often associated with dense hyphal strands in the mycelium; exudate salmon. Conidiogenous cells  $2-6\times3-5~\mu m$ , more or less globose or bottle-shaped. Conidia  $2.0-2.8\times1.6-2.0~\mu m$ , average  $2.4\times1.8~\mu m$ , Q=1.1-1.6, average Q=1.4, broadly ellipsoid with one large guttule.

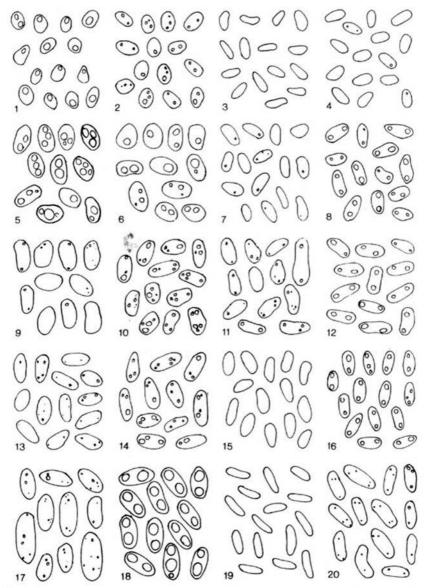
Chlamydospores few, about 6-15 µm in diameter, solitary, thick-walled, and usually also finely warty, generally formed at tip of hyphae, present after 4 weeks on OA.

NaOH spot-test: negative.

Ecology and distribution. Probably a common saprophytic soil fungus in south-west Asia, characterized by its extremely small conidia. Isolates made in India refer e.g. to dead leaf sheats of rice, Oryza sativa, bark of Ficus sp. and soil. The fungus is also reported as an opportunistic human pathogen.

Culture studied. CBS 509.91 (PD 77/920) ex saline soil, India; CBS 711.76 (PD 91/1445) ex bark of Ficus sp. (Moraceae), India.

Note. This avowed substitute for *Phoma oryzae* Cooke & Massee has been adopted from a study on an Indian soil-isolate of the fungus by P.N. Mathur (Thesis Univ. Agra, 1967).



Figs. 1–20. Conidia (x 1250). 1. Phoma minutispora; 2. P. anserina; 3. P. opunticola; 4. P. flavigena; 5. P. putaminum; 6. P. capitulum; 7. P. costarricensis; 8. P. valerianae; 9. P. multipora; 10. P. fimeti; 11. P. apiicola; 12. P. dorenboschii; 13. P. anigozanthi; 14. P. viburnicola; 15. P. eucalyptica; 16. P. tropica; 17. P. hedericola; 18. P. eupyrena; 19. P. incompta; 20. P. dictamnicola.

## 2. Phoma anserina El. Marchal. - Fig. 2

Phoma anserina El. Marchal, Champ. copr. (1891) 11.

Phoma marchali Sacc., Sylloge Fung. 10 (1892) 188.

Aposphaeria humicola Oud., Ned. kruidk. Archf III, 2 (3) (1902) 721.

Phoma radicis-callunae Rayner, Bot. Gaz. 73 (1922) 231.

Phoma suecica van Beyma, Antonie van Leeuwenhoek 8 (1942) 110-111.

Selected literature: Boerema (1985).

## Description in vitro

OA: growth-rate 49-53 mm, regular, without aerial mycelium; colony colour pale to dark olivaceous, grey-olivaceous or olivaceous-black; reverse grey-olivaceous to olivaceous-black, but with distinct lead-grey tinge.

MA: growth-rate 57-73 mm, regular, with floccose pale olivaceous-grey aerial mycelium; colony colour greenish-olivaceous to grey-olivaceous or olivaceous-black; reverse grey-olivaceous, olivaceous black.

CA: growth-rate 61–73 mm, regular, without aerial mycelium or with finely floccose-woolly olivaceous-grey aerial mycelium; colony colour ranging from pale olivaceous-grey to grey-olivaceous or olivaceous-black; reverse grey-olivaceous to olivaceous-black.

Pycnidia  $112-136\times112-176~\mu m$ , solitary, globose with 1, rarely 2 or 3 small ostioles, without or sometimes with a short neck; olivaceous to olivaceous-black, smooth, glabrous or with hyphal outgrowths ('semi-pilose'), abundant, scattered, mostly on, but sometimes partly in or entirely in the agar; exudate white to rosy-buff. Conidiogenous cells  $4-8\times3-7~\mu m$ , phialidic, globose to bottle-shaped. Conidia  $2.4-3.2(-5.5)\times1.8-2.4(-3.0)~\mu m$ , average  $2.8\times1.9~\mu m$ , Q=1.3-1.8, average Q=1.5, broadly ellipsoid with 1 or 2 (3), acentric guttules.

Chlamydospores absent, but swollen elements may occur in some strains.

NaOH spot-test: negative.

Ecology and distribution. This is an 'omnivorous' soil fungus in temperate Eurasia and North America, which is also recorded from northern Africa. Commonly found on roots of herbaceous and woody plants and cysts of nematodes. Also frequently isolated from seeds and fruits.

Culture studied. CBS 364.91 (PD 81/290) ex fruit of Ananas sativus (Bromeliaceae) with fruit-rot; origin unknown; CBS 365.91 (PD 84/108) ex seed of Cucumis sativus (Cucurbitaceae), the Netherlands; CBS 363.91 (PD 79/712) ex seed of Pisum sativum (Papilionaceae), the Netherlands.

Note. Culture CBS 364.91 (PD 81/290) is distinctly less intensely pigmented than the other two cultures studied. Also the size of the conidia varies considerably from one strain to the other; the length which usually ranges from  $2.4-3.2~\mu m$  may reach up to  $5.5~\mu m$ .

# 3. Phoma opunticola Boerema, de Gruyter & Noordel., spec. nov. - Fig. 3

Coloniae in agaro farinae avenae tarde crescentes (circa 30 mm), olivaceo-griseae vel citrinae sectoribus pallidis, reverso simili; pycnidia  $40-150~\mu m$  in diam., globosa vel compressa, collo longo praedito, atra; conidia hyalina, semplicia, plerumque  $2.4-3.6\times1.0-1.4~\mu m$ , eguttulata. Parasitica in cladodiis vivis Opuntiis. Holotypus: L 989.300-160 (siccus); CBS 376.91 (PD 77/1177) (vivus).

Description in vitro

OA: growth-rate 32 mm (14 days: 63 mm), regular, without aerial mycelium; colony colour olivaceous-grey to olivaceous-greenish or citrine with paler sectors, pycnidia in concentric zones, reverse similar.

MA: growth-rate 39-40 mm (14 days: 70 mm), regular, with felted, white aerial mycelium; colony greenish-olivaceous; reverse similar.

CA: growth-rate 36 mm (14 days: 75 mm), regular, with finely velvety, olivaceousgrey aerial mycelium; colony colour olivaceous to greenish-olivaceous, with distinct radially orientated hyphal strands; reverse similar.

Pycnidia 40–150 µm, globose or somewhat compressed, with long neck somewhat bottle-shaped, black, glabrous to somewhat hairy with mycelium; with white to pale olivaceous-grey exudate; abundant, mostly in concentric rings, and associated with the radiating hyphal strands, on and partly also in the agar. Conidiogenous cells  $2.5-6\times2-4$  µm in diameter, phialidic, broadly globose, thin-walled. Conidia  $2.4-3.6\times1.0-1.4$  µm, in average  $2.8\times1.2$  µm, Q=1.7-3.6, average Q=2.4, ellipsoid to subcylindrical, eguttulate.

NaOH spot-test: not a fairly strong reaction, but usually a slight sienna discolouration is observed, that slowly turns bluish-green.

Ecology and distribution. A pathogen of Opuntia spp. causing necrotic spots on the leaf-like stems (cladodes): 'Necrotic Spot'. The fungus is indigenous to South America, but probably also occurs in other areas where the host is cultivated. It easily may be confused with the Asteromella-spermatial state of Mycosphaerella opuntiae (Ell. & Ev.) Dearness, commonly known as Phyllosticta concava Seaver.

Culture studied. CBS 376.91 (PD 77/1177) ex Opuntia sp. (Cactaceae), Peru.

# 4. Phoma flavigena O. Const. & v.d. Aa - Fig. 4

Phoma flavigena O. Const. & v.d. Aa, Trans. Br. mycol. Soc. 79 (1982) 343.

Description in vitro (adopted from Constantinescu & v.d. Aa, 1982)

OA: growth-rate 18-23 mm, regular, with whitish, sparse aerial mycelium; colony colourless but the agar is strongly discoloured due to the release of a yellow pigment.

MA and CA: growth-rate 11-14 mm, regular, with abundant, greyish or yellowish aerial mycelium; colony colourless but the agar is strongly discoloured due to the release of a yellow pigment.

Pycnidia  $50-210~\mu m$  in diameter, solitary or confluent, globose, with a conspicuous neck, with distinct ostiole, brownish, abundant in concentric rings, immersed in the agar or in aerial mycelium; exudate buff or rosy-buff. Conidiogenous cells  $4-7\times3-6~\mu m$ , phialidic, truncate-conical or flask-shaped. Conidia  $2-4\times1-2~\mu m$ , Q=1.8-2.7, average Q=2.3, ellipsoid to cylindrical, sometimes with one or two guttules.

NaOH spot-test: negative.

Ecology and distribution. Once isolated from fresh water, Romania. Culture studied. CBS 314.80 (PD 91/1613).

## 5. Phoma putaminum Speg. - Fig. 5

Phoma putaminum Spegazzini, Atti Sov. crittogam. ital. 3 (1881) 66. — Aposphaeria putaminum (Speg.) Sacc., Sylloge fung. 3 (1884) 177. — Coniothyrium putaminum (Speg.) O. Kuntze, Revisio Gen. Pl. 3 (2) (1898) 459.

Phoma radicicola McAlpine, Fung. Dis. Stone-fruit Trees Melb. (1902) 126.

Phoma dunorum ten Houten, Kiemplziekt. Conif. [Thesis Univ. Utrecht] (1939) 88-89.

Selected literature. Boerema & Dorenbosch (1973).

## Description in vitro

OA: growth-rate 43-59 mm, regular, without aerial mycelium; colony colourless but exudating a pigment that stains the agar honey; reverse honey with greenish-olivaceous tinge.

MA: growth-rate 51-63 mm, regular, with sparse aerial mycelium of more or less erect hairs, that becomes floccose after 14 days; colony colour between ochraceous and umber with slight olivaceous tinge; reverse umber with honey margin.

CA: growth-rate 39-47 mm, somewhat irregular, with cottony, grey-olivaceous aerial mycelium; colony colour umber, towards margin hazel; reverse similar.

Pycnidia  $60-300~\mu m$ , mostly single but also confluent, globose, olivaceous to olivaceous-black, covered in mycelial hairs, especially in young pycnidia, with distinct ostiole, without or with a neck; scattered, especially towards margin of colony, and also in concentric zones, mostly partly in the agar, sometimes also entirely in the agar; exudate whitish-salmon. Conidiogenous cells  $3-7\times 3-7~\mu m$ , phialidic, broadly globose to slightly elongated, thin-walled. Conidia  $3.2-4.2\times 2.0-2.6$ , average  $3.8\times 2.4$ , Q=1.3-1.9, average Q=1.6, broadly ellipsoid, mostly with 2 or 3 greenish guttules.

NaOH spot-test: negative.

Ecology and distribution. A soil-borne fungus, isolated from the subterranean parts of various herbaceous and woody plants. Generally it is regarded as a saprophyte, but it may act as an opportunistic parasite on roots etc. Most isolates originate from Europe and North America, but the fungus has also been recorded from the southern hemisphere.

Culture studied. CBS 372.91 (PD 75/690) ex Ulmus sp. (Ulmaceae), the Netherlands; CBS 373.91 (PD 83/119) ex Buxus sp. (Buxaceae), West Virginia, U.S.A.

# 6. Phoma capitulum Pawar, Mathur & Thirumalachar - Fig. 6

Phoma capitulum Pawar, Mathur & Thirumalachar, Trans. Br. mycol. Soc. 50 (1967) 261.

Phoma ostiolata Pawar, Mathur & Thirumalachar, Trans. Br. mycol. Soc. 50 (1967) 262, var. ostiolata.

Phoma ostiolata var. brunnea Pawar, Mathur & Thirumalachar, Trans. Br. mycol. Soc. 50 (1967) 263.

Selected literature. Boerema (1985).

# Description in vitro

OA: growth-rate 37 mm (14 days: 66-71 mm), regular with fine velvety-floccose, white aerial mycelium; colony colour salmon to flesh, with grey concentric pycnidial zones; reverse similar.

MA: growth rate 38-40 mm (14 days: 75 mm); regular, with fine peluchy to floccose, white to pale olivaceous-grey aerial mycelium; colony colour peach to salmon with pale olivaceous-grey tinges zone full of pycnidia; reverse between salmon and apricot.

CA: growth-rate 27-28 mm (14 days: 55-58 mm), irregular, with peluchy, white aerial mycelium; colony colour between salmon and flesh with greyish concentric pycnidial zones; reverse similar.

Pycnidia  $50-105\times50-80~\mu m$ , usually in clusters of up to 20 specimens in a row, born on radiating, dense, blackish hyphal strands; globose with 1-3 ostioles on a short neck; citrine to honey when young then olivaceous to olivaceous-black, smooth, glabrous, with thin walls; readily developing in concentric zones or sectors after 7-14 days, mostly on the agar, but also partly or entirely in the agar; exudate grey-saffron. Conidiogenous cells  $3-7\times4-7~\mu m$ , phialidic, globose, thin-walled. Conidia  $3.2-4.4\times2.0-3.0~\mu m$ , average  $3.8\times2.6~\mu m$ , Q=1.2-2.0, average Q=1.5, broadly and shortly ellipsoid with one or two guttules.

NaOH spot-test: negative.

Ecology and distribution. Isolated from saline soil, marine environment and oakforest soil in India. It probably represents a common, 'halophilic' soil inhabiting fungus in south-west Asia.

Culture studied. CBS 337.65 (PD 91/1614; IMI 113 693; ATCC 16195; HACC 167) ex saline soil, India.

## 7. Phoma costarricensis Echandi — Fig. 7

Phoma costarricensis Echandi, Rev. Biol. Trop. 5 (1957) 83.

Phyllosticta coffeicola sensu Stevens, Ill. biol. Monographs 11 (2) )1927) 52-53; not Phyllosticta coffeicola Speg., Rev. Fac. Agron. Veter. La Plata (1896) 345 [= Phomopsis sp.].

## Description in vitro

OA: growth-rate 43 mm, regular, with scattered tufts of whitish aerial mycelium in marginal zone only; colony colour greenish-olivaceous; reverse grey-olivaceous.

MA: growth-rate 52 mm, regular, with floccose, olivaceous aerial mycelium; colony colour greenish-olivaceous, reverse olivaceous-black.

CA: growth-rate 57 mm, with poorly developed, more or less felted, adpressed, greyolivaceous aerial mycelium; colony colour grey-olivaceous, zonated; reverse similar.

Pycnidia  $50-200\times50-140~\mu m$ , globose to bottle-shaped, usually with one ostiole, glabrous, smooth, brownish-olivaceous to olivaceous-black, abundant on and partly in the agar; exudate whitish. Conidiogenous cells  $3-6\times3-5~\mu m$ , phialidic, globose to bottle-shaped, thin-walled. Conidia  $(1.4-)2.8-4.0\times(1.0-)1.6-1.8~\mu m$ , average  $2.6-3.3\times1.3-1.6~\mu m$ , Q = 1.3-3.0, in average 2.0, eguttulated, or sometimes with small guttules.

NaOH spot-test: not a specific reaction, but a slight discolouration to sienna may occur.

Ecology and distribution. A pathogen of Coffea arabica causing lesions on leaves, stems and fruits: 'Coffee Blight'. The fungus was until recently only known from South and Central America, but is now also recorded in other parts of the world.

Cultures studied. CBS 506.91 (IMI 215 229; PD 91/876) ex twig of Coffea sp. (Rubiaceae), Nicaragua.

Note. In vivo the conidia generally are larger, mostly  $5-6 \times 2-3 \, \mu m$ .

## 8. Phoma valerianae P. Henn. - Fig. 8

Phoma valerianae P. Hennings, Nyt. Mag. Naturvid. 42 (1904) 29.

Phyllosticta valerianae-tripteris f. minor Unamuno, Mems R. Soc. esp. Hist. nat. 15 (1929) 348-349.

### Description in vitro

OA: growth-rate 65 mm, colony rather regular, greenish-grey with greenish-grey, flat, finely woolly aerial mycelium; reverse zonated with alternating grey-olivaceous and olivaceous-black zones, margin more like greenish-grey.

MA: growth-rate 36-37 mm, colony regular, dark grey-olivaceous, olivaceous-grey or greenish-grey, with very fine, compact, velvety, greenish-grey or olivaceous-grey aerial mycelium; reverse iron grey with pallid olivaceous-buff centre and marginal zone.

CA: growth-rate 48-50 mm, colony rather regular, smoke-grey to olivaceous-grey, darker grey-olivaceous towards margin, entirely densely velvety with whitish-grey aerial mycelium; reverse olivaceous-grey at centre with darker zones towards margin, marginal zone paler towards greenish-olivaceous.

Pycnidia  $120-340~\mu m$ , single rarely confluent, globose with distinct neck, olivaceous to olivaceous-black, glabrous, abundant in and on the agar; exudate amber. Conidiogenous cells  $3-6\times4-6~\mu m$  in diameter, phialidic, globose, thin-walled. Conidia  $2.5-3.9\times1.4-2.1$ , average  $3.2\times1.8$ , Q=1.4-2.1, average 1.8, ellipsoid with two polar guttules. NaOH spot-test: negative.

Ecology and distribution. In Europe frequently occurring on dead stems of Valeriana spp., and apparently seed-borne. Incidentally also isolated from other not related herbaceous plants.

Culture studied. CBS 499.91 (PD 73.672) ex Valeriana officinalis (Valerianaceae), basal stemrot, the Netherlands.

# 9. Phoma multipora Pawar, Mathur & Thirumalachar - Figs. 9, 21

Phoma multipora Pawar, Mathur & Thirumalachar, Trans. Br. mycol. Soc. 50 (1967) 260-261.

#### Description in vitro

OA: growth-rate 20-23 mm (14 days: 39-48 mm), regular with weakly undulating outline, with only poorly developed, more or less felted, white to olivaceous-grey aerial mycelium; colony colour apricot to scarlet; reverse similar.

MA: growth-rate 18-22 mm (14 days: 34-43 mm), regular to irregular, with compact to fluffy, grey-olivaceous to olivaceous-grey aerial mycelium; colony colour grey-olivaceous at centre, towards margin olivaceous-grey with peach outer margin; reverse rust or sepia with apricot to cinnamon outer margin.

CA: growth-rate 20-22 mm (14 days: 42-46 mm), regular or slightly irregular in outline, with scanty, velvety, grey-olivaceous aerial mycelium; colony colour grey-olivaceous to olivaceous-grey, margin vinaceous; reverse dark vinaceous to dark brick or sepia.

Pycnidia  $120-300 \mu m$ , solitary, or confluent in groups of 2-5, globose, occasionally 'multi'-ostiolate, but usually with one or two wide ostioles without a distinct neck; citrine, then darker and more like olivaceous, glabrous or with very short swollen hyphal appendages around the ostiole ('semi pilose'), scattered in and on the agar; exudate rosy-buff or

vinaceous. Conidiogenous cells globose to bottle-shaped,  $3-6\times4-8~\mu m$ , phialidic. Conidia  $3.6-4.8\times1.6-2.4~\mu m$ , average  $4.3\times2.2~\mu m$ , Q=1.7-2.6, average Q=2.0, broadly ellipsoid with or without a few guttules.

Chlamydospores absent, but in the mycelium swollen cells, that can be pigmented, may be formed.

NaOH spot-test: quickly changing to purplish-blue.

Ecology and distribution. This fungus is recorded from the subtropical regions of south-east Asia (India) and northern Africa (Egypt), and it probably is a halophilic soil-borne saprophyte. The records from India refer to soil in mangrove vegetations near the Bombay-coast and dead herbaceous stems near Lahore.

Culture studied. CBS 501.91 (PD 83/888), substrate unknown, Egypt; CBS 353.65 (PD 91/1560) ex saline soil, India.

Note. CBS 353.65 forms small orange-yellow crystals on OA.

# 10. Phoma fimeti Brun. - Fig. 10

Phoma fimeti Brunaud, Bull. Soc. bot. Fr. 36 (1889) 338. Selected literature. Dorenbosch (1970), Boerema & Dorenbosch (1973).

Description in vitro

OA: growth-rate 20-21 mm (14 days: 39-45 mm), regular, with velvety pale olivaceous-grey aerial mycelium; colony colour grey-olivaceous to greenish-olivaceous with a yellow pigment diffundating into the medium, with concentric pycnidial zones, reverse greenish-olivaceous to olivaceous-grey.

MA: growth-rate 18-23 mm (14 days: 33-40 mm), with floccose, olivaceous-grey aerial mycelium; colony colour olivaceous-grey with citrine margin; reverse olivaceous-grey to olivaceous-black.

CA: growth-rate 20–23 mm (14 days: 29–40 mm), with velvety, pale olivaceousgrey aerial mycelium; colony colour olivaceous-grey to grey-olivaceous with regular pycnidial zones, margin sometimes distinctly paler more like citrine-honey; reverse olivaceous-black, sometimes with citrine-honey margin, sometimes with leaden-grey tinges in marginal zone.

Pycnidia  $65-200\times60-200~\mu m$ , solitary or confluent, globose, usually with one ostiole on short neck; olivaceous-black with white to ochraceous exudate, glabrous, smooth, abundant, in concentric zones, on the agar, sometimes entirely in the agar, or in aerial mycelium. Conidiogenous cells  $2-7\times2-8~\mu m$ , phialidic, broadly globose, thin-walled. Conidia  $2.8-5.1\times1.7-3.2~\mu m$ , average  $3.8-4.1\times2.2-2.7~\mu m$ , Q=1.0-2.4, average Q=1.4-1.8, broadly ellipsoid with two or more, polar guttules.

NaOH spot-test: negative.

Ecology and distribution. A saprophytic soil-borne fungus which has been isolated from dead tissue of various herbaceous and woody plants; widespread, almost cosmopolitan.

Culture studied. CBS 368.91 (PD 78/1096), ex Juniperus communis (Cupressaceae), Switzerland; CBS 369.91 (PD 88/614) ex Soutern Pine Pole, U.S.A.; CBS 370.91 (PD 70/999) ex Apium graveolens (Umbelliferae), the Netherlands.

# 11. Phoma apiicola Kleb. - Fig. 11

Phoma apiicola Kleb., Z. PflKrankh. 20 (1910) 22. Selected literature. Goossens (1928).

### Description in vitro

OA: growth-rate 17-22 mm, regular or with irregular margin, pale olivaceous-grey with concentrical zones of darker olivaceous-grey tinges, caused by numerous pycnidia; aerial mycelium poorly developed and compact, greyish; reverse smoke-grey to greyolivaceous, sometimes with greenish-olivaceous or olivaceous-buff tinges.

MA: growth-rate 17-21 mm, colony regular, pale olivaceous-grey, densely woollyhairy at centre with grey aerial mycelium; marginal zone almost without aerial mycelium; reverse ochraceous with umber centre.

CA: growth-rate 13-17 mm, regular, hazel with rather compact, greyish-olivaceous aerial mycelium; marginal zone without aerial mycelium; reverse isabelline-brownish, more like honey at margin.

Pycnidia  $168-304\times160-240~\mu m$ , globose with distinct, short neck and with distinct ostiole; olivaceous, smooth, glabrous, solitary or confluent, readily developing in concentrical zones, both in and on the agar, in places with hardly any aerial mycelium; exudate yellowish-white. Conidiogenous cells  $4-7\times3-6~\mu m$ , phialidic, globose to bottle shaped. Conidia  $3.4-4.4\times1.6-2.0~\mu m$ , average  $3.8\times1.7~\mu m$ , Q=2.0-2.5, average Q=2.3, ellipsoid, sometimes slightly constricted in the middle, usually with two very small guttules (occasionally a small percentage larger conidia may be produced:  $6.4-9.6\times2.0-4.0~\mu m$ , average  $7.5\times2.5~\mu m$ , Q=2.4-3.5, average Q=3.0).

Chlamydospores absent, but swollen cells may occur in the mycelium.

NaOH spot-test: negative.

Ecology and distribution. Wide-spread on celeriac and celery (Apium graveolens) in temperate regions of Europe and North America: 'Root Rot', 'Scab' and 'Crown Rot'. This fungus is also responsible for seedling canker. Soil-borne, sometimes also found on seed.

Culture studied. CBS 504.91 (PD 78/1073) ex Apium graveolens (Umbelliferae), the Netherlands; CBS 505.91 (PD 82/201) ex Apium graveolens (Umbelliferae), the Netherlands.

Note. On MA sometimes crystals are formed.

# 12. Phoma dorenboschii Noordel. & de Gruyter, spec. nov. — Fig. 12

Coloniae in agaro maltoso post septiem diem 47–55 mm in diam. marginem sinuata, pallide olivaceogriseae demum olivaceo-atrae, reversus similior. Chlamydosporae absentae. Crystalla in agaro hyalina, stellata vel filiformia dichotoma. Pycnidia in agaro et mycelio aerio copiosa, subglobosa vel elongata, solitaria vel agglutinata, ochracea demum olivaceo-atra, 88–360 µm in diam. Conidiophora phyalidea. Conidia hyalina, glabra, cylindracea, unicellulata, biguttulata, 3.0–5.5 × 1.5–2.0(–2.5) µm. Typus L 988.202-121 (siccus); CBS 426.90 (PD 86/551) (vivus); ex *Physostegia virginiana* (Labiatae), the Netherlands.

## Description in vitro

OA: growth-rate 52-54 mm, colony regular with sinuate outline; with sparse subfelty to floccose aerial mycelium; pale buff to buff, dull green or olivaceous-grey finally olivaceous-grey; reverse similar.

MA: growth-rate 47–55 mm, colony regular with sinuate outline, with compact, felted or finely floccose, aerial mycelium; whitish, greenish-olivaceous to greenish-black, sometimes concentrically zonate; reverse honey, olivaceous-black or greenish-olivaceous.

CA: growth-rate 60-70 mm, colony regular with sinuate outline, with whitish, floccose aerial mycelium; pale grey-olivaceous to grey-olivaceous or iron-grey with greenisholivaceous outer margin; reverse cinnamon to grey-olivaceous or olivaceous black.

Pycnidia  $88-360~\mu m$ , globose or elongate with up to 5 ostioles with an elongated neck; ochraceous then greenish-black, glabrous, single or confluent, abundant in aerial mycelium and both on and in the agar; exudate buff. Conidiogenous cells phialidic, globose,  $4-6\times3-6~\mu m$ . Conidia  $3.0-5.5\times1.5-2.0(-2.5)~\mu m$ , average  $4.2-4.4\times1.7-1.8~\mu m$ , Q=1.5-3.5, average Q=2.5, oblong to subcylindrical, usually with two small, polar guttules.

Crystals: whitish, bryoid 'ice-fern' crystals are formed within one week in malt-agar at centre of colony.

NaOH spot-test: negative.

Ecology and distribution. On stems and leaves of *Physostegia virginiana* and *Callistephus* sp., causing leaf spots and anthracnoses; found in the Netherlands only, but probably of foreign origin.

Culture studied. CBS 426.90 (PD 86/551, type-strain) ex Physostegia virginiana (Labiatae), the Netherlands; CBS 320.90 (PD 86/932) ex Physostegia virginiana (Labiatae), the Netherlands.

Note. Phoma dorenboschii is named in honour of Ms. Miek Dorenbosch who contributed substantially to our knowledge of Phoma taxa in pure culture during her about 30 years career as a mycologist at the Plant Protection Service, Wageningen. So far known it is the only species in sect. Phoma that forms ice-fern crystals in pure culture (Noordeloos et al., 1992).

# Phoma anigozanthi Tassi — Fig. 13

Phoma anigozanthi Tassi, Boll. R. Orto bot. (Boll. Lab. Orto Bot.) Siena 3 (2) (1900 ['1899']) 148.

Description in vitro

OA: growth-rate 43-44 mm, regular, flat, without aerial mycelium; colony distinctly zonated with concentric zones, citrine to greenish-olivaceous with paler margin; reverse similar.

MA: growth-rate 51 mm, regular, with abundant hairy to floccose-woolly, white then grey-olivaceous aerial mycelium; colony greenish-olivaceous towards margin more like citrine; reverse similar.

CA: growth-rate 53-54 mm, regular, with hardly any aerial mycelium; colony colour greenish-olivaceous then olivaceous-black with citrine margin; reverse similar.

Pycnidia  $120-240 \times 128-240 \,\mu m$ , solitary or confluent, 2-5 pycnidia together, irregularly globose to bottle-shaped, with one to three ostioles on a short neck (that develops into a longer neck in a later stage); olivaceous to olivaceous-black, glabrous, smooth, exudating a pale vinaceous or salmon-saffron conidial slime; readily developing on the agar and in the aerial mycelium, rarely in the agar, abundant after 7 days in distinct concentrical rings. Conidiogenous cells phialidic, globose or lageniform, sometimes elon-

gated,  $2.5-6\times2.5-5$  µm. Conidia  $3.2-4.2\times1.6-2.0$  µm, average  $3.7\times1.8$  µm, Q=1.8-2.6, average Q=2.1, ellipsoid, sometimes with very small polar guttules.

NaOH spot-test: green at first, then turning orange-red.

Ecology and distribution. A pathogen of Anigozanthus spp. (Kangaroo-Paw; Amaryllidaceae), causing leafspots and dieback: 'Leaf Blotch'. The fungus is probably indigenous to Australia, but so far only recorded in Europe.

Culture studied. CBS 381.91 (PD 79/1110) ex Anigozanthus sp. (Amaryllidaceae),

the Netherlands.

# 14. Phoma viburnicola Oud. - Fig. 14

Phoma viburnicola Oud., Versl. gewone Vergad. wis- en natuurk. Afd. K. Akad. Wet. Amst. 9 (1900) 298; Ned. Kruidk. Archf III, 2 (1) (1900) 247.

Phyllosticta opuli Sacc., Michelia 1 (2) (1878) 146; not Phoma opuli Thüm., Hedwigia 2 (1882) 24.

Selected literature. Bocrema & Griffin (1974).

### Description in vitro

OA: growth-rate 47-48 mm, regular, with some greyish aerial mycelium at centre, colony colour pale olivaceous-grey to grey-olivaceous with more or less colourless margin; reverse similar.

MA: growth-rate 47-49 mm, regular, with dense woolly, white or olivaceous aerial mycelium; especially in central part; colony colour olivaceous-grey to greenish-olivaceous, margin olivaceous-black or grey-olivaceous; reverse grey-olivaceous to olivaceous-black.

CA: growth-rate 56-58 mm, regular, with dense woolly or floccose, white to grey aerial mycelium; colony colour olivaceous-grey to grey-olivaceous; reverse grey-olivaceous to olivaceous-black.

Pycnidia  $140-300 \times 100-260~\mu m$ , globose to elongated, bottle-shaped, solitary or confluent in clusters of 2-3 specimens, olivaceous to olivaceous-black round ostiole, with 1 or 2 to 3 ostioles, with very short neck, that develops into a longer neck in a later stage; glabrous, smooth, scattered or in concentric rings, both in and on the agar as well as in the aerial mycelium; exudate whitish. Conidiogenous cells  $3-6 \times 3-6~\mu m$ , phialidic, globose, thin-walled. Conidia  $3.6-5.6 \times 1.6-2.2~\mu m$ , average  $4.5 \times 1.8$ , Q = 2.0-3.0, average Q = 2.5, ellipsoid, with polar guttules.

NaOH spot-test: not specific, a slight discolouring to sienna may occur.

Ecology and distribution. In Europe a widespread occurring opportunistic pathogen of woody plants (in our work encountered in members of Caprifoliaceae, Cupressaceae, Hippocastanaceae, Aquifoliaceae, Rubiaceae, Liliaceae and Rosaceae). The fungus is originally described from leaf spots and stem lesions on Viburnum spp., but serious disease symptoms on those shrubs are usually caused by Phoma viburni (Roum. & Sacc.) Boerema & Griffin, a quite different species with significantly larger and sometimes in part uniseptate conidia.

Culture studied. CBS 371.91 (PD 81/413) ex Chamaecyparus lawsoniana (Cupressaceae), The Netherlands; CBS 500.91 (PD 83/322) ex Ilex aquifolium (Aquifoliaceae), the Netherlands.

## Phoma eucalyptica Sacc. — Fig. 15

Phoma eucalyptica Sacc., Sylloge Fung. 3 (1884) 78 [as '(Thüm.)', but nom. nov.]. — Coniothyrium eucalypti Thüm. in Instituto Coimbra 27 sub Contr. Fl. myc. Lusit. II n. 341. (1880 ['1879 e 1880']); quoted in Hedwigia 19 (1880) 151; not Phoma eucalypti Cooke & Kickx, Sylloge Fung. 3 (1884) 78.

### Description in vitro

OA: growth-rate 43-47 mm, regular, with or without adpressed and sparse, woolly, olivaceous-grey aerial mycelium; with distinct radiating hyphal strands or not; colony colour olivaceous-grey with citrine outer margin; reverse olivaceous with citrine margin.

MA: growth-rate 50-54 mm, regular, with white to olivaceous-grey, woolly aerial mycelium; colony colour olivaceous-black with brown tone with slightly paler margin; reverse similar.

CA: growth-rate 50-53 mm, regular with distinct radially orientated hyphal strands, with sparse, finely floccose, olivaceous-grey aerial mycelium; colony colour brown-olivaceous to olivaceous, often with paler, citrine margin; reverse similar.

Pycnidia  $120-250\times80-200~\mu m$ , solitary or confluent in clusters of 2-5 specimens, globose, with 1-5 ostioles, usually papillate or with distinct neck, olivaceous to olivaceous-black, with white to vinaceous-buff or saffron exudate; glabrous, abundant, scattered or in concentric zones, on and in the agar. Conidiogenous cells  $3-7\times2-8~\mu m$ , phialidic, broadly globose to bottle-shaped, thin-walled. Conidia  $2.8-4.2\times1.0-2.0~\mu m$ , in average  $3.2-3.5\times1.4-1.8~\mu m$ , Q=1.7-3.4, in average Q=2.0-2.5, eguttulate, ellipsoid to subcylindrical.

Chlamydospores absent, but simple, globose or ellipsoid swollen cells may be present in MA after 4 weeks.

NaOH spot-test: not a specific reaction, but a slight discolouration to sienna may occur.

Ecology and distribution. Opportunistically parasitic on members of the Myrtaceae in association with 'Shoot Wilt' of Eucalyptus spp., especially E. globulus (Australian Gumtree), and with 'Leaf Necrosis' and blister symptoms on Eugenia spp., especially E. aromatica (clove-tree). Common in Australasia; once also isolated from seawater near Yugoslavia.

Culture studied. CBS 508.91 (PD 73/1413) ex seawater, Yugoslavia; CBS 377.91 (PD 79/210) ex Eucalyptus sp. (Myrtaceae), leaf, W. Australia; CBS 378.91 (PD 82/107) ex Eugenia aromatica (Myrtaceae); 'Blister disease'.

# 16. Phoma tropica R. Schneid. & Boerema - Fig. 16

Phoma tropica R. Schneider & Boerema, Phytopath. Z. 83 (1975) 361-365.

#### Description in vitro

OA: growth-rate 50-53 mm, regular, without or with poorly developed downy, dark olivaceous aerial mycelium; colony colourless to greenish-olivaceous to dull green, outer margin light green; reverse similar.

MA: growth-rate 52-54, regular, without or with poorly developed aerial mycelium, colony dull green, more olivaceous at centre and more honey to fawn at margin or entirely olivaceous-grey; reverse similar or slightly darker.

CA: growth-rate 53-60 mm, regular, without or with poorly developed aerial mycelium; colony greenish-olivaceous to olivaceous or grey-olivaceous to olivaceous-black, with greenish-olivaceous margin; reverse similar or darker.

Pycnidia 100-350(-400) µm, single or confluent, with 1 to 5 distinct ostioles, but hardly any neck; greenish-olivaceous to olivaceous black, at first around ostiole, glabrous, very abundant, on and partly in the agar, entirely scattered over colony or in concentric rings; exudate white-yellowish. Conidiogenous cells  $2-6\times3-6$  µm, phialidic, globose, thin-walled. Conidia  $3.0-5.9\times1.2-2.1$  µm, average  $3.7-4.4\times1.6-2.0$  µm, Q=1.6-3.2, average Q=2.2, ellipsoid with two distinct, polar guttules.

NaOH spot-test: negative

Ecology and distribution. A saprophyte from tropical regions that is commonly found in heated glasshouses in Europe, where it occurs on dead tissue of a wide variety of ornamental plants.

Culture studied. CBS 497.91 (PD 79/209) ex Coffea arabica (Rubiaceae), origin unknown; CBS 498.91 (PD 75/698) ex Poinsettia sp. (Euphorbiaceae), the Netherlands.

## 17. Phoma hedericola (Dur. & Mont.) Boerema - Fig. 17

Phoma hedericola (Dur. & Mont.) Boerema, Trans. Br. mycol. Soc. 67 (1976) 295. — Phyllosticta hedericola Dur. & Mont., Flora d'Algérie crypt. 1 (1849) 611 [as 'hederaecola']. — Phyllosticta destructiva var. hederae (Dur. & Mont.) Oudemans, Ned. Kruidk. Archf II, 1 (3) (1873) 257 [name change].

Selected literature. Bocrema (1976).

#### Description in vitro

OA: growth-rate 55-61 mm, regular, with or without sparse, whitish aerial mycelium; colony colourless with olivaceous sectors or centre, with numerous clustered pycnidia over whole colony; reverse similar.

MA: growth-rate 40-52 mm, regular, with or without floccose, white aerial mycelium; colony more or less colourless or with olivaceous sector; reverse pale luteous.

CA: growth-rate 35-61 mm, slighty irregular, with floccose, white to olivaceous-grey aerial mycelium; colony colourless with olivaceous sectors or entirely grey-olivaceous to olivaceous-black; reverse similar.

Pycnidia  $90-140\times80-140~\mu m$ , solitary or confluent, globose or irregularly shaped, with 1 to 2, sometimes 3 ostioles, without distinct neck; olivaceous, with white conidial slime, glabrous or with hyphal strands, readily or slowly developing, scattered all over the colony, mostly on the agar, sometimes in the agar or in the aerial mycelium; exudate whitish. Conidiogenous cells  $4-6\times3-6~\mu m$ , globose, thin-walled. Conidia  $(3.5-)4.0-6.0\times(1.5-)2.0-2.8~\mu m$ , average  $4.8\times2.3~\mu m$ , Q=1.7-2.3, average Q=2.0, broadly ellipsoid with two or more, small, usually polar guttules.

NaOH spot-test: yellowish-green turning into reddish-rust.

Ecology and distribution. A pathogen of Hedera spp. (Araliaceae), causing necroses on leaves and stems: 'Leaf Spot'. Probably cosmopolitan.

Culture studied. CBS 366.91 (PD 70/811) ex Hedera helix (Araliaceae), the Netherlands; CBS 367.91 (PD 87/229) ex Hedera helix (Araliaceae), the Netherlands.

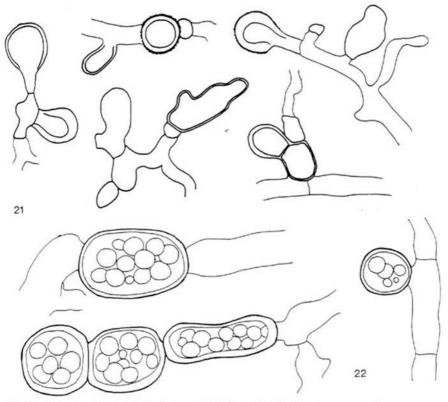


Fig. 21. Phoma multipora. Chlamydospores (x 1250). — Fig. 22. Phoma eupyrena. Chlamydospores (x 1250).

# 18. Phoma eupyrena Sacc. - Figs. 18, 22

Phoma eupyrena Sacc., Michelia 1 (5) (1879) 525. Selected literature. Dorenbosch (1970), Morgan-Jones & Burch (1988).

# Description in vitro

OA: growth-rate 48-54 mm, regular, with small tuft of olivaceous aerial mycelium at centre only; colony colour dull green or herbage green to olivaceous-black; reverse similar.

MA: growth-rate 48-60 mm, regular, with abundant woolly, whitish-grey to olivaceous-grey aerial mycelium; colony colour variable from dull green to olivaceous-buff, greenish-olivaceous to olivaceous-black; reverse greenish-olivaceous to olivaceous-black.

CA: growth-rate 53-56 mm, regular, with woolly to floccose, white to grey-olivaceous aerial mycelium; colony colour dull green to olivaceous-grey or grey-olivaceous, with some olivaceous-black sectors; reverse similar.

Pycnidia  $120-260 \times 100-260 \,\mu\text{m}$ , very variable in size, solitary or confluent, more or less globose with very distinct neck; ochraceous then olivaceous to olivaceous-black, glabrous, smooth, abundant on and (partly) in the agar, scattered or arranged in more or less concentric rings; exudate whitish. Conidiogenous cells  $3-8 \times 3-7 \,\mu\text{m}$ , phialidic, broadly globose, thin-walled. Conidia  $4.2-5.6 \times 1.8-2.4 \,\mu\text{m}$ , average  $4.9 \times 2.1 \,\mu\text{m}$ , 0 = 2.0-2.9, average 0 = 2.4, ellipsoid with two large, polar guttules.

Chlamydospores simple,  $8-20\times6-15$  µm, intercalary or terminal, single or in chains of globose, relatively thick-walled elements, ochraceous-olivaceous with numerous dark greenish guttules.

NaOH spot-test: negative.

Ecology and distribution. A cosmopolitan, soil-inhabiting fungus, which may cause damping-off of seedlings of herbaceous and woody plants; formerly considered as a specific fungus of Potato.

Culture studied. CBS 375.91 (PD 78/745) ex Phaseolus vulgaris (Papilionaceae), the Netherlands; CBS 374.91 (PD 78/391) ex Solanum tuberosum (Solanaceae), the Netherlands.

# Section Sclerophomella (compare Boerema, Loerakker & Wittern, 1986)

Species characterized by thick-walled pycnidia with late formation of an opening (pore instead of ostiole).

# 19. Phoma incompta Sacc. & Mart. - Fig. 19

Phoma incompta Sacc. & Mart., Sylloge Fung. 10 (1892) 146. Selected literature. Malathrakis (1979).

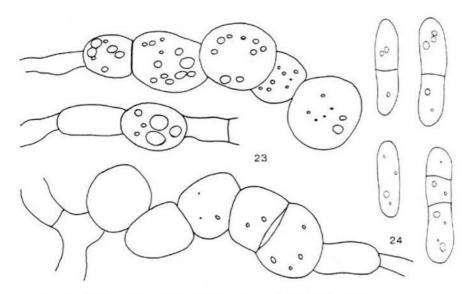
## Description in vitro

OA: growth-rate 21–22 mm (14 days: 40–42 mm), regular, without or with sparse, woolly, grey-olivaceous aerial mycelium; colony colour chestnut, towards margin ochraceous; reverse similar or with weak greenish tinge, distinctly zonated from pycnidial concentric rings.

MA: growth-rate 19-21 mm (14 days: 24-35 mm), regular, with abundant, white to olivaceous buff or greenish-olivaceous aerial mycelium; colony colour greenish-olivaceous to dark herbage green with darker centre (towards olivaceous-grey); reverse between sepia and olivaceous, with outer margin more like citrine.

CA: growth-rate 14–19 mm (14 days: 27–34 mm), regular, with woolly, pale green aerial mycelium; colony colour pale greenish-olivaceous with amber coloured spots; reverse umber to fuscous-black with ochraceous margin.

Pycnidia  $50-300 \times 50-250$ , single but usually confluent in dense clusters, thick-walled, dark olivaceous to rusty-blackish, with an indistinct opening visible as a pallid spot, and occurring only late in the pycnidial development; exudate sordid white to pale violaceous; covered in mycelial hairs, abundant in concentric rings, mainly on, but also in the agar. Conidiogenous cells  $3-7 \times 2-8 \,\mu\text{m}$ , phialidic, globose, thin-walled. Conidia  $3.0-3.6 \times 0.8-1.0 \,\mu\text{m}$ , av.  $3.2 \times 0.9 \,\mu\text{m}$ , Q = 3.0-4.5, av. 3.7, slenderly cylindrical, eguttulate.



Figs. 23-24. Phoma dictamnicola. 23. Chlamydospores (x 1250); 24. conidia in vivo (x 1250).

Chlamydospores absent, but swollen cells may occur in the mycelium.

NaOH spot-test: red-brown with bluish margin.

Ecology and distribution. A pathogen of Olea europaea (olive) in southern Europe (Greece, Italy). In naturally infected trees it may cause the characteristic symptoms of a vascular wilt disease: 'Shoot Wilt'. It has also been found on the fruits.

Culture studied. CBS 652.77 (PD 76/1013) ex Olea europaea (Oleaceae), Greece; CBS 526.82 (PD 82/786) ex Olea europaea (Oleaceae), Italy.

# Phoma dictamnicola Boerema, de Gruyter & Noordel., nom. nov. — Figs. 20, 23, 24.

Ascochyta nobilis Kabát & Bubák, Ost. bot. Z. 54 (1904) 3; not Phoma nobilis Sacc., Michelia 2 (3) (1882) 616 [= Phomopsis sp.].

Phyllosticta dictamni Fairman, Annls mycol. 8 (1910) 324; not Phoma dictamni Fuckel, Jb. nassau. Ver. Naturk. 23-24 [= Symb. mycol.] (1870 ['1869 und 1870']) 125.

## Description in vitro

OA: growth rate 47–48 mm, colonies regular, with very conspicuous whitish-greyish, adpressedly woolly-felted aerial mycelium all over the colony, total colour greyish-olivaceous to greenish-olivaceous at margin, reverse beige at centre, towards margin greenish-olivaceous.

MA: growth rate 54-67 mm, colony irregular, with densely woolly-felted, whitish aerial mycelium; total impression pale olivaceous-grey; reverse honey with olivaceous-black patches, caused by abundant production of pycnidia.

CA: growth rate 68 mm, colony regular, with abundant, whitish, floccose aerial mycelium; reverse rather pale beige.

Pycnidia  $250-450~\mu m$ , solitary or in clusters of 2-3, thick-walled, more or less globose, without distinct neck; opening occurs only late in the pycnidial development, greenish-olivaceous; abundant, especially towards the margin of the colony, usually in the agar, part of them half in the agar; exudate not observed. Conidiogenous cells  $4-7 \times 4-7 \mu m$ , phialidic, globose, thin-walled. Conidia [in vitro always aseptate and relatively small]  $3.8-5.4 \times 1.4-2.4~\mu m$ , average  $4.6 \times 1.8~\mu m$ , Q = 2.0-3.1, average Q = 2.6, ellipsoid, sometimes reniform, without visible guttules.

Chlamydospores 8-12 µm in diameter, simple, usually intercalary chains of globose, greenish-olivaceous cells.

NaOH spot-test: negative.

Ecology and distribution. A serious pathogen of Dictamnus albus, frequently found in Eurasia and North America: 'Leaf Spot'. The small conidial dimension in vitro may led to confusion with the spermatial state Asteromella dictamni Petrak (syn. Phyllosticta dictamnicola Lobik) of Mycosphaerella dictamni Petrak (anam. Septoria dictamni Fuckel).

Culture studied. CBS 507.91 (PD 74/148) ex Dictamnus albus (Rutaceae), the Netherlands.

Note. Phoma dictamnicola produces in vivo on the leaf spots often variable number of relatively large 1-septate conidia. In vitro, and on dead stems, however, the conidia always remain small and aseptate.

#### REFERENCES

- Aa, H.A. van der, M.E. Noordeloos & J. de Gruyter. 1990. Species concept in some larger genera of the Coelomycetes. Stud. Mycol. 32: 3-19.
- Boerema, G.H. 1976. The Phoma species studied in culture by Dr. R.W.G. Dennis. Trans. Br. mycol. Soc. 67: 289-319.
- Boerema, G.H. 1985. Mycologisch-Taxonomisch onderzoek aan Bodem Phoma's. Versl. Meded. Plziektenk. Dienst Wageningen 163 (Jaarb. 1984): 34–40.
- Boerema, G.H. & M.M.J. Dorenbosch. 1973. The Phoma and Ascochyta species described by Wollenweber and Hochapfel in their study on fruit-rotting. Stud. Mycol. 3.
- Boerema, G.H. & M.J. Griffin. 1974. Phoma species from Viburnum. Trans. Br. mycol. Soc. 63: 109–114.
- Boerema, G.H., J. de Gruyter & M.E. Noordeloos (in prep). Specific and infraspecific taxa of Phoma differentiated on account of cultural characteristics.
- Boerema, G.H., W.M. Loerakker & I. Wittern. 1986. Zum Auftreten von Phoma nigrificans (P. Karst.) comb. nov. (Teleomorph Didymella macropodii Petrak) an Winterraps (Brassica napus L. var. oleifera Metzger). JI Phytopath.15: 269–273.
- Constantinescu, O. & H.A. van der Aa. 1982. Phoma flavigena sp. nov., from fresh water in Romania. Trans. Br. mycol. Soc. 79: 343-345.
- Dorenbosch, M.M.J. 1970. Key to nine ubiquitous soil-borne Phoma-like fungi. Persoonia 6: 1-4.
- Goossens, J. A. A. M. H. 1928. Onderzoek over de door Phoma apiicola Klebahn veroorzaakte schurftziekte van knolselderij en over synergetische vormen en locale rassen van deze zwam. Tijdschr. PlZiekt. 34: 271–348.
- Malathrakis, N.E. 1979. A study of an olive tree disease caused by the fungus Phoma incompta Sacc. et Mart. [in Greek]. 123 pp. Thesis Agric. Coll. Athens.
- Morgan-Jones, G. & K.B. Burch. 1988. Studies in the genus Phoma. X. Concerning Phoma eupyrena, an ubiquitous, soil-borne species. Mycotaxon 31 (2): 427–434.

Noordeloos, M.E., J. de Gruyter, G.W. van Eijk & H.J. Roeijmans (in manuscr.). Phoma-like fungi forming Ice-fern crystals in pure culture: taxonomic implications of morphology, cultural characteristics, pathology and biochemistry.

Padwick, G.W. 1950. Manual of rice diseases. CMI Kew.

Rayner, R.W. 1970. A mycological colour chart. CMI Kew.

Shukla, N.P., R.K. Rajak, G.P. Agarwal & D.K. Gupta. 1984. Phoma minutispora as a human pathogen. Mykosen 27: 255-258.

#### PERSOONIA

Published by Rijksherbarium / Hortus Botanicus, Leiden Volume 15, Part 1, pp. 93-99 (1992)

# STUDIES ON THE CHARACTER VARIABILITY IN THE RAMSBOTTOMIA CREC'HQUERAULTII COMPLEX (PEZIZALES)

B. KULLMAN<sup>1</sup> & J. VAN BRUMMELEN<sup>2</sup>

The variability of the margin of the apothecium and the spore ornamentation of the Ramsbottomia cree' hqueraultii complex has been studied by light microscopy and scanning electron microscopy (SEM) to estimate their taxonomic value.

The taxonomic value of the structure of the apothecial margin is a controversial issue in the systematics of Pezizales. The most important distinguishing characters of the genera are the presence or absence of hairs and their structure and colour.

The genus Ramsbottomia W.D. Buckley emend. Benkert & Schumacher (1985) is especially interesting in this respect since the apothecium of its R. crec' hqueraultii complex has been described as 1) externally smooth, with an indistinct margin of textura porrecta by Boudier (1904-1911), Seaver (1928), Rifai (1968), and Gamundi (1975) for Lamprospora crec'hqueraultii (Crouan) Boud, and by Eckblad (1968) for L. ovalispora (Svrček & Kub.) Eckbl., as well as 2) covered with hairs by Seaver (1928) for Sphaerosporella perplexa Seav., by Gamundi (1975) for L. crec'haueraultii (Crouan) Boud, var. modesta (P. Karst.) Gamundi, by Dennis (1978) for L. crec'hqueraultii (Crouan) Boud., and by Caillet & Moyne (1980) for Octospora crec' hqueraultii (Crouan) Caillet & Moyne. This character has also been described as 3) somewhat intermediate between both possibilities mentioned: "... apothecia ... smooth or with hardly visible light brownish hyphal element at the thick margin" by Moravec (1978) for L. crec'hqueraultii (Crouan) Boud. var. modesta (P. Karst.) Gamundi. Benkert (1976) described Lamprospora crec'hqueraultii (Crouan) Boud, as having hair-like hyphae at the margin of the apothecium. The hyphae are hyaline, more or less brown and wavy, varying within the specimen, but Benkert wrote: "Man kann diese Hyphen zwar nicht als echte Haaren bezeichnen."

Buckley (1923), however, described *Ramsbottomia lamprosporoidea* as having a margin covered with pale brown hairs 160–285 μm long and 11–18 μm wide.

In the present paper the main attention is paid to the variability of the margin of the apothecium and of the spore ornamentation in order to estimate the taxonomic value of these characters.

#### MATERIALS AND METHODS

In the course of this study 30 specimens of the Ramsbottomia crec'hqueraultii complex in the herbarium of the Institute of Zoology and Botany of the Academy of Sciences of

Institute of Zoology and Botany of the Academy of Sciences of Estonia, 21 Vanemuise St., EE 2400 Tartu, Estonia.

<sup>2)</sup> Rijksherbarium / Hortus Botanicus, P.O. Box 9514, 2300 RA Leiden, The Netherlands.

Estonia (TAA) were studied. Another part of the material had been obtained from other herbaria (Benkert's Herbarium, CO, CUP, H, L, NY, O, and PC). The following specimens were examined.

WEST EUROPE. France: Finistère, s. loc., on soil of banks of brooks, IX and X.1858, Crouan (lectotype of Ascobolus crec'hqueraultii, PC); Finistère, Marais de Pontanéven, on moist ground, 18.V.1865, Crouan (as Ascobolus crec'hqueraultii, CO). — The Netherlands: prov. N.-Brabant, Nuenen, among mosses, 4.VI.1991, H. Huijser (TAA, L); Urkhoven, among mosses, 9.VI.1991, H. Huijser & B. Kullman (TAA).

CENTRAL EUROPE. Germany: near Suhl, Hargrund, Friedberg, on the ground, 17.IX.1981, D. Benkert 5430/1 (as Ramsbottomia crec'hqueraultii, TAA); Rostock, Mooskuhler by Neuhirsberg, on moist sand, 19.IX.1985, D. Benkert (as R. crec'hqueraultii, TAA); Potsdam, Fresdorf bog, on the ground, 29.X.1969, D. Benkert (as R. crec'hqueraultii, TAA); Zechengrund bei Oberwiesental, on moist ground among Pnellia, 23.IX.1986, D. Benkert (as R. crec'hqueraultii, TAA).

NORTH EUROPE. Norway: Hedmark, Os, Tufsinga, Straumvollen, on silt among Carex aquacilis, 22.VII.1976, T. Schumacher 134/76 (as Lamprospora ovalispora, TAA ex O). — Russian Rep.: Karelia, Kalevala, Mikkola, among Pinetum myrtillosum on the ground, 9.VIII.1965, E. Parmasto (TAA 18143). — Sweden: Torne Lappen, Jukkasj rvi s:n, Abisko, on the ground among mosses, path to Njakajure, 26.VII.1927, J.A. Nannfeldt & H. Smith (as Peziza modesta P. Karst., det. J.A. Nannfeldt; Lamprospora crec'hqueraultii det. R.P. Korf; CUP 49535). — Finland: Mustiala, 1876, P.A. Karsten (as Humaria modesta P. Karst., det. P.A. Karsten, H; as L. exapetala, NY).

EAST EUROPE. Estonia: Dist. Võru, Vana-Kasaritsa, on silt among Carex sp., 8.VI.1986, B. Kullman (TAA 116172); Dist. Tartu, Ahunapalu, Apnasaar, on the ground, 19.VIII.1990, K. Kalamees (TAA 144733). — Russian Rep.: Komi, Cherdynsk, Visay, on the ground among mosses, 9.IX.1988, B. Kullman (TAA 117118).

WEST SIBERIA. Russian Rep.: Yamal-Nenets A. Okr., Krasnosel'kup, river Taz, on the ground among Laricetum ledosum, 31.VII.1964, E. Parmasto & I. Maasik (as R. ovalispora, TAA 17003); Gavotuy, Polar Urals, on the ground, 5.VII.1966, A. Sirko (TAA); Shurush, Ovgort, on the ground, 27.VII.1976, M. Murdvee (TAA 110047).

ARCTIC. Russian Rep.: Yamal-Nenets A. Okr., Slantsevaya, Krasny Kamen, Polar Urals, on the ground, 11.VIII.1966, A. Sirko (TAA).

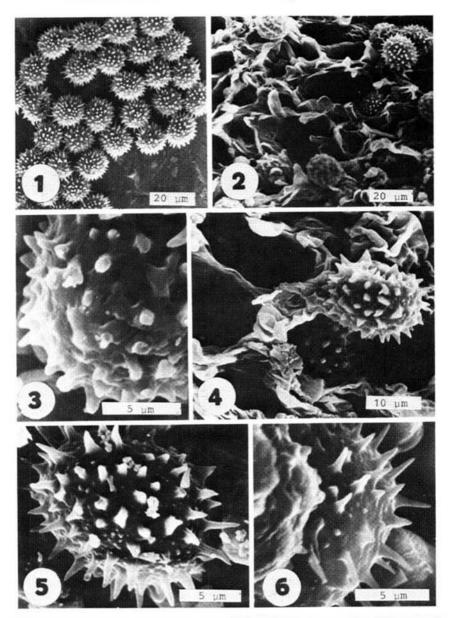
THE FAR EAST. Russian Rep.: Amur Dist., Dzeltulakskiy Region, Mogot, 200–400 m high, on the ground of a forest path, 27.VII.1961, A. Raitviir (TAA 42026); Mogot, on the ground, 29.VII.1961, A. Raitviir (TAA 42079); Svobodny, on the ground, 15.VIII.1959, B. Tomilin (TAA); Khabarovsk Dist., Oblutse Region, on sandy ground in a wood of Picea abies, 8.VIII.1961, A. Raitviir (TAA 42165); Selikhino, on the ground in a birch wood, 22.VII.1961, A. Raitviir (TAA 42355); Yuzny, Sikhote-Alin' mountains, near the river Matay, on the ground, 14.VII.1973, B. Kullman (TAA 66495); Primor'ye Dist., Partisanskiy, Sikhote-Alin' mountains, on the ground in the wood, 16.VIII.1986, I. Parmasto (TAA 125955); Reservation Lazo, Amerika, near a river, on the ground among mosses, 9.VIII.1986, B. Kullman (TAA 116338); on the ground, 12.VIII.1986, B. Kullman (TAA 116403); Kamchatka Dist., Reservation Kronokskiy, Uzon Caldera, Pineto-Alnetum, on the ground, 22.VIII.1978, K. Kalamees (TAA 120188).

NORTH AMERICA. U.S.A.: New York, near Yonkers, on the ground in the wood, 2.X.1919, F.J. Seaver (as Sphaerospora perplexa Seav., NY).

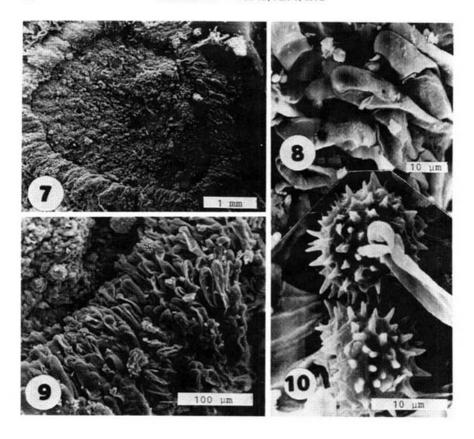
# Light and scanning electron microscopy (SEM)

The fungal fruit-bodies were observed and measured with an 'Amplival' microscope. A watery solution of 2% potassium hydroxide (KOH) was used as an observation medium for fruit-bodies. The spore wall markings were stained in a solution of cotton blue in lacto-phenol and the spores were measured using the immersion objective HI 100. At least ten spores were measured in each specimen.

The ornamentation of the spores was studied and scanning electron micrographs were taken with a Tesla BS 301.



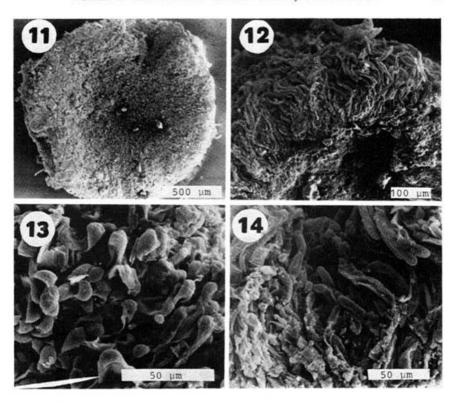
Figs. 1–6. Ramsbottomia crec'hqueraultii: 1, spores (TAA 117118); 2, 4, spores (TAA 110047); 3, 5, 6, spores (T. Schumacher, 134/76; TAA ex O).



Figs. 7-10. Ramsbottomia crec'hqueraultii: 7, apothecium and 9, margin (TAA 116338); 8, hairs and 10, spores (TAA 66495).

#### RESULTS AND DISCUSSION

Rifai (1968) wrote that "it might well be necessary to uphold Ramsbottomia for the reception of Lamprospora crec'hqueraultii and its related species now classified as Lamprospora or Octospora, which should be distinguished from the last two genera by the difference in the structure of its excipular tissue, especially in the absence of a thin and compact prosenchymatous layer on the outer surface of the receptacle." Taking this into account, Benkert & Schumacher (1985) have described Ramsbottomia W.D. Buckley emend. Benkert & T. Schumacher. In addition, they stress two important circumstances, viz. differently from the genus Lamprospora De Not. the genus Ramsbottomia is not bryo-parasitic and it has prominent, brownish, hyphoid hairs towards the margin, arising from outermost excipular cells.



Figs. 11-14. Ramsbottomia crec'hqueraultii: 11, apothecium (TAA 110047); 12, margin and 14, hairs (TAA 125955); 13, hairs (B. Tomilin, 15.VIII.1995; TAA).

In the opinion of the present authors it is the latter morphological character that seems to be the most important. The margin may be more or less distinct (Figs. 7, 11) but the presence of the hairs similar to those of the genera *Aleuria* or *Melastiza* is obligatory (Figs. 7–9, 11–14).

The statistical test applied to the variability of hair length dimensions shows that it may be homogeneous in the investigated material.

The spore ornamentation of the whole studied materials is relatively uniform and does not vary notably in different geographical populations (Figs. 1–6, 10), when we take into account that the spores in a single apothecium can already be rather different.

The shape of the spores varies from a perfect sphere to subglobose. A more detailed account of the variability of spores in *Ramsbottomia crec'hqueraultii* will be presented in a forthcoming paper.

On the basis of comparative morphological study the authors consider all specimens studied to belong to the same species. It is a holarctic arcto-boreo-temporate species (Fig. 15) which has evidently been distributed from its centre of diversity in the Far East (Sikhote Alin' mountains), where the variability of its characters is remarkably larger than that of its neighbouring populations, to Europe during the late-glacial period and the Lower Holocene, like the Polyporaceae (Laasimer, 1965). Ramsbottomia crec'hqueraultii has quite a wide ecological amplitude.

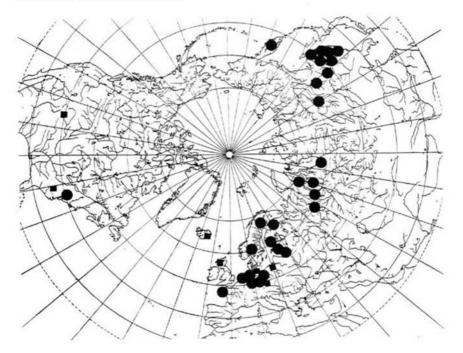


Fig. 15. Holarctic arcto-boreo-temporate distribution of Ramsbottomia crec'hqueraultii ( $\bullet$  = according to the specimens examined during this study;  $\blacksquare$  = taken from literature).

## Ramsbottomia crec'hqueraultii (Crouan) Benkert & T. Schumach.

Ascobolus crec'hqueraultii Crouan, Annls Sci. nat. (Bot.) IV, 10 (1858) 194, pl. 130 figs. 12–16. — Lamprospora crec'hqueraultii (Crouan) Boudier, Icon. mycol., Sér. I, livr. 2 (1904), Liste préliminaire, unnumbered page. — Ramsbottomia crec'hqueraultii (Crouan) Benkert & T. Schumach., Agarica 6 (12) (1985) 33.

Peziza asperior Nyl., Obs. Peziz. fenn. (1868) 21. — Ciliaria asperior Boud., Hist. class. Discom. Eur. (1907) 62. — Ramsbottomia asperior (Nyl.) Benkert & T. Schumach., Agarica 6 (12) (1985) 35. Peziza modesta P. Karst., Not. Sällsk. F. Fl. fenn. Förh. 10 (1869) 122. — Lamprospora crec'hqueraultii (Crouan) Boud. var. modesta (P. Karst.) Gamundi, Fl. cript. Tierra del Fuego 10 (3) (1975) 130. Sphaerospora perplexa Seaver, The North American Cup-Fungi (Operculates) (1928) 45.

Apothecia 1–6 mm in diameter. Disc orange to orange-yellow, flat to convex. Receptacle saucer-shaped; margin even, rather thick, sometimes exceeding the hymenial level; often with more or less evident hyphoid hairs arising from the excipulum towards the margin. Ectal excipulum of textura globulosa. Hairs hyphoid, cylindrical, obtuse, thick-walled, 2–4-celled, subhyaline or pale brownish, 64–260  $\mu$ m long and 6.4–22.4  $\mu$ m wide. Hymenium 230–350  $\mu$ m thick. Asci clavate, cylindrical, 8-spored. Ascospores uniseriate when dried, with many small oil drops when fresh, hyaline, subglobose or globose,  $(14.5-)14.8-18.4(-19.5)\times(12.0-)13.8-16.0(-16.4)$   $\mu$ m, ornamented with sharply pointed spines, 1.3-3.5  $\mu$ m long, up to 1.7  $\mu$ m wide, sometimes with fine warts between the spines. Paraphyses enlarged to 5.0-6.3  $\mu$ m wide at the tip, septate, straight.

Habitat. From June to October, on the ground, sometimes among mosses or species of Carex; North America, Europe, and Asia.

#### **ACKNOWLEDGEMENTS**

The first author wishes to express her sincere gratitude to Dr. D. Benkert and Dr. T. Schumacher for providing the opportunity of using the specimens collected and determined by them, to Dr. M. Rahi for his help in the work with the SEM and to Mrs. U. Martinson for linguistic help. Gratitude is expressed to the Netherlands Organization for Scientific Research (N.W.O), who subsidized the visit of the first author to the Rijksherbarium / Hortus Botanicus at Leiden.

#### REFERENCES

Benkert, D. 1976. Bemerkenswerte Ascomyceten der DDR. I. Zu einigen Arten der Gattung Lamprospora de Not. Feddes Reprium 87: 611–642.

Benkert, D. & T. Schumacher. 1985. Emendierung der Gattung Ramsbottomia (Pezizales). Agarica 6: 28-46.

Boudier, J.L. 1904-1911. Icones mycologicae, ou iconographie des champignons de la France. Paris.

Buckley, W.D. 1923. New British Discomycetes. Trans. Br. mycol. Soc. 9: 43-47.

Caillet, M. & G. Moyne. 1980. Contribution à l'étude du genre Octospora Hedw. ex S.F. Gray emend. Le Gal. Espèces à spores ornamentées, globuleuses ou subglobuleuses. Bull. trimest. Soc. mycol. Fr. 96: 175-211.

Crouan, P.L. & H.M. Crouan. 1858. Notes sur neuf Ascobolus nouveaux. Annls Sci. nat. (Bot.) IV, 10: 193–199, pl. 13.

Dennis, R.W.G. 1978. British Ascomycetes. Vaduz.

Eckblad, F.-E. 1968. The genera of the operculate Discomycetes. A re-evaluation of their taxonomy, phylogeny and nomenclature. Nytt. Mag. Bot. 15.

Gamundi, I.J. 1975. Fungi, Ascomycetes, Pezizales. Fl. cript. Tierra del Fuego 10 (3).

Grelet, L.-J. 1943. Les Discomycètes de France d'après la classification de Boudier. Dixième fascicule. Rev. Mycol. 8: 1–25.

Laasimer, L. 1965. Eesti NSV taimkate. Tallinn.

Moravec, J. 1978. Fungi of Kilimanjaro. I. Discomycetes, Pezizales. Česká Mykol. 32: 70–78.

Rifai, M.A. 1968. The Australasian Pezizales in the herbarium of the Royal Botanic Gardens Kew. Verh. K. Ned. Akad. Wet. (Natuurk.) 57 (3).

Seaver, F.J. 1928. The North American Cup-Fungi (Operculates). New York.

#### PERSOONIA

Published by Rijksherbarium / Hortus Botanicus, Leiden Volume 15, Part 1, pp. 101-103 (1992)

# MYCENA DASYPUS, A NEW MEMBER OF SECTION POLYADELPHIA

R.A. MAAS GEESTERANUS1 & T. LÆSSØE2

Mycena dasypus, collected in Surrey (England), is described as a new species in section Polyadelphia.

The species described in this short note is one of those humble fungi which are easily passed over unnoticed. And the area where Mycena dasypus was found – old heathland now under management to clear encrouching trees such as Pinus silvestris, Quercus, and Betula – hardly gives the impression of being prime terrain for mycological novelties. In fungi, however, and more especially if one concentrates on the smaller denizens, the unexpected is the rule.

# Mycena dasypus Maas G. & Læssøe, spec. nov.3 — Figs. 1-5

Basidiomata solitaria vel bina. Pileus (siccatus) 2-3.5 mm latus, hemisphericus vel plano-convexus, centro depressus, sulcatus, minute pruinosus, griseo-albidus. Caro tenuis, pallida, odore saporeque ignotis. Lamellae c. 10 stipitem attingentes, adscendentes, adnatae, albae, margine concolores. Stipes  $8-15 \times 1$  mm, apice minute pruinosus, albidus, deorsum purpureo-brunneus, basi fibrillis radiantibus, crassis, albidis substrato affixus.

Basidia (immatura) c.  $22.5 \times 6.5$ –7 µm, clavata, 4-spora, fibulata. Sporae 7.2– $9.8 \times 3.6$ –4.6 µm, amyloideae. Cheilocystidia 12.5– $14.5 \times 6.3$ –10.5 µm, clavata vel obpyriformia, fibulata, surculis simplicibus, cylindraceis 2– $4.5 \times 1$  µm instructa. Pleurocystidia nulla. Trama lamellarum iodi ope vinescens. Hyphae pileipellis 2.5–4.5 µm latae, fibulatae, surculis simplicibus vel ramosis 1.8– $5.5 \times 1$ –2 µm obtectae. Hyphae stipitis corticales 1.8–3.5 µm latae, fibulatae, surculis sparsis, simplicibus, cylindraceis 1.8– $2.7 \times 1.3$ –2 µm munitae.

Ad sarmenta Pini et Quercus.

Holotypus: T. Læssøe & B.M. Spooner TL-2360 (K).

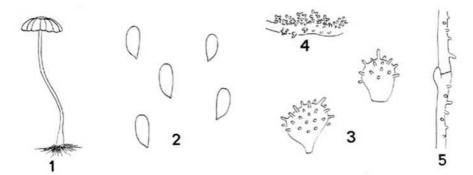
Basidiomata solitary or in twos. Pileus (dried) 2-3.5 mm across, hemispherical to plano-convex, somewhat depressed at the centre, sulcate, minutely pruinose, greyish white. Flesh thin, pallid. Odour not noted, taste not recorded. Lamellae c. 10 reaching the stipe, tender, ascending, less than 0.5 mm broad, somewhat ventricose, adnate, white, the edge convex, concolorous. Stipe  $8-15 \times 1$  mm, hollow, fragile, equal for the greater part, terete, curved, apically minutely pruinose, glabrescent farther below (but see remarks), whitish above, purplish brown below, arising from a dense patch of radiating, long, coarse, whitish fibrils.

Basidia (none seen mature) c.  $22.5 \times 6.5$ –7 µm, clavate, with four incipient sterigmata, clamped. Spores 7.2– $9.8 \times 3.6$ –4.6 µm, elongated pip-shaped, almost cylindri-

Rijksherbarium / Hortus Botanicus, P.O. Box 9514, 2300 RA Leiden, The Netherlands.

Royal Botanic Gardens, Kew, England.

Etymology: δάσύσ, hairy; πούσ, foot.



Figs. 1–5. Mycena dasypus (holotype). 1. Habit; 2. spores; 3. cheilocystidia; 4. fragment of a hypha of the pileipellis; 5. hypha of the cortical layer of the stipe. (Fig. 1, c. × 2.5; all others, × 700.)

cal, smooth, amyloid. Cheilocystidia  $12.5-14.5\times6.3-10.5~\mu m$ , forming a sterile band (lamellar edge homogeneous), clavate to obpyriform, clamped, covered with fairly few, evenly spaced, simple, cylindrical, straight excrescences  $2-4.5\times1~\mu m$ . Pleurocystidia absent. Lamellar trama vinescent in Melzer's reagent. Hyphae of the pileipellis  $2.5-4.5~\mu m$  wide, clamped, covered with simple to much branched excrescences  $1.8-5.5\times1-2~\mu m$ . Hyphae of the cortical layer of the stipe  $1.8-3.5~\mu m$  wide, clamped, sparsely covered with simple, cylindrical, straight excrescences  $1.8-2.7\times1.3-2~\mu m$ . Hyphae of the basal patch  $2.7-4.5~\mu m$  wide, aseptate, with thickened, colourless cell-walls.

Very common on site, mostly on Pinus litter but also on twigs of Quercus.

Holotype: 'Fungi britannici / Mycena dasypus Maas G. & Læssøe / England, Surrey, Esher Common / 21 Jan. 1990 / T. Læssøe & B.M. Spooner TL 2360' (K).

Additional material. 'Essex, Epping Forest, High Beeches / 13 Oct. 1991 / A. Henrici, on Rubus twig in litter' (K).

Except for the colours of the pileus and the lower part of the stipe mentioned by the collector, the description of the species is entirely based on the dried material.

Unfortunately, the type collection is somewhat scanty, while two of the specimens investigated proved to have been invaded by a parasitic fungus whose coarse hyphal ends, sticking out on all sides, simulate cheilocystidia and caulocystidia. It is not impossible that under the influence of the impaired condition large portions of the surface layers of pileus and stipe of the *Mycena* appear collapsed or covered with amorphous matter which greatly hampers observation. In spite of these shortcomings, however, the species is unmistakably recognizable as a member of section *Polyadelphia* Sing. ex Maas G. It comes near *Mycena juncicola* (Fr.) Gillet and *M. culmigena* Maas G. on account of a purplish colour, the presence of clamps, and the large spores, but it differs from both. From the former it can be separated by the greyish white pileus, the regularly shaped cheilocystidia, the aseptate hyphae of the basal patch, and the different substratum. *Mycena dasypus* differs from the latter by the colour of both its pileus and lamellae, longer spores and cheilocystidia, and its very conspicuous basal patch made up of aseptate hyphae.

If for some reason the purplish brown shade of the stipe in *M. dasypus* should prove to be not the natural or the usual colour, the species keys out (Maas Geesteranus, 1986: 161) near *Mycena herbarum* Sing. and *M. lohwagii* Sing., both of which are equally easily separable from the present taxon.

#### REFERENCE

Maas Geesteranus, R.A. 1986. Conspectus of the Mycenas of the Northern Hemisphere-6. Sections Polyadelphia and Saetulipedes. Proc. K. Ned. Akad. Wet. (Ser. C) 89: 159-182.

#### PERSOONIA

Published by Rijksherbarium / Hortus Botanicus, Leiden Volume 15, Part 1, pp. 105-107 (1992)

# MYCENA TERENA, A NEW MEMBER OF SECTION POLYADELPHIA FROM SOUTHERN NORWAY

A. ARONSEN1 & R.A. MAAS GEESTERANUS2

Mycena terena is proposed as a new species belonging to section Polyadelphia. It deviates from other members of this section on account of its smooth cheilocystidia.

It is hard to explain what it is that drives one (the first author) to rummage among fallen willow leaves in search of Mycenas, even though his experience says that much more promising habitats are to be found elsewhere. And yet, there they were. Minute specimens of a manifestly unknown *Mycena* arising from long dead, curled-up willow leaves, lifting their tiny heads on very slender black-tipped stipes.

# Mycena terena Aronsen & Maas G., spec. nov.3 — Figs. 1-11

Basidiomata solitaria vel subcongregata. Pileus usque ad 3 mm latus, campanulatus vel hemisphericus, interdum papillatus, deinde obtuse umbonatus, aetate raro depressus, plerumque sulcatus, haud translucente striatus, minute pruinosus, glabrescens, siccus, e argillaceo albido-pallescens, centro obscurior. Caro tenuis, pallida, odore nullo. Lamellae 0-8 stipitem attingentes, adnatae vel late adnatae, albae. Stipes  $5-10(-23) \times < 0.2$  mm, minute pruinosus, initio apice niger, deorsum albido-pallescens, basi fibrillis albis instructus.

Basidia  $17-25 \times 7-10 \, \mu m$ , clavata, 4-spora, fibulata, sterigmatibus  $4.5-6.5 \, \mu m$  longis munita. Sporae  $7.2-9.0 \times 4.5-5.6 \, \mu m$ , amyloideae. Cheilocystidia  $17-30 \times 6.5-10 \times 0-4.5 \, \mu m$ , sparsa, cylindracea, subclavata, fusiformia, sublageniformia, fibulata, laevia. Pleurocystidia nulla. Trama lamellarum iodi ope vinescens. Hyphae pileipellis  $4.5-10.5 \, \mu m$  latae, fibulatae, diverticulatae. Hyphae stipitis corticales  $1.5-4.5 \, \mu m$  latae, fibulatae, diverticulatae.

Ad Salicis folia decisa.

Holotypus: "Fungi norvegici / Mycena terena Aronsen & Maas G. / leg. A. Aronsen, no. A 26/90 / 14 Oct. 1990 Vestfold: Tjøme, Hvasser, Sønstegård / on buried Salix leaves on the ground" (L, no. 990. 198-234).

Basidiomata solitary or in small groups. Pileus up to 3 mm across, conical, parabolical, campanulate, hemispherical, young occasionally with a small papilla, later mostly obtusely umbonate, flattening with age, rarely plano-convex and with the centre somewhat depressed, generally shallowly sulcate, not translucent-striate, minutely pruinose, glabrescent, dry, at first beige or pale grey, often with the centre darker grey, then fading to pale grey or white, often with the centre yellowish or ochraceous brown. Flesh thin, pallid. Odour none, taste not recorded. Lamellae 0–8 reaching the stipe, usually well developed (although less than 0.5 mm broad) but occasionally only showing as faint ridges, tender, ascending, becoming subhorizontal, adnate to broadly adnate, smooth,

Etymology: τέρενα, slender.

<sup>1)</sup> Torødveien 54, N-3135 Torød, Norway.

<sup>2)</sup> Rijksherbarium / Hortus Botanicus, P.O. Box 9514, 2300 RA Leiden, The Netherlands.

white, the edge more or less concave, white. Stipe  $5-10(-23) \times < 0.2$  mm, equal, terete, flexuous, smooth, minutely pruinose, entirely black in very young specimens except for the base which is whitish, retaining the black colour only at the very apex when growing older, grey to dark grey farther below, finally fading to greyish white or watery white throughout, the base attached to the substratum by shorter or longer, fine, white fibrils.

Basidia  $17-25\times7-10~\mu m$ , clavate, 4-spored, clamped, with plump sterigmata  $4.5-6.5~\mu m$  long. Spores  $7.2-9.0\times4.5-5.6~\mu m$ , pip-shaped, smooth, amyloid. Cheilocystidia  $17-30\times6.5-10\times0-4.5~\mu m$ , occurring mixed with basidia (lamellar edge heterogeneous), cylindrical, subclavate, fusiform, sublageniform, occasionally sigmoid, clamped (but clamps easily missed), smooth, rarely with a few, very coarse excrescences, apically generally broadly rounded but also more or less narrowed to form a neck. Pleurocystidia absent. Lamellar trama vinescent in Melzer's reagent. Hyphae of the pileipellis  $4.5-10.5~\mu m$  wide, clamped (but clamps not present at every septum), densely covered with cylindrical, straight excrescences  $1-2.5\times0.9~\mu m$ . Hyphae of the cortical layer of the stipe  $1.5-4.5~\mu m$  wide, clamped (but clamps infrequent), more or less densely covered with cylindrical, sometimes more thorn-like, straight to curved excrescences  $1-3.5\times0.9~\mu m$ .

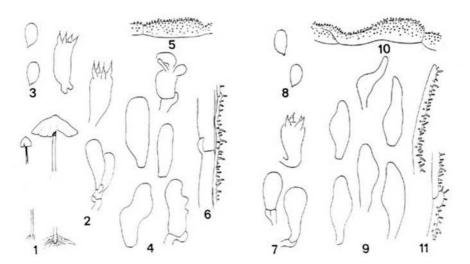
Growing on fallen leaves of Salix spec.

Collections examined. NORWAY. Vestfold, Tjøme, Hvasser, Sønstegård: 26 Oct. 1989, A. Aronsen A51/89 (L, no. 990.198-189); same locality: 4 Nov. 1989, A. Aronsen A66/89 (L, no. 990.198-251); same locality: 14 Oct. 1990, A. Aronsen A26/90 (holotype; L, no. 990.198-234).

The macroscopic description of the species has been made by the first author, at some points complemented by the second author's observations on the dried material, while the microscopic details are based on the second author's reexamination of the collections mentioned above.

Mycena terena is a member of section Polyadelphia Sing. ex Maas G. and has many features in common with the section's type species, M. polyadelpha (Lasch) Kühn., such as minute size, small number of lamellae, somewhat concave lamellar edge, very narrow stipe, relatively broad basidia, plump sterigmata, lack of pleurocystidia, wide hyphae of the pileipellis and their characteristic ornamentation of densely spaced, short excrescences and, finally, occurrence on decayed leaves of a deciduous tree. There is one character, however, which will necessitate an emendation of the sectional diagnosis and by which the species can at once be differentiated from other members of section Polyadelphia: the cheilocystidia are smooth. Although this may seem a fundamental difference, it should be remembered that the cheilocystidia of Mycena riparia Maas G. appear not nearly as densely diverticulate as their counterparts in M. polyadelpha or M. quercus-ilicis Kühn. (Maas Geesteranus, 1986), thus occupying an intermediate position.

An unusual character shared by *M. terena* and *M. albiceps* (Peck) Gilliam (Maas Geesteranus, I.c.) is their black stipe. The difference is that in the former species, the stipe, except for its whitish base, is entirely black in very young specimens, then gradually fades with age from the base upwards to grey or white although remaining black at the apex for some time longer. In *M. albiceps*, the stipe is black for the greater part but it is the apex which is paler, while the black colour seems to be retained even in aged specimens. The decisive difference separating *M. terena* from *M. albiceps* is in the cheilocystidia of the latter species. Owing to the scantiness of the type material of *M. albiceps* cheilo-



Figs. 1-6. Mycena terena (holotype, Aronsen A 26/90; L). 1. Habitus of young and old specimen (dried); 2. basidia; 3. spores; 4. cheilocystidia; 5. hypha of the pileipellis; 6. hypha of the cortical layer of the stipe. — Figs. 7-11. Mycena terena (Norway: Aronsen A 51/89; L). 7. Basidia; 8. spores; 9. cheilocystidia; 10. hypha of the pileipellis; 11. hyphae of the cortical layer of the stipe. (Fig. 1, × 10; all others, × 700.)

cystidia were not observed (Maas Geesteranus, 1986: 162), but unpublished illustrations executed by Dr D. E. Desjardin (San Francisco) show the cheilocystidia of *M. albiceps* to be apically covered with rather numerous, coarse, more or less curved excrescences.

If one were to judge *Mycena terena* by its cheilocystidia alone, the species could easily be mistaken for a member of section *Fragilipedes* (Fr.) Quél. However, there is not a single species of the *Fragilipedes* which combines such characters as extremely small size, distant lamellae (generally less than 10), and comparatively broad hyphae of the pileipellis that are very densely covered with wart-like excrescences.

#### ACKNOWLEDGEMENT

Appreciative acknowledgment is made to Dr D.E. Desjardin (San Francisco State University) who very kindly allowed the second author of the present paper the perusal of a part of one of his future publications.

#### REFERENCE

Maas Geesteranus, R.A. 1986. Conspectus of the Mycenas of the Northern Hemisphere-6. Sections Polyadelphia and Saetulipedes. Proc. K. Ned. Akad. Wet. (Ser. C) 89: 159-182.

#### PERSOONIA

Published by Rijksherbarium / Hortus Botanicus, Leiden Volume 15, Part 1, pp. 109-115 (1992)

# ERLÄUTERUNGEN ZU RAMARIA OBTUSISSIMA UND ZU RAMARIA SCHILDII

#### E. SCHILD

Borgo Treviso 177, I-31033 Castelfranco, Italy

Es wird festgestellt, daß Clavaria obtusissima Peck warzige Sporen hat. Das Taxon welches in der Monographie von Corner fälschlicherweise unter diesem Namen aufgeführt ist hat glatte Sporen; es wurde von Petersen unter dem Namen Ramaria schildii neu beschrieben und wird in dieser Arbeit emendiert.

Seit Jahren finde ich in den Fichtenwäldern hoch oberhalb des südlichen Brienzersee-Ufers eine recht groß werdende, wenn jung leuchtend gelbe *Ramaria*, deren Sporen in Baumwollblau betrachtet wie auch auf Rasterfotos absolut glatt sind (Abb. 1a, b).

Nach Corner (1950: 609) bestimmte ich diesen Pilz jeweils als Ramaria obtusissima, da ich die makroskopische Beschreibung einigermaßen passend fand, und die Sporen ebenfalls als glatt angegeben werden. Um sicher zu sein, beschaffte ich mir aber aus dem Herbar von Peck Typusmaterial von Clavaria obtusissima. An diesem konnte ich feststellen, daß die Sporen im Lichtmikroskop – wenn nur mit Wasser oder KOH-Lösung betrachtet – zwar glatt erscheinen (worauf sich wohl Corners Angabe gründen dürfte); in Baumwollblau lassen sich jedoch feine Riebelungen und Warzen erkennen, was auch auf Rasterfotos deutlich bestätigt wird (Abb. 1c). Somit war mir klar, daß meine glattsporigen Pilze aus Brienz nichts mit Pecks C. obtusissima zu tun haben können, dies umsomehr, da Peck (1913: 39) in seiner Diagnose über die Farbe nur "ochracei" schreibt. Weitere Nachforschungen überzeugten mich außerdem, daß es sich bei meinen Pilzfunden tatsächlich um dieselbe Art handelt, welche bei Corner unter dem falsch angewendeten Namen Ramaria obtusissima aufgeführt ist. Da mir in der einschlägigen Literatur, im besonderen für europäische Verhältnisse, keine andere gelbe Ramaria mit glatten Sporen bekannt ist, sah ich es als angezeigt, daß dieses Taxon neu beschrieben werden muß.

In einer Co-Arbeit mit Dr. J. Keller, Neuchâtel, der mir die schönen Rasterfotos der Sporen anfertigte, hatte ich vor, dieses glattsporige Taxon unter dem Namen "Ramaria leiospora" zu beschreiben. Inzwischen publizierte Petersen (1988: 229) diesen Pilz unter dem Namen Ramaria schildii Petersen, wobei seine Beschreibung offensichtlich nur auf einer einzigen Aufsammlung, derjenigen von Italien: "vic. Trento, 21.IX.1972, Coll. Gruppo G. Bresadola (TENN No. 36847)" basiert war.

Anhand dieser Aufsammlung schreibt Petersen zum Strunk: "... stipes slender (not more than 8 mm thick) rooting to some extent, fasciculate or with a very small point of union at the base, or solitary." Zu den Tramahyphen im Strunk schreibt er: "... stipe tramal hyphae hyaline, thin-walled, clamped ... and dendrohyphidia 1.6–2.0 µm diam."

Petersen meint, daß R. schildii wegen des dünnen, fast etwas büschellig aussehenden Stiels und den vorkommenden Dendrohyphidien eine, von 'R. leiospora' verschiedene Art sei, "denn meine Pilze hätten einen einfachen, dickeren Stiel und keine Dendrohyphi-

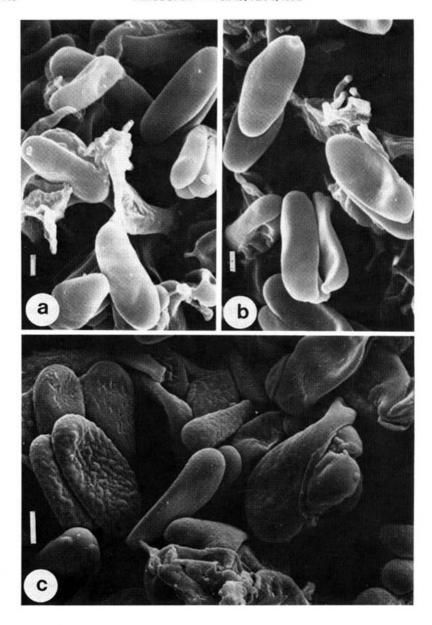


Abb. 1. Ramaria Sporen. a. R. schildii (Herb. Schild 778); b. R. schildii (Herb. Schild 1401); c. R. obtusissima (Holotypus, Herb. Peck, NYS). Strich = 2 µm.

dien, darum solle ich dieses Taxon unter dem von mir früher gewählten Namen, R. leiospora, beschreiben." Von diesen meinen Pilzfunden selbst gibt Petersen auch noch eine Beschreibung, diese unter der Bezeichnung 'Ramaria European taxon 1', wobei er hier unter 'Specimens examined' nun einige wenige Sammlungen angibt.

Zu Petersens Auffassung ist folgendes zu entgegnen. Da sich die Beschreibung von R. schildii nur auf eine einzige Aufsammlung stützt und die Beschreibung von 'Ramaria European taxon 1' offenbar auf nur wenige Sammlungen, ist die Variationsbreite der Makro- und Mikro-Charaktere wesentlich zu kurz gekommen. Besonders der Strunk, auch von nahe beieinander gewachsenen Fruchtkörpern, zeigt alle Übergänge, von schmächtig bis kurz und dick. In Anbetracht der von Petersen bei R. schildii beschriebenen Dendrohyphidien, ist nur zu erwähnen, daß zwei Aufsammlungen (aus Trento und Brienz), welche auf Grund des lang ausgezogenen, beziehungsweise auch fast büschelligen Stieles gut zu Petersens Beschreibung paßten, keine Dendrohyphidien festgestellt werden konnten. Somit dürfte es gerechtfertigt sein anzunehmen, daß das Vorhandensein von Dendrohyphidien nicht mit dem Makromerkmal Dünnstieligkeit zusammen hängt. Auch wurden bei keiner anderen Sammlung dieser glattsporigen Pilzart Dendrohyphidien beobachtet, was darauf hindeuten könnte, daß diesem Merkmal vielleicht nicht immer einen so großen Wert beigemessen werden muß. Im weiteren ist zu vermerken, daß zwischen den Sporen von R. schildii und denjenigen bei allen meinen Aufsammlungen kein Unterschied festzustellen ist. Jedenfalls gibt es keinen Grund nebst R. schildii noch eine zweite solche glattsporige Art neu zu beschreiben. Nur ist es hier angebracht die Beschreibung der Art zu emendieren. Anhand meiner reichlichen Aufsammlungen gebe ich folgende Beschreibung.

#### Ramaria schildii Petersen

Ramaria schildii Petersen, Mycologia 80 (1988) 229.

Falsch angewendeter Name: Ramaria obtusissima (Peck) Corner sensu Corner, Ann. Bot. Mem. 1 (1950) 609.

### Makroskopische Merkmale

Fruchtkörper 10-160(-180) mm hoch, 80-140 mm breit, reich verästelt (Abb. 2a). Strunk sehr unterschiedlich gestaltet; entweder einfach, allgemein zwischen 30-60 mm hoch, oben 20-35 mm dick dann oft lang ausgezogen und abwärts sich konisch verjüngend bis zuspitzend (charakteristisch), Basis bisweilen noch leicht knollig, oder auch aus mehreren, 5-15 mm dicken Individuen bestehend die unten miteinander verwachsen sind, sodaß der Strunk manchmal fast büschelig aussieht, meist glatt, seltener mit verkümmerten Seitenästchen (Abb. 2a). Farbe: abwärts schmutzig weiß, Basis mehr oder weniger deutlich mit weißem Mycelfilz behaftet, oben in die Farbe der Äste übergehend, auf Druck allgemein leicht bräunlich-weinbräunlich anlaufend, wenn alt oft ähnlich einer reifenden Clavariadelphus pistillaris besonders im oberen Teil, manchmal mit incarnaten, schmutzig weinrötlichen oder purpurnen Partien oder Flecken, vor allem an früheren Druckstellen oder wenn verletzt. Äste meist sehr dicht, daher bei jungen Pilzen oben oft blumenkohlartig gedrängt, dann sich streckend, unten (3-)5-13(-17) mm dick, aufwärts etwas divergierend seltener etwas parallel, aufrecht oder mit Tendenz zu schräger Wuchsform, aus etwa gleicher Höhe meist zwei oder drei mal geteilt, rundlich oder unregelmäßig, an oberen Ästen oft fast glatt, sonst zart runzelig (Lupe), stellenweise schwach längsgerillt

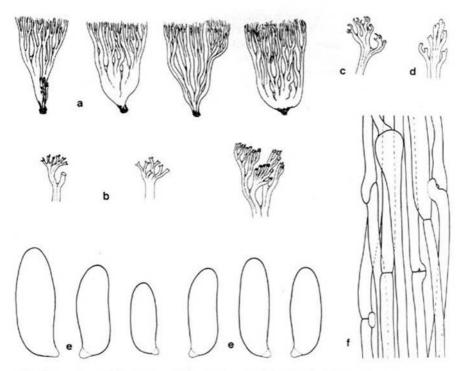


Abb. 2. Ramaria schildii. a. Skizzen der Fruchtkörper-Basis (verkl.); b-d. Endästchen-Spitzen. b. palmate, c. krallenartige, d. stumpfe Enden (verkl.); e. Sporen (× 2000); f. Hyphen der Ast-Trama (ca. × 1000).

besonders unterhalb den Astteilungen, manchmal jedoch ausgeprägt längsgerillt. Astwinkel gemischt; breit bis eng U-förmig, stumpfspitzig oder offenspitzig. Endästchen oft mit leicht palmaten, handförmig flach gedrückten Teilstellen (charakteristisch) (Abb. 2b). Enden-Spitzen sehr unterschiedlich gestaltet; wenn jung oft etwas zahnförmig, später meist mit zwei bis drei, 1–3 mm langen, bisweilen fast hühnerkrallenartig gestalteten Spitzen die an sich oft noch je zwei kurze, fransig dünne Spitzchen aufweisen (Lupe) (Abb. 2c), seltener mit stumpfen oder sogar plumpen Enden (Abb. 2d). Astfarbe an jungen Pilzen leuchtend hell primelgelb (nahe S. 290, 289, 319; Séguy, 1936); Spitzen noch intensiver, bei zunehmender Reife an Ästen mit einem Hauch ockergelb-crémeocker, Greif- und Legstellen werden später mehr oder weniger deutlich wässerig weinbräunlich, blaß incarnat, bräunlichpurpur oder bräunlichweinrötlich bis schmutzig weinrötlich (nahe S. 250, 249, 200, 199, 190, 189), alternde Pilze können auch gänzlich weinrosabräunlich werden (nahe S. 205, 204), oder von oben am Strunk bis in die mittleren Aste oft auch ähnlich einer reifenden Clavariadelphus pistillaris oder C. truncatus. Die Spitzen bleiben bei dieser Art meist bis ins Alter schön gelb, bisw. leicht grünlichgelb (charakteristisch!).

Fleisch bei feuchten Pilzen schmutzig weiß, zart wässerig marmoriert besonders im Strunk, unter dem Hymenium bisweilen leicht gelblich durchgefärbt, bei antrocknenden Pilzen crémeweiß, rahmweiß, unveränderlich, gegen die Spitzen weichbrüchig, sonst in Ästen Tendenz zu Längsspaltung da etwas faserig, meist nicht gelatinös.

Geruch aus mehreren, ungleich stark ausgeprägten Komponenten bestehend daher schwer definierbar, am ehesten vielleicht an Hypholoma fasciculare erinnernd (manchmal fast stechend) auch irgendwie staub- oder erdartig, entfernt bisweilen auch mit kampferartigen oder maggi-artigen Komponenten, seltener fast etwas angenehm-mild riechend.

Geschmack im Strunk oft korallenpilzartig mild, aufwärts etwas herb, gegen die Spitzen herb-bitterlich besonders im Alter.

Sporenpulver in Masse gelbocker.

## Makrochemische Reaktionen

Fe SO<sub>4</sub>: Hymenium allmählich schmutzig olivgrün bis grünspangrün. Fleisch langsam sehr blaß grünlich, dann oft leicht nachdunkelnd, bisweilen mit Hauch blaugrün.

KOH (10-20%): Hymenium schnell schmutzig bräunlich bis orangeockerlich, dann langsam satter schmutzig bräunlichorange-bräunlich. Fleisch schnell blaß schmutzig orangegelblich, dann vertieft.

Phenolliquefact: Hymenium sofort satter ockerlich oder schmutzig ockerlichbeige. Fleisch meist sofort ziemlich deutlich weinrosa dann vertieft.

Eisenchlorid: Hymenium sofort graugrün-blaugrün, dann nachdunkelnd. Fleisch nur blaß grau-blaugrünlich.

H<sub>2</sub>SO<sub>4</sub> (60%): Hymenium schnell blaß ockergelb, gelb- bis gelbgrünlich aufhellend, auf bereits weinbräunlichem Hymenium älterer Pilze eher orangegelblich-ockerorangelich reagierend. Fleisch nicht reagierend oder blaß olivgelblich bis oliv, dann vertieft.

# Mikroskopische Merkmale

Hymenium die Äste allseitig bedeckend, an den Spitzen etwas verkümmert, meist zwischen 75–90 μm dick, olivgrünlich. Subhymenium irregulär, etwa zwischen 15–35 μm dick, jedoch gegen die Tramahyphen sehr undeutlich abgegrenzt. Sporen glatt, (9,6-)  $10,4-16(-16,8) \times (3,5-)3,7-5,6$  μm (ohne Apiculus), im Schnitt  $13,2 \times 4,5$  μm, blaß meergrünlich bis olivgelblich, mit plasmatischem Inhalt und meist einem großen oder mehreren kleinen Tropfen, Tendenz zu zylindrischer Form dabei oft mit geradem bis eingedrücktem 'Rücken'; Sporenwand 0,2-0,3(-0,4) μm dick, stark cyanophil, Apiculus meist zwischen 0,7-1 μm lang (Abb. 2e). \*Basidien\* keulenförmig  $(38-)48-68(-75) \times (6,4-)7,2-12,3$  μm, olivgrünlich, Inhalt feintropfig bis körnig rauh, mit 4 (3) Sterigmen (3,6-)4,8-7,6(-8) μm lang, Basis mit Schnallen, cyanophil. Basidiolen meist dünner, sonst gleich.

Hyphen im ganzen Fruchtkörper da und dort mit Schnallen, hyalin, bisweilen vakuolär sonst Inhalt glatt, Membran blaß olivlich oder beigegrünlich, glatt, etwas cyanophil. Im Subhymenium irregulär, zwischen 2,5–5 μm dick, relativ kurzgliedrig, parallelwandig oder unregelmäßig, Membran zwischen 0,2–0,4 μm. In der Ast-Trama im allgemeinen 2,5–16,5 μm dick, nahe dem Subhymenium stellenweise fast regulär und allgemein dünner, gegen das Astinnere dicker, mehr oder weniger irregulär, Hyphenglieder parallel-

<sup>1)</sup> Alle Maße wurden in 'L4' nach Clémençon (1972: 49) ermittelt. Es möge erwähnt werden, daß von frischen Pilzen in purem Wasser gemessene Sporen ca. das gleiche brauchbare Resultat ergeben.

wandig bis unregelmäßig, oft auch langbauchig oder gegen Septen verjüngt, bei Septen etwas verengt bis leicht eingeschnürt (Abb. 2f). Membran in oberen Ästen zwischen 0,2–0,5 μm dick, in unteren Asten manchmal verdickt, einzeln bis 0,6–1,2 μm. Im Strunk: in der Rinde meist gedrängt-verflochten, oft relativ dünn (1,6–)2–6 μm, mit 0,3–0,5 μm dicken, manchmal gelblichen Membranen dann in Massen blaß ockergelblich erscheinend, gegen innen hyalin, 2,5–12(–16) μm dick, irregulär, Membran 0,4–1,6 μm, im übrigen sonst wie in Ästen. Sowohl an den Hyphen des Strunkes wie in Ästen oft ampullenförmige Anschwellungen mit meistens verdickter Membran und ornamentierter Innenwand. Besonders oben im Strunk und in unteren Ästen manchmal mit zerstreut herumliegenden bizarr-polymorphen bis nadelförmigen Kristallkörper. Bisweilen an gewissen Stellen im Fleisch mit etwas gelatinösen Elementen. Basisfilz-Hyphen hyalin, öfters mit Schnallen, zwischen 1,5–8 μm dick, Membran 0,3–0,6 μm, bisweilen leicht incrustiert, bei Septen manchmal mit ampullenförmigen Anschwellungen bis etwa 11 μm, mit oder ohne Fortführung der Hyphen. Oleiferen in einigen Fruchtkörpern spärlich, in anderen dagegen mäßig vertreten, 2,5–5 μm dick, an kopfigen Enden bis 8–10(–25) μm, stark cyanophil.

Standort. Diese Art ist mir bis jetzt nur aus dem montanen Nadelwald in etwa 1000–1350 m Höhe bekannt, wobei in der Nähe auch einzelne eingestreute Laubbäume und Gebüsche vorkommen können, jedoch immer unter *Picea* stehend, meist zwischen Moos, Gras und Kräutern im Nadelhumus, gerne an Waldrändern oder in Waldlichtungen die gegen Norden gerichtet sind.

## Untersuchtes Material

SCHWEIZ: Bauwald bei Brienz, 8. Aug. 1971, E. Schild & K. Kehrli (Herb. Schild 286). Einsiedeln, Sept. 1972, E. Schild & R.H. Petersen (Herb. Schild 541). Tiefentalgebiet bei Brienz, 31. Juli 1973, E. Schild (Herb. Schild 642). Bauwald bei Brienz, 2. Sept. 1973, E. Schild & K. Kehrli (Herb. Schild 673). Tiefental Brienz, 11. Aug. 1974, E. Schild & K. Kehrli (Herb. Schild 778). Tiefental Brienz, Aug. 1976, E. Schild (Herb. Schild 1040). Scharnachtal-Gebiet, Sept. 1979, W. Wäfler (Herb. Schild 1304). Scharnachtal, Flühmäder, 7. Aug. 1982, W. Wäfler (Herb. Schild 1547). Gau-Gebiet bei Brienz, 31. Aug. 1984, E. Schild & W. Wäfler (Herb. Schild 1637). — TSCHECHOSLOWAKEI: Polana prope Detva, 26. Aug. 1951, A. Pilat (als Ramaria obtusissima, Herb. Schild 42). — FRANKREICH: Piccetum du Jura, Forêt de la Faye, 29. Aug. 1973, H. Romagnesi (Herb. Schild 751). — ITALIA: Trentogebiet, 13. Sept. 1981, B. Cetto (Ausstellung) (Herb. Schild 1401). — DEUTSCHLAND: am 'Mostleberg', 9. Sept. 1976, A. Runge (Herb. Schild 1057), Berchtesgadener Alpen, 13. Aug. 1982, Schmid-Heckel (als R. aurea, Herb. Schild 1650). Bayern, Kaltenbrunnen, 16. Sept. 1973, A. Einhellinger (als R. flava; Herb. Schild 1656). — USA: West Roxbury, Massachusetts, Sept. 1911, Ms. A. Hibbark (Holotypus von Clavaria obtusissima, Herb. C.H. Peck, NYS; auch Fragmente im Herb. Schild 1165).

Leute die sich nicht eigens mit dieser Gattung befassen, können jüngere, noch gelbe Fruchtkörper von R. schildii mit allen anderen gelben Ramarien verwechseln.

Außer R. schildii deren Sporen auch im RE-Mikroskop glatt sind, ist mir in Zentraleuropa noch eine große, gelbe Ramaria-Art aus der Flava-Gruppe bekannt deren Sporen im Lichtmikroskop – wenn nur im Wasser oder KOH-Lösung betrachtet – mehr oder weniger glatt erscheinen und auch fast dieselbe Größe haben. In Baumwollblau aber ist deutlich eine warzige Ornamentation zu erkennen. Es handelt sich hier um die neutypisierte und wiederbeschriebene Art, Ramaria flava (Schaeff.) Quél. (Schild, 1991). Alle übrigen gelben Ramarien in Europa haben warzige Sporen was bereits in purem Wasser betrachtet sichtbar ist. Zwei weitere, aus Europa beschriebene Arten mit glatten Sporen gehören nicht zur Flava-Gruppe. Es ist dies R. gypsea Schild (1982: 33) sie ist gänzlich gibsweiß und R. terrea Schild (1990: 134) diese ist erdbraun.

#### VERDANKUNG

Folgenden Herren möchte ich meinen Dank aussprechen: Prof. R.H. Petersen in Tennessee, für die Besorgung und Zusendung des Ex-Typus-Materials von *Clavaria obtusissima* aus dem Herbar Peck; Dr. J. Keller, Neuchâtel, und J. Christan, München, für die jeweiligen elektromikroskopischen Aufnahmen der Sporen; K. Kehrli, Schwanden bei Brienz, für seine stets uneigennützige Bereitschaft mich an Fundorte zu führen; und W. Wäfler, Kehrsatz bei Bern, für Exkursionsführungen und Zusendung von Frischmaterial

## Summary

It has been ascertained that Clavaria obtusissima Peck has warted spores. The taxon wrongly presented under this name in Corner's Monograph has smooth spores; it was newly described by Petersen as Ramaria schildii and is emended here.

#### LITERATUR

Clémençon, H. 1972. Zwei verbesserte Präparierlösungen für die mikroskopische Untersuchung. Z. Pilzk. 38: 49-53.

Corner, E.J.H. 1950. A monograph of Clavaria and allied genera. Ann. Bot. Mem. 1.

Peck, C.H. 1913. Report of the state botanist 1912. Bull. N.Y. St. Mus. 167: 5-137.

Petersen, R.H. 1988. Contribution toward a monograph of Ramaria. VII. New taxa and miscellany. Mycologia 80: 223-234.

Schild, E. 1982. Studie über Ramarien. Schweiz. Z. Pilzk., Sondernummer 123: 1-48.

Schild, E. 1990. Ramaria-Studien. Z. Mykol. 56: 1-216.

Schild, E. 1991. Zur Typisierung von Ramaria flava (Schaeff.) Quélet und Ramaria sanguinea (Pers.) Quélet. Z. Mykol. 57: 229-248.

Séguy, E. 1936. Code universel des couleurs. Paris.

### PERSOONIA

Published by Rijksherbarium / Hortus Botanicus, Leiden Volume 15, Part 1, pp. 117-121 (1992)

# PSILOCIN, PSILOCYBIN, SEROTONIN AND UREA IN PANAEOLUS CYANESCENS FROM VARIOUS ORIGIN

#### T. STIJVE

Quality Assurance Department of Nestec Ltd, Avenue Nestlé 55, 1800 Vevey, Switzerland

The occurrence of tryptamine derivatives and urea in *Panaeolus cyanescens*, also known as *Copelandia cyanescens*, from Australia, Hawaii and Thailand was investigated. All 70 collections contained psilocin, serotonin and urea. Those from Hawaii were also relatively rich in psilocybin, whereas the species from Australia and Thailand were virtually exempt of this compound. Many collections also contained detectable amounts of precursors as tryptophan, tryptamine and baeocystin, but 5-hydroxytryptophan – widely encountered in many other Panaeoloideae – was found to be absent in all samples. The role of these 4- and 5-hydroxylated tryptamine derivatives in the metabolism of the fungus and their possible chemotaxonomic significance is briefly discussed. Volunteers ingesting samples of *Panaeolus cyanescens* reported a stronger psychotropic effect than that experienced with the same amount of *Psilocybe semilanceata*.

Panaeolus cyanescens (Bk. & Br.) Sacc., also known as Copelandia cyanescens (Bk. & Br.) Sing., is a primarily coprophilic Panaeolus of average size encountered in the tropics and neotropics of both hemispheres (Singer, 1960). It is characterised by the bluestaining reaction of its flesh on exposure to air, and by the fact that the gills possess honey-coloured ventricose metuloids (Gerhardt, 1987; Young, 1989).

Panaeolus cyanescens was already recognised as a hallucinogenic mushroom about 25 years ago (Heim, 1967). Its recreational use on the Samoan islands has been reported (Cox, 1981), and, more recently, also on the island of Bali in Indonesia, and in the Hawaiian archipelago (Allen & Merlin, 1989).

The mushroom is reported to contain both psilocin and psilocybin in unusually high quantities (Schultes & Hoffmann, 1980), but published analytical data are few. Panaeoloideae contain appreciable levels of both urea and serotonin, 5-hydroxytryptamin (Stijve, 1985), and it has been suggested that their presence could have chemotaxonomic significance, since these compounds are absent in other Coprinaceae, such as *Psathyrella* and *Coprinus* (Stijve, 1987). It was therefore considered of interest to check whether *P. cyanescens* would share these chemical characteristics.

Since the chromatographic techniques used for the assay of urea, serotonin and their precursors permit the simultaneous determination of psilocin and psilocybin, the possible fluctuation in the concentrations of the latter hallucinogens according to geographical origin could also be monitored. The results of the investigation are reported in this paper.

## MATERIAL AND METHODS

The available P. cyanescens collections had been gathered and identified by competent mycologists. Material from Queensland, Australia was supplied by Dr. A. Young, Black

butt. Three abundant collections from Hawaii were sent by John Allen, who was also able to provide us with some dried carpophores from the island Koh Samui in Thailand. Carpophores from the three locations were sent to Dr. E. Gerhardt in Berlin, who confirmed their identity as *P. cyanescens* (Bk. & Br.) Sacc. The material was received in airdried condition and stored at 5°C in air-tight containers. Considering that some of the possible present compounds such as psilocin are sensitive to oxygen and enzymatic activity, the fungi were analyzed soon after receipt. Just prior to extraction, the individual carpophores were weighed and ground to a fine powder. When large, a single fruit-body was taken for analysis, when small, several specimens. Methanol extraction and subsequent TLC and HPLC analyses were performed as reported earlier (Stijve, 1985).

Psychotropic effects of the dried powdered material from Hawaii (of which an ample supply was available) were tested by ingestion of 1 g amounts of the finely ground material. Among the four volunteers involved in this experiment all had previous experience with hallucinogenic mushrooms, mainly *Psilocybe semilanceata*.

### RESULTS

The results of the chromatography analyses of the individual carpophores are listed in Table I. In all samples varying amounts of psilocin, serotonin and urea were found using two chromatographic systems.

There was no indication of significant degradation of the material having occurred between collection and analysis. In fact, the most labile compound, psilocin, was invariably found to be accompanied by its primary oxidation product observed as a greenish spot just below psilocin during TLC in the cellulose/BAW system (Stijve et al., 1984) – but its concentration rarely exceeded 0.02%, which added little to the total psilocin content. The limit of detection for psilocybin and its precursors tryptophan, tryptamine, and baeocystin fluctuated somewhat with the sensitivity of the chromatography systems, but it was always adequate. No other tryptamine derivatives such as methylserotonin or bufotenin were observed, although the limit of detection was often better than 0.01%.

During the self-experiments, taking 1 g of the powdered pooled Hawaiian carpophores (containing 0.6 percent psilocin and 0.2 percent psilocybin) produced a most powerful psychotropic effect. The initial symptoms were felt within 20 minutes: accentuation of visual patterns, slight euphoria and intensifying of colour perception. These effects became stronger for about 4 hours, but since the session was held indoors, the participants experienced more a soulsearching trip with much introspection than a strongly visual adventure.

# DISCUSSION

The results of the investigation (Table I) indicate that the tryptamine derivative composition of *P. cyanescens* varies appreciably according to origin. The dimensions and thus the weight of the carpophores fluctuated widely, even within a collection. However, a small specimen weighing about 20 mg is not necessarily in the primordial stage. It can be adult and sporulating as witnessed by an often high urea content. Not surprisingly, all carpophores contained psilocin, the predominant tryptamine derivative. Collections from Australia and Thailand were found virtually exempt of psilocybin, which can perhaps be

Table. I. Tryptamine derivatives and urea in Panaeolus cyanescens from	m various origin.
--	-------------------

Collections	Australia Queensland 1989	Thailand Koh Samui Isl. 1990	Hawaii Sept. 1989	Hawaii Dec. 1989	Hawaii Coral Kingdom Oahu, July 1990
	N = 11	N = 5	N = 13	N = 14	N = 27
Weight of carpophores (mg)	65-340 (125)	-	24-260 (154)	15-300 (103)	6-250 (56)
Psilocin	0.025-0.71 (0.31)	0.40-1.05 (0.95)	0.055-0.33 (0.17)	0.38-1.30 (0.71)	0.04-0.60 (0.26)
Psilocybin	< 0.012 - 0.04	< 0.025	0.03-0.28 (0.16)	0.07-0.44 (0.19)	0.01-0.73 (0.16)
Baeocystin	< 0.01	< 0.025	<0.01-0.025 (0.020)	<0.005-0.026 (0.016)	< 0.005 – 0.035 (0.015)
Tryptophan	< 0.01-0.03	< 0.01	0.006-0.014 (0.011)	< 0.01-0.02	< 0.01
Serotonin	0.023-0.45 (0.17)	0.026-0.038 (0.031)	0.025-0.11 (0.062)	0.02-0.24 (0.064)	0.005-0.10 (0.035)
Tryptamine	< 0.004 - 0.02	0.002-0.008 (0.005)	< 0.005	< 0.005	
Urea	0.20-4.5 (1.65)	1.8-3.3 (2.66)	0.40-2.50 (1.33)	0.53-1.87 (1.20)	0.07-3.0 (1.97)
All values in mg	kg on dry weigh	nt. Mean values i	n brackets.		

explained by a lack of a phosphorylating enzyme. On the other hand, contrary to our observations for the Hawaiian material, we found often small but detectable amounts of tryptamine which may be a precursor both in the biosynthesis of psilocin and serotonin. Interestingly, the alternate serotonin precursor, 5-hydroxytryptophan, which is present in substantial amounts in most Panaeoloideae, was found to lack completely in *P. cyanescens*. Although we paid special attention to its possible presence it was even found absent in the stipes of the carpophores in which it is usually concentrated (Stijve, 1987). The detection limit was 0.005% or better.

When analyzing separately stipe and pileus it was found that psilocin was about equally distributed between both parts of the carpophore. Serotonin was found exclusively in the pileus, but the stipe contained  $3 \times \text{more}$  psilocybin than the pileus. That most of the urea (80-90%) is concentrated in the cap was already known (Stijve, 1987).

The collections rich in psilocybin often contained detectable amounts of the mono-methyl analogue baeocystin, indicating that in the biosynthesis phosphorylation precedes methy-

lation, although to only a modest extent. The specificity of the biosynthesis routes for psilocin and serotonin is highlighted by the total absence of bufotenin (dimethylserotonin), the 5-substituted analogue of psilocin. Similarly, no phosphate ester of serotonin was observed either.

It has been suggested (Stijve, 1987) that the biosynthesis of both urea and serotonin could be ways of neutralising toxic ammonia which is invariably present in the nitrogenrich substrate of the Panaeoloideae. It was already noticed that some of the species low in urea had often, although not always, a higher than average serotonin content. It could well be that psilocin and psilocybin are similar waste products. Not seldom a high concentration of the said compounds in *P. cyanescens* was accompanied by a low serotonin content in the individual carpophores. Even the average values point in that direction (Table I). For example, the Australian collections are low or average in psilocin, but definitely higher in serotonin than the psilocin-rich material from Thailand and Hawaii.

It is not unthinkable that the ability to biosynthesize psilocin in a number of *Panaeolus* is a consequence of a genetical accident: initially, these members of the genus probably produced more or less important quantities of serotonin until a mutation conferred the ability to produce 4-hydroxylated tryptamines in addition to the 5-substituted ones. So far, no other genus or species has been found to accomplish this feat.

The ability to produce both urea and serotonin is even shared by members of the genus that grow on rotten wood, an unusual substrate for *Panaeolus*. We found 0.18 percent serotonin, 0.05 percent 5-hydroxytryptophan and 0.5 percent urea in a collection of *P. bernicii* Young, only known from the Bunya pine rainforest in Queensland, Australia, where it grows gregarious to subcaespitose on very rotten wood (Young, 1989). This remarkable species did not contain any psilocin or psilocybin.

Singer (1986) created the genus *Copelandia* for those Panaeoloideae that have characteristic coloured metuloid cystidia and blueing tissues. Gerhardt (1987) has rightly pleaded for a broader generic concept, since the metuloids are also found in many Inocybes, and species with blue- or blue green-staining flesh occur in many genera (Stijve & Kuyper, 1985).

The results presented in this paper show that *P. cyanescens* has also the chemical characteristics (urea, serotonin) of a true *Panaeolus*. In addition, recent studies on the bioconcentration of metals indicate that *P. cyanescens* shares the ability to accumulate manganese with the other members of the genus. No other dark brown to black spored agarics seem to have a marked affinity for this metal (Stijve, 1990).

The four volunteers testing the psychotropic effects of *P. cyanescens* agreed that the 1 g portion ingested proved it to be more potent than *Psilocybe semilanceata*. The effects were felt sooner after ingestion, but the duration of the trip was shorter, which can be explained by the high psilocin content of the *Panaeolus*. Unlike psilocybin, which has first to be hydrolysed, psilocin works directly on the neurotransmitter receptors. (*Psilocybe semilanceata* contains much psilocybin, but no psilocin.) No disagreeable side-effects were noted. Initially, one person got slightly nauseous when told that the fungi had been growing on cow dung, but she recovered real soon and later qualified the experience as 'rewarding'.

#### ACKNOWLEDGEMENTS

The author thanks John W. Allen (University of Hawaii, in Honolulu), Anthony Young (Blackbutt, Queensland, Australia) and Dr. Ewald Gerhardt (Botanisches Museum, Berlin, Dahlem, Germany) for either supplying *Panaeolus* collections or checking botanical identity.

Thanks are also due to Beowulf Glutzenbaum, Heinrich Hodenthaler, Cécile Tire-Boudin and Héléne la Main for participating in the self-experiments; and to Karima Ziaoullah for her help in preparing the manuscript.

### REFERENCES

- Allen, J.W. & M.D. Merlin. 1989. Copelandia and other psychoactive fungi in Hawaii. Newsl. Hawaiian bot. Soc. 28 (2): 27–30.
- Cox, P. 1981. Use of a hallucinogenic mushroom, Copelandia cyanescens. Samoa. J. of Ethnopharmacology 4: 115–116.
- Gerhardt, E. 1987. Panaeolus cyanescens (Bk. & Br.) Sacc. und Panaeolus antillarum (Fr.) Dennis, zwei Adventivarten in Mitteleuropa. Beitr. Kenntn. Pilze Mitteleur. III: 223-227.
- Heim, R. 1967. Nouvelles investigations sur les champignons hallucinogènes. Eds. Mus. nat. Hist. nat. Paris.
- Schultes, R. & A. Hoffmann. 1980. The botany and chemistry of hallucinogens, 2nd ed. Charles C. Thomas, Springfield.
- Singer, R. 1960. Hongos psicotropicos. Lilloa 30: 124-126.
- Singer, R. 1986. The Agaricales in Modern Taxonomy, 4th. ed. Koenigstein.
- Stijve, T. 1985. Een chemische verkenning van het geslacht Panaeolus. Coolia 28 (4): 81-89.
- Stijve, T. 1987. Vorkommen von Serotonin, Psilocybin und Harnstoff in Panaeoloideae. Beitr. Kenntn. Pilze Mitteleur. III: 229-234.
- Stijve, T. 1990. Unpublished results.
- Stijve, T., C. Hischenhuber & D. Ashley. 1984. Occurrence of 5-hydroxylated indole derivatives in Panaeolina foenisecii (Fries) Kühner from various origin. Z. Mykol. 50 (2): 361–368.
- Stijve, T. & Th.W. Kuyper. 1985. Occurrence of psilocybin in various higher fungi from several European countries. Planta Medica 5: 385–387.
- Young, A.M. 1989. The Panaeoloideae (Fungi, Basidiomycetes) of Australia. Aust. Syst. Bot. 2: 75–97.

## PERSOONIA

Published by Rijksherbarium / Hortus Botanicus, Leiden Volume 15, Part 1, pp. 123-125 (1992)

# A NEW SPECIES OF ENTOLOMA SUBGENUS POUZARELLA FROM SPAIN

MACHIEL E. NOORDELOOS1, MANUEL TABARÉS2 & AUGUST ROCABRUNA3

A new species of Entoloma subgen. Pouzarella sect. Dysthales is described, viz. E. pseudodysthales Noordel., Tabarés & Rocabruna from Spain, distinguished by the fetid smell, large, broad spores, and lack of cheilocystidia.

The subgenus *Pouzarella* is a rather distinctive group within the large genus *Entoloma*, and is characterized by the usually small, mycenoid habit of the fruit-bodies, distinctive long, encrusted and often setiform hairs on pileus and stipe, the nodulose spores, clampless hyphae, and rather well differentiated trichodermal pileipellis with encrusted, and sometimes also intracellular pigments (Noordeloos, 1979, 1987). In Europe the subgenus *Pouzarella* is represented with two sections, viz. section *Dysthales*, with six species, and section *Versatilia* with three species only.

The two junior authors, exploring the mycoflora of Catalonia, Spain, recently came across a species from section *Dysthales* from coastal sand dunes, that could not be named with the existing literature. It comes close to *Entoloma dysthales* (Peck) Sacc. from which it differs not only by the lack of cheilocystidia, but also by its rather broad spores and fetid smell. *Entoloma dysthales* f. *acystidiosum* Noordel. originally decribed from Portugal, and since then known from various other places in Europe, including Scandinavia, differs by having differently shaped spores with a length/width ratio (Q) ranging from 1.6–1.9, and the lack of a distinct smell. *Pouzarella foetida* Mazzer, described from North America, also has a fetid smell, though rather weak, but this taxon has spores and cystidia similar to *Entoloma dysthales*, and is considered a variant of that species (Noordeloos, 1988). Concluding this we think that an undescribed *Entoloma* species is involved, that is described below.

# Entoloma pseudodysthales Noordel., Tabarés & Rocabruna, spec. nov. - Fig. 1

Pilcus 3–10 mm latus, 5–20 mm altus, conicus, haud expansus, haud hygrophanus, haud translucidostriatus, obscure brunneus, toto hirsutus vel subsquamulosus, margine juventute fibrillosus. Lamellae distantes, adnatae, obscure brunneae. Stipes  $10-30\times1-3$  mm, pilco concolorus, fibrillosus. Odore foetulentus. Sporae  $14-21\times(10-)12-16$  µm, Q=1.2-1.4, noduloso-angulatae. Basidia 4-sporigera, efibulata. Cystidia desunt. Pileipellis cutis vel trichoderma hyphis septatis, 8-20 µm latis pigmentis incrustantibus. Stipitipellis similis. Fibulae desunt. Habitat ad terram in silvis mediterraneis (*Pinus halepensis, Pistacia lentiscus, Juniperus phoenicea*). — Holotypus: *M. Tabarés*, 31.I.1988, 'Playa Larga, Tarragona, Spain' (L).

<sup>1)</sup> Rijksherbarium / Hortus Botanicus, P.O. Box 9514, 2300 RA Leiden, The Netherlands.

<sup>2)</sup> Avda Coll del Portell, 44 bajos 6a, Barcelona, 08024 Spain.

<sup>3)</sup> Gran Via, III-Atic 1 a, Premià de Mar, Barcelona, Spain.

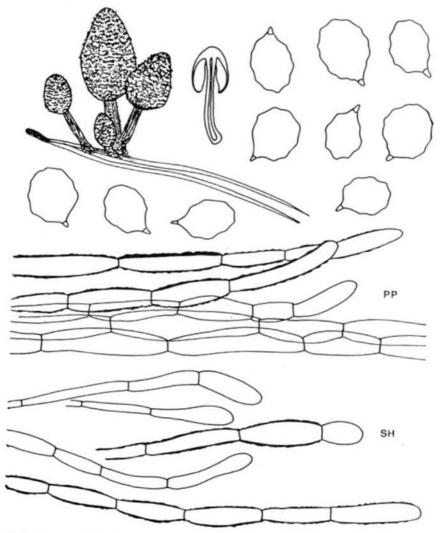


Fig. 1. Entoloma pseudodysthales. Habit ( $\times$  1), spores ( $\times$  2000), pileipellis (PP;  $\times$  500), and hairs of stipe (SH,  $\times$  500). All figures from holotype.

Pileus 3–10 mm broad, 5–20 mm high, conical, not expanding, not hygrophanous, not translucently striate, dark brown, entirely hirsute to subsquamulose, margin when young fringed with fine fibrils. Lamellae distant, adnate, 1–2 mm wide, dark brown. Stipe  $10-30\times1-3$  mm, slightly broadened towards base, dark brown concolorous with pileus, entirely covered with fine white fibils. Smell strong, like a mixture of garlic and rotten cabbage (reminiscent of the smell of *Collybia impudica*).

Spores  $14-21\times(10-)12-16~\mu m$ , Q=1.2-1.4, average Q=1.3, many-angled nodulose in side-view with rather thick walls, brownish in water. Basidia  $30-60\times12-17~\mu m$ , 4-spored, clampless. Lamella edge fertile. Cheilocystidia and pleurocystidia absent. Hymenophoral trama regular, made up of long, cylindrical elements with encrusted walls. Pileipellis a cutis with transitions to a trichoderm, made up of long, septate hyphae, gradually tapering towards end,  $8-20~\mu m$  wide. Pigment coarsely encrusting all parts of pileipellis and pileitrama. Stipitipellis a cutis with transitions to a trichoderm, made up of long, multiseptate hairs,  $7-16~\mu m$  wide with brown, coarsely encrusted walls. Clamp-connections absent from all tissues.

Habitat. In forest of Pinus halepensis, Pistacia lentiscus and Juniperus phoenicea on sandy soil in coastal dunes.

Collection examined. Spain, Tarragona, Playa Larga, 31.I.1988, M. Tabarés (holotype, L).

#### REFERENCES

Noordeloos, M.E. 1979. Entoloma subgenus Pouzaromyces emend. in Europe. Persoonia 10: 207–243. Noordeloos, M.E. 1987. Entoloma in Europe. Beih. Nova Hedwigia 91.

Noordeloos, M.E. 1988. Entoloma in North America. Cryptogamic Studies, vol. 2. Fischer Verlag, Stuttgart.

#### PERSOONIA

Published by Rijksherbarium / Hortus Botanicus, Leiden Volume 15, Part 1, pp. 126-128 (1992)

# BOOKS RECEIVED BY THE RIJKSHERBARIUM LIBRARY

R. Agerer. Colour Atlas of Ectomycorrhizae. Issue 5. (Einhorn Verlag, P.O. Box 1280, D-7070 Schwäbisch-Gmünd, Germany. 1991.) 16 pls. Price: DM 52.80.

The fifth issue of this series comprises extended keys for the determination of 148 species of Ectomycorrhizae (Abies 1, Betula 17, Carpinus 1, Fagus 31, Larix 10, Picea 51, Pinus 33, Pseudotsuga 4, and Quercus 2). The glossary, synoptic tables, literature as well as proposals for the arrangement of the plates are updated. Volume 5 comprises 16 plates of ectomycorrhiza with photo in colour, half tone photographs showing diagnostic details, and extensive legends. This series is an excellent piece of work and recommended to all working with ectomycorrhizae.

M. Bon. Flore mycologique d'Europe. 2. Les Tricholomes et Ressemblants. (Docum. Mycol. Mémoirs hors série 2, Lille. 1991.) Pp. 163, numerous text-figs, 5 col. pls. Price: FF 130.-.

This publication contains updated keys to the genera of the tribes Tricholomoideae and Leucopaxilloideae (*Tricholoma*, *Callistosporium*, *Tricholomopsis*, *Porpoloma*, *Floccularia*, *Leucopaxillus*, and *Melanoleuca*), following the concept of Bon's so-called monographic keys, that contain much information on macro- and microscopical characters and relevant literature. Numerous line-drawings of microscopical details are given and the coloured plates give good pictures of 40 species.

Cuadernos de Trabajo de Flora Micologica Iberica.

This series, edited and distributed by the Botanical Garden, Madrid, is published in the scope of the Flora Micologica Iberica project. The aim of the first five volumes is to present the information hiding in various databases that form the backbone of the project. The following issues have been received:

Vol. 1 – F. Pando , M. Duenas, C. Lado & M.T. Telleria (Eds.). Información bibliográfica. I. Espana peninsular e Islas Baleares. (Madrid. 1990.) Pp. 156. Price: 1000 Ptas.

This booklet lists 2281 publications dealing with fungi from mainland Spain and the Balearic Islands. Fungi pathogenic to Man and Animals are excluded. Included are necrologies of Mycologists who contributed to the knowledge of the mycoflora of Spain.

Vol. 2 – F. Pando. Manual de las bases de datos de Flora Micologica Iberica. (Madrid. 1991.) Pp. 67. Price: 750 Ptas.

This issue treats the criteria used to create the databases for the Flora Micologica Iberica project.

- Vol. 3 M.T. Telleria (Ed.). Bases corológicas de Flora Micologica Iberica, 1–132. (Madrid. 1991.) Pp. 160. Price: 1000 Ptas.
  - This is the first issue dealing with chorological data of fungi that have been found fructificating in the Iberian Peninsula. From the Aphyllophorales 132 taxa are listed with the localities where they have been found, and references to herbaria and literature.
- Vol. 4. M. T. Telleria (Ed.). Bases corologicas de Flora Micologica Iberica, 133–249. (Madrid. 1992.) Pp. 208. Price: 1000 Ptas. This is a continuation of volume 3, treating 116 species of the Aphyllophorales.
- Value of the second of volume 5, deating 110 species of the Aphyliopholates.
- Vol. 5. J. Cardoso & I. Melo. Informação bibliográfica. II. Portugal. Price: 1000 Ptas. This part, similar in concept to volume 1 of this series, contains more than 1000 bibliographic references referring to Portugal.
- G. Gulden & E.W. Hanssen. Distribution and ecology of stipitate hydraceous fungi in Norway with special reference to the question of decline. (Sommerfeltia 13, Botanical Garden and Museum, University of Oslo, Trondheimsveien 23B, N-0562 Oslo 5, Norway. 1992.) Pp. 58, numerous tables, distr. maps. Price: NOK 110.

Twenty-eight species of stipitate hydnaceous fungi (Auriscalpium, Bankera, Hydnellum, Hydnum, Phellodon, and Sarcodon) are treated in this volume. Data from about 1200 collections deposited in Norwegian herbaria, supplemented with personal observations and literature sources form the base of this study. Distribution maps and information on the ecology of each species is given. The species are ranged in five groups according to their distribution pattern: southern coastal species, southern species, south-eastern species, eastern species, and ubiquitous species. An evaluation of the frequency of the species through the decades back to 1950 is presented. Contrary to the situation in western and central Europe, there is no strong evidence for decline of these fungi. However, three Hydnellum species, viz. H. aurantiacum, H. peckii, and H. suaveolens seem to be less frequently observed in recent times.

G.J. Krieglsteiner. Verbreitungsatlas der Groszpilze Deutschlands (West). Band 1. Ständerpilze, Teil A. Nichtblätterpilze, Teil B. Blätterpilze. (Verlag Eugen Ulmer, Wollgrasweg 41, D-7000 Stuttgart 70, Hohenheim, Germany. 1991.). Pp. 1016, 3511 distr. maps in full colour. Price: DM 98,-.

Under the supervision of the author, numerous mycologists, mainly amateurs, in west Germany started with a mapping program of higher fungi, on a scale of 1:25.000. Three million data were gathered and resulted in distribution maps of 3511 taxa of higher basidiomycetes (Agaricales s.l. and Aphyllophorales). The maps give the distributional data that can be correlated with geography and altitude. No comments on distribution patterns and/or ecology are given.

M. Moberg & J. Holmåsen. Flechten von Nord- und Mitteleuropa. Ein Bestimmungsbuch. Aus dem Schwedischen übersetzt von Ute Jülich. (Gustav Fischer Verlag, Stuttgart, Jena, New York. 1992.) Pp. 237, numerous black-and-white illustr., 324 col. pls. (13 x 23.5 cm). Price: DM 78.-. The text part includes a Foreword, Table of Contents, Introduction, chapters on the construction of lichens, propagation, growth, ecology and distribution, chemistry, practical use, collecting and preservation. Then follow a list of the genera treated and 31 pages of generic descriptions of fruticose and foliose lichens and, for some of the genera, keys to the species. The coloured plates constitute the central, and most important, part of the book which is terminated by a key to lichen genera and an Index.

It is a pleasure for the reviewer to announce this book, even though a few comments cannot be avoided.

The key to the lichen genera (Bestimmungsschlüssel der Flechtengattungen, p. 223) would more sensibly have been placed before the generic descriptions (Gattungsbeschreibungen ..., p. 34). Reference to the pages where the species are keyed out would have been helpful. Similarly, rapid use of the book would have been facilitated if the species mentioned and their relevant illustrations are cross-referenced. It is rather confusing that the generic descriptions follow a systematic sequence, whereas in the index (p. 226) they are given in alphabetic order, again without page indication. Another source of confusion is that species like *Melanelia acetabulum* (p. 40) and *M. disjuncta* (p. 41) are illustrated with the legends *Parmelia acetabulum* (p. 86) and *P. disjuncta* (p. 89). The lichen called *Cornicularia aculeata* (p. 223) is illustrated as *Coelocaulon aculeatum* (p. 79).

The majority of the lichen illustrations are good to very good. Quite a number are excellent, e.g. *Hypogymnia bitteriana* (p. 84), *Aspicilia cenotea* (p. 110), *Cladonia cenotea* (p. 140). In some pictures, however, magnification is insufficient to show much detail of the marginal lobes of the thallus, e.g. *Parmelia glabratula* (p. 90) and *P. taractica* (p. 96).

All told, this book is certain to prove its usefulness, and the price is no serious obstacle.

R.A. Maas Geesteranus

R.E. Tulloss, C.L. Ovrebo & R.E. Halling. Studies on Amanita (Amanitaceae) from Andean Colombia. (Memoirs of the New York Botanical Garden 66. 1992.) Pp. 46, 28 text-figs., 8 black-and-white photogr. Price: US\$ 19.90.

With the publication of descriptions of 12 species and varieties of Amanita, 11 of which are new, from Colombia, mainly from Quercus humboldtii forests, the number of Amanitas known from that country is raised from 6 to 13. A few of the newly described taxa have been recorded before under misapplied names. Two of the Amanitas known now from Colombia belong to sect. Amanita, four to sect. Vaginatae, one to sect. Lepidella, two to sect. Phalloideae and four to sect. Validae; only sect. Amidella is not represented.

All taxa are extensively and thoroughly described and illustrated, whereas they are carefully compared to possible relatives occurring elsewhere in the world.