The systematics of gasteroid, auricularioid Heterobasidiomycetes *)

F. Oberwinkler & R. Bauer

Lehrstuhl Spezielle Botanik der Universität Tübingen, 7400 Tübingen, Federal Republic of Germany

Oberwinkler, F. & R. Bauer (1989). The systematics of gasteroid, auricularioid Heterobasidiomycetes. – SYDOWIA 41: 224–256.

Most species of the gasteroid, auricularioid Heterobasidiomycetes have stipitate, capitate basidiocarps and simple septal pores. They differ in characteristics of basidia, basidiospores, conidia, hyphae, haploid states (i.e., yeasts or hyphal), and substrate specifities, and in the properties of the 5S rRNAs. Systematically important characters are interpreted comparatively with special reference to septal pores and spindle pole bodies. An unknown membrane complex, the symplechosome, is described and evaluated taxonomically. It is concluded that the gasteroid, auricularioid Heterobasidiomycetes are heterogeneous. Therefore several new taxa are proposed to accommodate natural relationships: Agaricostilbales, Agaricostilbaceae, Pachnocybaceae, and Atractogloeaceae.

Oberwinkler & Bandoni (1982.a) classified auricularioid fungi with gasteroid basidia and related holobasidiate taxa in the Atractiellales. The circumscription of the order was based on comparative morphology of basidiocarps, hyphal systems, basidial ontogeny, spore development, and ultrastructure of the septal pores. It was definitely concluded that *Agaricostilbum* has basidiomycetous affinities, and that the Atractiellales seems necessary to accommodate those species which differ markedly from *Auricularia,* the type genus of the Auriculariales. Also *Atractiella* could be transferred from the Deuteromycetes to Basidiomycetes. *Pilacrella* was found to be a synonym of *Atractiella.* The Chionosphaeraceae were proposed for the holobasidiate *Chionosphaera* and the phragmobasidiate *Stilbum. Pachnocybe ferruginea* was considered to be a heterobasidiomycete of uncertain taxonomic position. The genus *Atractogloea* was erected (Oberwinkler & Bandoni, 1982.b) for a species with pulvinate basidiocarps, clamped hyphae with simple septal pores and agaricostilboid basidia. For illustrations of morphological details we refer to the publications of Oberwinkler & Bandoni (1982.a,b).

*) Part 56 of a series "Studies in Heterobasidiomycetes" of the Botanisches Institut, Lehrstuhl Spezielle Botanik, Universität Tübingen.

Materials and Methods

Organisms

For descriptions and illustration of the species, the following collections were used:

Agaricostilbum cf. *hyphaenes* (HAR. & PAT.) OBERW. & BANDONI, Indonesia, Java, Bogor Botanical Gardens, on the inflorescences of *Latania loddigesii* (Palmae), 250 m, June 1962, M.A. RIFAI 322. – *Agaricostilbum pulcherrimum* (BERK. & BR.) BRADY, SUTTON & SAMSON, Argentina, Buenos Aires, Sta. Catalina; en espata de palmera (Phoenix sp.), leg. WRIGHT, DESCHAMPS & DEL BUSTO, 27.4.1969. – Argentina, Entre Rios, Palmar de Colón, leg. A. MANRANTA, 11.11.1979; culture isolated from this collection by R.J. BANDONI, 13.7.1981. – Australia, New South Wales, Hastings Forest Reservation north of Wauchope, Wilson River Reservation, 250 m, on *Archontophoenix cunninghamiana*, 18.8.1981, leg. F. OBERWINKLER FO 32292. – *Pilacre hyphaenes* HAR. & PAT., Champignon sur les écailles d'une inflorescence mâle morte et tombée de *Hyphaene guineensis;* bords du fleuve Congo près de Boma (Bas Congo Belge); 14.7.1902, coll. MARTRET. Aug. CHEVALIER, type, PC.

Atractiella brunaudiana SACC., Saintes (BRUNAUD), in sedimento Coffeae arabicae humi dejecto, putrescente, type, PAD. – *Atractiella* sp., New Orleans, Louisiana, 2.2.1974, leg. and cult. R.J. BANDONI 6066 A. – *Atractiella solani* (COHN & SCHROET.) OBERW. & BANDONI, Scotland, St. Kilda, 9.1967, isolated from soil by R. WATLING. – *Atractiella* sp., Canada, British Columbia, Vancouver Island, 16.4.1981, leg. K. SEIFERT. – *Pilacrella delectans* MÖLLER, Brasilien, Blumenau, on *Euterpe oleracea*, type, HBG. – *Pilacrella solani* COHN & SCHROET., CBS 277–32.

Atractogloea stillata OBERW. & BANDONI, Stanford, California, on weathered spathe of *Phoenix canariensis*, coll. W. SHOFIELD, Dec. 27, 1980. Soaked in sterile, distilled water on Dec. 28, 1980. Mature fructifications were observed on Dec. 30, 1980 by R.J. BANDONI.

Chionosphaera apobasidialis COX, Illinois, Effingham Co., on *Quercus stellata*, 28.2.1971, coll. D. COX, type; specimens determined as *Stilbum vulgare* TODE: Ontario, Granton, on *Carpinus americana*, 18.1.1894, coll. J. DEARNESS, NY. – Jennings Gap, Virginia, on a hyphomycete on *Castanea dentata*, 13.9.1926, coll. W.W. DIEHL, det. F.K. CHARLES, BPI. – Butternut Brook, Litchfield Co., woods of White Memorial Foundation, on dead bark of *Carpinus caroliniana*, 10.1.1978, coll. C.T. ROGERSON, cult. R.J. BANDONI.

Pachnocybe ferruginea (SOW.:FR.) BERK., DAOM 33804, dried culture obtained from sapwood rot from 10' level of a *Picea engelmanni*, Victoria, British Columbia, 28.11.1951 (BBH-825 B). Culture from P.R. KROPP, Oregon State University (mis. W. WALKER).

Phleogena faginea (FR.) LINK, Speldhurst, Danemore Park, on burnt wood, standing trunks of coppiced *Alnus*, coll. B.J. COPPINS 2491 and C.B. RICHARDSON, mis. R. WATLING. – Österreich, Steiermark, Possruckgebirge, Heilig Geist Klamm südlich Leutasch, stehender Faulstamm von *Acer*, 18.5.1980, leg. J. POELT. – *Phleogena* sp., Australia, New South Wales, Hastings Forest near Wauchope, 1000 m, 15.8.1981, leg. F. OBERWINKLER 32014. – *Pilacre faginea* FR., on oak, Iowa City, Iowa, 29.9.1929, G.W. MARTIN. – Sao Leopoldo, Rio Grande do Sul, J. RICK, 1932, Mo.Bot.G.Herb. 150748, BPI.

Stilbum vulgare TODE, auf Buchenrinde bei Achimsthal (?), 25.3.1862, NIESSL, M. – FUCKEL, Fungi rhenani 2302, ad truncos putridos betulinos, raro, hieme, Hallgarten, M, NY. – Australia, New South Wales, Hastings Forest near Wauchope, Wilson River Reserve 250 m, 18.8.1981, on *Archontophoenix cunninghamiana*, leg. F. OBERWINKLER 32291; yeast cultures from R.J. BANDONI F 356, F 357.

Tulostoma brumale PERS., Deutschland, Baden-Württemberg, Tübingen, Spitzberg bei der Ödenburg, 4.5.1976, leg. T. ANKE & F. OBERWINKLER 23594; 18.9.1976, FO 24052.

Light and electron microscopy

Living and untreated material of different developmental stages was studied with a Zeiss photoscope III, using phase optics and Nomarski interference contrast optics. For transmission electron microscopy samples were fixed in 2% glutaraldehyde in O.1 M sodium cacodylate buffer at pH 7.2 overnight or during several days. Following six transfers in 0.1 M sodium cacodylate buffer, the material was postfixed in 1% OsO4 in the same buffer for 2 hours in the dark, washed in distilled water, and prestained in 1% uranyl acetate solution for 1 hour in the dark. After 5 washes in distilled water, the material was dehydrated in acetone, using 10 minute changes at 25%, 50%, 70%, 95% and 3 × 100% acetone. The material was embedded in SPURR's (1969) plastic. Series of sections were cut on a Reichert ultramicrotome using a diamond knife and, after mounting on Formvar coated single slot copper grids, stained with lead citrate (REYNOLDS, 1963) at room temperature for 3 to 5 minutes, and washed again with water. The thin sections were examined with a Zeiss EM 109 transmission electronmicroscope at 80 kV.

Results and Discussion

1. Basidiocarps

Except for *Atractogloea stillata,* all presently known species of the gasteroid, auricularoid Heterobasidiomycetes have stipitate, capitate basidiocarps. Gasteroid basidiomycetes, e.g. *Phleogena* species commonly have angiocarpous basidiocarps within which maturation of basidia and spores occurs. Under these circumstances, active spore abstriction would be meaningless. Although most angiocarpous Basidiomycetes have gasteroid basidia, species with gymnocarpous basidiocarps and gasteroid basidia also are known. In species of *Agaricostilbum, Atractiella, Atractogloea, Chionosphaera, Pachnocybe,* and *Stilbum* the basidia are predominantly arranged in an exposed hymenium on the superior surface of the capitulum. In gross morphology of fruiting bodies, *Agaricostilbum* species are comparable with those of *Atractiella, Stilbum vulgare,* and *Chionosphaera apobasidialis*. However, these taxa are quite distinct in basidial morphology and basidiospore production.

Stilboid fructifications are not only wide-spread in numerous species of higher fungi, e.g. Ascomycetes, Deuteromycetes, Hetero- and Homobasidiomycetes, but also, e.g. in the lichenized Caliciales, and in many of the Myxomycetes. In general, stalked and stalked-capitate fructifications certainly evolved many times convergently in Basidiomycetes and also in Ascomycetes. Therefore, basidiocarp gross morphology is not applicable for delimitations of higher, natural taxa in Basidiomycetes, i.e. stilboid fructifications are not neces-

sarily a proof for a single and close natural relationship of atractielloid fungi.

Pustulate basidiocarps, such as those of *Atractogloea stillata,* are present in species of a variety of heterobasidiomycetous genera, e.g. *Platygloea,* and *Mycogloea* of the Auriculariales s.l. In the Auriculariales s.str. (BANDONI, 1984) such fructifications are known from species of the genera *Myxarium, Stypella, Pseudostypella,* and *Efibulobasidium.* Various species of *Tremella* have pustulate basidiocarps as do *Christiansenia pallida* (Tremellales), *Dicellomyces gloeosporus* (Exobasidiales), and a variety of *Dacrymyces* species. OBERWINKLER & BANDONI (1982.b) suggested that the pustulate basidiocarp form is primitive, a possibility also supported by its predominantly heterobasidiomycetous distribution. In Homobasidiomycetes such basidiocarps are relatively rare. Pustulate, gelatinous fructifications occur also in anamorphic taxa with heterobasidiomycetous affinities, as *Infundibura adhaerens* and *Pleurocolla compressa.*

2. Hyphae, hyphidia and hyphal systems

Thin-walled, hyaline hyphae predominate in gasteroid, auricularioid fungi and also in other Heterobasidiomycetes. However, variously thick-walled hyphae occur in several taxa, e.g. in species of *Agaricostilbum* and *Atractiella.* Dimitic hyphal systems are lacking in atractielloid species, and they are rarely found in other Heterobasidiomycetes. In contrast, in the Heterobasidiomycetes there is an unusually large number of taxa with gelatinizing hyphae which are said to be responsible for soft, jelly-like fruitbodies. With the exception of *Atractogloea stillata,* all other gasteroid, auricularioid species have non-gelatinized basidiocarps. Anastomoses of hyphae can be found frequently in stalks and heads of fructifications. In conclusion, hyphal characteristics are of rather limited value for taxonomic interpretations in atractielloid Heterobasidiomycetes. They cannot be used for delimiting higher taxonomic units.

Most atractielloid species have gymnocarpic hymenia in which also hyphidia are lacking. *Phleogena faginea* is clearly distinct in hymenial morphology. Branched hyphidia overgrow the basidial surface, thus forming a dense peridial layer. The taxon Phleogenaceae appears to be justified by the gasteroid basidiocarp structure. There are scattered hyphidia which protrude from the hymenium in *Atractiella solani,* or they are arranged marginally as in *Atractiella* spp. The taxonomic significance of simple hyphidial elements is difficult to evaluate.

3. Septa and septal pores

Heterobasidiomycetous fungi with abundant primitive characteristics lack clamps. This is true of all Uredinales, Septobasidiales, and the majoritiy of the Atractiellales s.l. In the gasteroid, auricularioid group, only *Atractogloea stillata* (Fig. 1) and species of the genus *Phleogena* have clamped hyphae. Thus, in large groups of the Heterobasidiomycetes, septal morphology is taxonomically important.

Two clearly distinct septal pore types are present in the auricularioid Basidiomycetes. Dolipores with continous parenthesomes have been reported from *Hirneola auricula-judae* by SEBALD (1977) and MOORE (1978), from species of the *Auricularia* complex by PATTON & MARCHANT (1978), TU & KIMBROUGH (1978), and MCLAUGHLIN (1980). Rust-like septal pores were demonstrated for *Herpobasidium filicinum* (SEBALD, 1977) and *Eocronartium muscicola* (SEBALD, 1977; KHAN & KIMBROUGH, 1980). All species examined in the present study were found to have simple pores, as shown in Figs. 1, 2, 7, 8, 13, 20, 23, 26, 31. It is assumed that the pore types together with basidial morphology, spore production, and germination, indicate that the Atractiellales are affiliated with the Ustilaginaceae, Uredinales, Septobasidiales, and Auriculariales s.l. Surprisingly, the simple septal pore type is shared even by the holobasidiate *Chionosphaera apobasidialis* (Fig. 26) and *Pachnocybe ferruginea* (Fig. 31).

The cell and septal walls of *Agaricostilbum* species are multilayered not only in thick-walled hyphae, but also in young and thin-walled ones. The septal pore openings are associated with electron dense globules which resemble ascomycetous Woronin-bodies (Figs. 7, 8) lacking peripheral membranes. Therefore, concerning ultrastructural characteristics of septal pores, *Agaricostilbum* appears to be rather close to Ascomycetes.

OBERWINKLER & BANDONI (1982.a, Figs. 34–36) illustrated median sections of septal pores of an *Atractiella* sp. in which the simple pores are often associated with electron dense globose compartments. An isolate, named *Pilacrella solani*, (CBS 277–32) was also examined with the transmission electron microscope (Fig. 13). Almost identical septal pore details were found in this species. It could be proven that the hyphal cells were dikaryotic. A fragment of the generic type, *A. brunaudiana*, was also used for a TEM examination. In the almost hundred years old herbarium material, simple septal pores could be found. The globose vesicles in *Atractiella* species (Fig. 13) and *Phleogena faginea* (Fig. 20) seem not to be comparable to Woronin-bodies in Ascomycetes because of an other structural differentiation in electron-dense peripheral layers and electron-transparent central parts.

It is remarkable, that in *Atractogloea stillata* (Fig. 2), a swollen zone immediately surrounding the pore tapers abruptly toward the center (OBERWINKLER & BANDONI, 1982.b). The authors compared that pore type with those found in *Eocronartium muscicola* (SEBALD, 1977; KHAN & KIMBROUGH, 1980), and in *Septobasidium* (DYKSTRA, 1974), but remarked for the latter taxon that "the bulge surrounding the pore in *Eocronartium muscicola* (PERS. ex FR.) FITZP. is not always present and is possibly characteristic of a special ontogenetic stage". Nevertheless, it can be assumed that the central swellings of dolipores may have evolved by gradual flaring of septal wall portions adjacent to the pore. Therefore, the ultrastructure of the *A. stillata* pore is taxonomically and phylogenetically important.

Comparative morphological studies led OBERWINKLER & BANDONI (1982.a) to conclude that *Pachnocybe ferruginea* is not a deuteromycete, but a basidiomycete. Herbarium material was used to study the septal ultrastructure which was found to have a simple pore. In further studies with material from cultures simple septal pores (Fig. 31) were found again. The septal wall shows basidiomycetous multilayered lamellation.

Four distinct types of simple septal pores can be recognized in the gasteroid, auricularioid fungi: (1) *Agaricostilbum*-type in species of the genus *Agaricostilbum*. (2) *Atractiella*-type in species of the genera *Atractiella* and *Phleogena*. (3) *Stilbum*-type in *Stilbum vulgare, Chionosphaera apobasidialis,* and *Pachnocybe ferruginea*. (4) *Atractogloea*-type in *Atractogloea stillata*. The characteristics of each type appear to be constant and therefore applicable for systematic evaluations.

4. Spindle pole bodies

The morphology of spindle pole bodies (SPBs) is heterogeneous in heterobasidiomycetous fungi. For the best comparisons, ontogenetic cycles of mitoses and/or meioses are needed. Such data are not yet available for gasteroid, auricularioid fungi. Therefore, we can only compare interphase and early prophase SPBs. The figures are selected from pictures of serial sections.

The SPBs of *Agaricostilbum pulcherrimum* (Fig. 12), *Atractiella solani* (Fig. 14), *Phleogena faginea* (Fig. 21), *Stilbum vulgare* (Fig. 24), *Chionosphaera apobasidialis* (Fig. 27), and *Pachnocybe ferruginea* (Figs. 32, 33) consist of two lateral discs connected by midpieces. Two types of disc orientation in SPBs of these taxa can be recognized: (1) The discs have an approximately vertical position in respect to the nuclear envelope in *A. pulcherrimum* (Fig. 12), *St. vulgare* (Fig. 24), and *Ch. apobasidialis* (Fig. 27). They are in close proximity to the nucleus only with its narrow sides. *A. solani* (Fig.

14), *Ph. faginea* (Fig. 21), and *Pachnocybe ferruginea* (Figs. 32, 33) have obliquely positioned discs in interphase and prophase SPBs. Discs of prophase I SPBs in *P. ferruginea* (Fig. 32) and interphase SPBs in *A. solani* (Fig. 14) have electron dense central layers. Interphase and prophase SPBs with discs are known from Basidiomycetes only in the Uredinales (HEATH & HEATH, 1976; HEATH, 1981; O'DONNELL & MCLAUGHLIN, 1981.a,b,c; BAUER, 1987) and in *Helicobasidium mompa* (BOURETT & MCLAUGHLIN, 1985). SPBs with discs in *Ch. apobasidialis* and *P. ferruginea* are the first records from holobasidiate taxa.

In contrast to all other atractielloid taxa, the interphase SPB in *Atractogloea stillata* is diglobular (Fig. 4). An electron dense midpiece connects the homogeneous, non lamellated, globular elements. Similar SPBs have been described and illustrated from several Homobasidiomycetes (WELLS, 1977), *Bullera alba* (TAYLOR & WELLS, 1979), *Auricularia fuscosuccinea* (MCLAUGHLIN, 1980), *Tremella globospora* (BERBEE & WELLS, 1988). However, the globular element in SPBs of *Leucosporidium scottii* (MCCULLY & ROBINOW, 1972.a), *Sporidiobolus salmonicolor* (MCCULLY & ROBINOW, 1972.b), and *Ustilago maydis* (O'DONNELL & MCLAUGHLIN, 1984.a,b) contain an electron dense layer. So far, SPBs with homogeneous globular elements were only known from Basidiomycetes with dolipores.

Interphase and prophase SPBs in Basidiomycetes show stable morphological characteristics. Therefore they are applicable to taxonomic evaluations. Two major types can be distinguished, the disc-SPB and the diglobular SPB. Since all other atractielloid species have disc-SPBs, *A. stillata* with a diglobular SPB cannot be retained in the Atractiellales.

5. Membrane complexes

In *Atractiella solani* (Figs. 15, 16) and *Phleogena faginea* (Figs. 18, 19) we found membrane complexes which have not been described so far. Several layers of endoplasmatic reticulum are positioned in close proximity and interconnected by electron dense bars. The distance of the ER-membranes is ca. 0.03–0.04 µm in the connected region. Connecting bars appear to be rather regularly arranged, with a distance from each other of approximately 0.01-0.02 µm. Bars of half the length of interconnecting ones occur at the outer membrane surfaces. Within the connected area ER-membranes are flattened and tightly narrowed. At the periphery of the complex, outside of the connecting structures, ER-membranes often are flare, thus resembling dictyosomes. However, membranes of dictyosomes are not connected by electron dense bars. Therefore we

propose the term **symplechosome** for the interconnected membrane complexes in certain heterobasidiomycetous species.

These findings were presented and discussed for the first time by the senior author in the annual lecture of the 1985 annual meeting of the American Mycological Society in Gainesville, Florida. Further studies have proven the constancy of the symplechosome. However, its function is still unknown.

The presence of simple septal pores, associated with globular bodies, and the symplechosomes in species of *Atractiella* and *Phleogena* appears to be of considerable taxonomic importance. We have found these characteristics also in species of the genera *Helicogloea* and *Saccoblastia* (unpubl.). A comprehensive taxonomic evaluation will be published elsewhere.

Often mitochondria in subbasidial cells of *Pachnocybe ferruginea* (Fig. 30) are interconnected by electron dense bars, 0.04 µm long, and separated from each other approximately 0.01-0.02 µm. Mitochondrial complexes of such a type are not known from other Basidiomycetes.

6. Basidia

In the auricularioid fungi, the basidium typically is elongate, cylindrical, and usually transversely septate, as in *Atractogloea, Agaricostilbum, Atractiella* and *Stilbum* species. Although four-celled basidia predominate in this group, a single septum is typical after meiosis in *Stilbum vulgare* basidia. Phylogenetic connections to non-septate, i.e. holobasidiate species, such as *Chionosphaera apobasidialis* have been proposed (Oberwinkler & Bandoni, 1982.a).

In contrast to the Homobasidiomycetes, spore formation in gasteroid Heterobasidiomycetes is comparable to budding in basidiomycetous yeasts. The apparent "septation" of the basidium actually involves deposition of complete new walls around each of the several protoplasts within the old basidial wall. The latter wall is ruptured as sterigmata are formed or basidiospores are budded off (Figs. 9–11). The rupture of the outer wall is plainly visible by light microscopy in *Atractogloea stillata* and *Agaricostilbum pulcherrimum,* species exhibiting multiple spore production by each basidial cell (Wright & al., 1981; Oberwinkler & Bandoni, 1982.a). Multiple spore production of this type also occurs in the Ustilaginaceae. There is some evidence that basidia budding off numerous spores represent relics of original meiosporangia. The residual nuclei in the meiosporangial cells are capable of further mitotic divisions for producing haploid successors of primary meiospores. In *Agaricostilbum* species, the number of spores produced by a single basidium is high, but probably not constant (Ober-

WINKLER & BANDONI, 1982.a). Up to 12 spores of approximately the same developmental stage can be found attached to a single sporogenous locus in *A. pulcherrimum*. Younger spores attached to the basidium together with older ones indicate that spore production can continue for some time. Budding of the basidiospores while still attached, also occurs (WRIGHT et al., 1981). Basidia can be found with single basidiospores attached to the loci, although additional spores probably would be produced.

In *Atractiella* and *Stilbum* species, spore formation is accompanied by migration of the entire protoplast from the basidial cell into the spore. The number of basidiospores thus equals the number of basidial cells, i.e. commonly four in *Atractiella*. A distinctive characteristic of *Stilbum vulgare* is basidial morphology. Mature basidia are long stalked and apically enlarged into two spore-producing cells. This upper part of the basidium is curved, thus enabling both basidial cells to develop short sterigmata to the outside. However, the spores apparently are passively released. This is deduced from the morphology of sterigma apex, apiculus and attachment of basidiospore. JUEL (1898) was the first and so far the only investigator who studied *St. vulgare* in detail. He did not only describe the morphology of the species adequately, but examined also the karyology, and found the spore-building cells to be meiosporangia. He also gave reasonable arguments for identifying the species studied by him with *St. vulgare* TODE. Later authors unanimously accepted these conclusions.

The holobasidiate *Chionosphaera apobasidialis* (Fig. 25) has variable numbers of apical basidiospores, mostly 4-6. Inconstant numbers of basidiospores are known also in Homobasidiomycetes, but they are exceptional and the spores are mostly formed synchronously. This can be considered as an as yet unfixed feature in a primitive genus such as *Chionosphaera*, and as an aberrant pattern in the more highly evolved Homobasidiomycetes. It is, at present, difficult to interpret the varying numbers of basidiospores of some homobasidiomycetous gasteroid forms.

BREFELD (1908) succeeded in obtaining the whole life cycle, including basidiocarps of *Phleogena faginea* in pure culture. SHEAR & DODGE (1925) verified BREFELD's findings in careful investigations during which also karyological studies were carried out. "It is shown that this fungus has true basidia, each arising from a clamp connection, which is a two-nucleate structure. These nuclei fuse and undergo two divisions so that the mature basidium consists of four uninucleate cells". In the course of further experiments, BECKWITH (1929) "... proved that single spore cultures do not develop basidia. They are produced only when two strains of opposite sex are grown together." She concluded that "From the behavior of these cultures,

it seems evident that the fungus is heterothallic,...". The relationship of *Phleogena* has been discussed by most authors dealing with that fungus. But only since BREFELD (1888) elucidated the morphology of the basidiocarp and basidia, and studied the whole life cycle of *Ph. faginea,* sufficient facts were available for an adequate interpretation.

Another holobasidiate species of the gasteroid Heterobasidiomycetes is *Pachnocybe ferruginea.* Basidiospores are budded off apically (Figs. 28, 29) from a short-cylindrical basidium. Spore formation is similar to that in *Chionosphaera apobasidialis.*

7. Basidiospores

In the Atractiellales, the blastogenous basidiospores are sessile in *Atractogloea stillata, Phleogena faginea* (Fig. 17), and in the species of *Atractiella.* Short sterigmata are present in *Stilbum vulgare,* as was already illustrated by JUEL (1898). *Chionosphaera apobasidialis* also has short sterigmata and its spores, like those of *Stilbum vulgare,* are thick, rough-walled, and hyaline. Short, sterigma-like outgrowths are present in *Agaricostilbum* species (Figs. 9–11). The thickened, hyaline basidiospore walls are characteristic of species of *Agaricostilbum.* Basidiospores of *Atractiella* species are thin-walled or only moderately thickened and hyaline. Exceptional for all Heterobasidiomycetes are the thick-walled and brown basidiospores in *Ph. faginea* and *P. ferruginea.* These spore characteristics can be used, inter alia, for delimitations of higher taxa, i.e. the families Phleogenaceae and Pachnocybaceae.

In the auricularioid, gasteroid Basidiomycetes, including the Ustilaginales s.str., forcible discharge of basidiospores is lacking; both the primary spores and secondary spores are formed blastogenously. In other words, the "sexual" and "asexual" propagules are produced in the same way. Therefore also secondary ballistospores are lacking.

8. Yeasts

Knowledge of the essential parts of the life cycle is necessary for correct assessment of the importance of yeast stages. Budding most often occurs at the beginning of ontogenetic development, i.e. at basidiospore germination, and a yeast phase may develop. In basidiomycetous yeasts, budding results in rupture of the outer wall layer of the mother cell. Budding is commonly polar, the successive buds at any locus resulting in a series of annular scars that sometimes resemble those produced in formation of annelloconidia. In the atractielloid fungi, yeast phases are present in *Atractogloea still-*

ata, Agaricostilbum species, *Stilbum vulgare,* and in the holobasidiate *Chionosphaera apobasidialis.* According to JUEL (1898), germinating basidiospores of *St. vulgare* remain aseptate and form germ tubes. JUEL's description is not clear, but it can be suggested that conidia may be produced. During the present investigations, yeast stages were developing from germinating spores of *St. vulgare.*

In the course of further investigations in *A. stillata,* a yeast stage was obtained. Budding of this strain (Fig. 3) is comparable to what was described earlier from *Rhodotorula* spp. (THYAGARAJAN & al., 1962; MARQUARDT, 1962), *Sporobolomyces roseus* (PRUSSO & WELLS, 1967), *Tremella* (BANDONI & BISALPUTRA, 1971), and *Agaricostilbum pulcherrimum* (OBERWINKLER & BANDONI, 1982.a). During budding, the mother cell wall ruptures and the daughter cell enlarges by continuous growth of the inner mother cell wall layer which extends into the wall of the bud.

Another unusual and unexpected characteristic is endospore production in yeasts of *A. pulcherrimum.* One strain producing endospores was studied with the transmission electron microscope. Ultrastructural characteristics of the yeast cell wall and yeast-budding (Fig. 5) clearly show basidiomycetous characteristics. It is rather surprising that budding cells can also produce endospores. Obviously both processes can occur in a short sequence of time, but not fixed in a regular order. Several four-nucleate stages (Fig. 6) were found, but no synaptonemal complexes could be detected. Therefore, it is uncertain whether *Agaricostilbum* endospore formation is tied to meiosis or is the result of mitoses. That question is important (1) in comparison with the life cycle and its nuclear phases, and (2) phylogenetically, concerning the origin of the basidiomycetous meiosporangium. WRIGHT et al. (1981) stated "A fusion nucleus can be found in young basidia (Fig. 7). Following meiosis, each basidial compartment is uninucleate, as are the basidiospores". Thus, meiosis occurs in basidia. It would be unique and a highly important pecularity, if a second meiosis would take place in one individual life cycle. It is rather certain that the yeasts capable of endospore formation are *Agaricostilbum* yeasts, and not contaminants. Attention should be drawn to four other unusual taxa: (1) *Cystofilobasidium capitatum* produces filobasidiaceous basidia from germinating teliospores. Under certain conditions these resting spores can produce endospores. (OBERWINKLER & al., 1983). (2) *Sporidiobolus johnsonii* and *S. salmonicolor* teliospores can germinate with external basidia or by endospore formation. BANDONI (1984) suggests that the endospores of *Sporidiobolus* species are meiospores. (3) There is an unparalleled situation in *Platygloea sebacea* with auricularioid basidia that are capable of endospore formation (OBERWINKLER, unpubl.). It can be suggested that species

still exist in which meiospore ontogeny is variable. Such taxa might be descendents of old and probably also primitive Basidiomycetes.

Pachnocybe ferruginea completes its entire life history in culture (KROPP & CORDEN, 1986). Basidiospores produce short germination tubes from which single, allantoid cells are budded off. We found that budding may continue for a certain period. However, yeast colonies have not been observed.

Atractiella species and *Phleogena faginea* are the only taxa in the gasteroid, auricularioid group without a dimorphic ontogeny. This is considered of high taxonomic importance and could be used for an emended interpretation of the Atractiellales.

9. Conidia

Basidiospores of *Atractiella* species often germinate by short germ tubes which produce "microconidia". This was first reported by MÖLLER (1895) for *Pilacrella (Atractiella) delectans,* in which continuous production of microconidia occurs. BOIDIN & al. (1979) reported microconidial production on conidiophores in a species of *Hoehnelomyces (Atractiella).* It is not known whether the microconidia of these species are capable of budding.

BREFELD (1888) was the first who cultivated *Phleogena faginea* (as *Pilacre petersii*), described teleomorph and anamorph stages in detail, and illustrated the species outstandingly. BREFELD's theory, derived from a study of the whole life history of *Ph. faginea,* was the evolution of the basidium from the variable conidiophore to the definite, four-spored basidium. SHEAR & DODGE (1925) compared the anamorph stage of *Ph. faginea* with the hyphomycetous genera *Rhinotrichum* (nomen dubium according to HUGHES, 1958) and *Haplaria* (= *Botrytis,* fide HUGHES, 1958). KENDRICK & WATLING (1979) mentioned, that the anamorph stage of *Ph. faginea* "... resembles a *Nodulisporium,* though that form genus is usually associated with xylariaceous Ascomycetes".

Allantoid conidia develop acropetally on branched conidiophores in *Pachnocybe ferruginea* in pure culture.

The lack of conidial stages in *Atractogloea stillata, Agaricostilbum spp., Stilbum vulgare,* and *Chionosphaera apobasidialis* is substituted for yeast phases.

10. 5S ribosomal RNAs

Nucleic acids are considered to be rather conservative molecules, remaining unchanged during long periods of evolution. According to KIMURA (1968, 1969, 1980, 1983), KING & JUKES (1969), LANGLEY & FITCH (1974), SMITH & COSS (1984) molecular evolution is caused by neutral mutations. Only functional sites of the molecules

appear to be selected, while so-called unfunctional ones may evolve continuously. The slow rate of nucleotide substitution is considered to be a suitable tool for delimiting phylogenetically distant groups. 5S rRNA characteristics have been used by several workers for taxonomic interpretations. So far the 5S rRNAs of more than 500 organisms have been sequenced (WOLTERS & ERDMANN, 1988). The universal alignment which is numbered according the E. coli sequence (WOLTERS & ERDMANN, 1988) is adopted for the following discussion.

The 5S rRNA primary sequences of 57 species of Basidiomycetes have been identified by WALKER & DOOLITTLE (1982, 1983), HUYSMANS & al. (1983), LIU & NAZAR (1983), BLANZ & GOTTSCHALK (1984), GOTTSCHALK & BLANZ (1984, 1985), and WALKER (1984). Nucleotide exchanges of more than 50% within Basidiomycetes have been found. These exchanges can be calculated in different ways. WALKER & DOOLITTLE (1982) distinguished 5 clusters which were also used in an enlarged calculation by GOTTSCHALK & BLANZ (1985). A phenogram of the fungal subtree compiled by HUYSMANS & DE WACHTER (1986) does not contain (inter alia) the atractielloid taxa *Agaricostilbum pulcherrimum, Atractiella solani,* and *Phleogena faginea.* In a "phylogenetic tree" of 5S rRNAs from Basidiomycetes by HORI & OSAWA (1987) Atractiella and Phleogena are also lacking.

A model of 5S rRNA secondary structure for Eukaryotes was first published by NISHIKAWA & TAKEMURA (1974). This structure is rather conservative. It contains 5 helices and 5 loops. HUYSMANS et al. (1983) proposed a secondary structure model for Basidiomycetes. Type A and type B secondary structures were distinguished by GOTTSCHALK (1985). These types differ in positions 4:116 (UG in type A; UU in type B), 16:68 (CG in type A, GC in type B), 105.1 (insertion U in type A), and 102 (A in type A; C in type B).

WALKER (1984) sequenced the 5S rRNA of *Agaricostilbum pulcherrimum.* There are 40 to 62 nucleotide exchanges in comparison with other Basidiomycetes (GOTTSCHALK & BLANZ, 1985). *A. pulcherrimum* 5S rRNA has a unique A insertion in position 105.1 which is unknown from any other organism and therefore also deviates from the U-insertion of type A secondary structure. CG in position 73:103 is not known from other Eukaryotes; consequently it does neither correspond with type A nor with type B secondary structures, both with a nucleotide pair GC in this position. The nucleotide pair GC in position 16:68 is shared with type B-structures (CG in type A). Type B sequences correspond in UU (4:116), however *A. pulcherrimum* deviates by AU. Comparisons of the nucleotide sequence with other Basidiomycetes, as well as evaluations of the secondary structure and signature nucleotides (WALKER, 1984; GOTTSCHALK, 1985; BLANZ & GOTTSCHALK, 1986) clearly result in an isolated position of *A. pulcherrimum.*

GOTTSCHALK (1985) found 16 nucleotide exchanges in 5S rRNAs of *Atractiella solani* and *Phleogena faginea*. The comparatively low exchange rate groups the two species in two closely positioned clusters (BLANZ & GOTTSCHALK, 1986) within the range characterized by type A secondary structure. The nucleotide pairing GC in position 5:115 is unique within all sequenced Basidiomycetes. It is considered to represent an apomorphy by WOLTERS (1987).

Pachnocybe ferruginea was sequenced by WALKER (1984) and referred to cluster 2, containing in addition two *"Rhizoctonia"* species, i.e. *Helicobasidium purpureum* and a certainly misinterpreted *"Rhizoctonia hiemalis"*. GOTTSCHALK (1985) added *Septobasidium carestianum* to this group with type A secondary structure. Cladistic analysis (WOLTERS, 1987) supports this cluster by the apomorphic GC in position 8:112.

Atractielloid taxa were favorites for 5S rRNA sequencing, since they were correctly estimated to play a major role in understanding evolution in Basidiomycetes. *Chionosphaera apobasidialis* is one of the holobasidiate taxa in this group according to the original concept of OBERWINKLER & BANDONI (1982.a). A sequence published by WALKER (1984) led to a grouping together with *Sterigmatomyces penicillatus, Tremella mesenterica, Bullera alba,* and *Agaricus "edulis"*. According to GOTTSCHALK (1985), GOTTSCHALK & BLANZ (1985), BLANZ & GOTTSCHALK (1986), and BLANZ & UNSELD (1987) *Ch. apobasidialis* is in close proximity to cluster 5, containing doliporoid Heterobasidiomycetes and all so far sequenced Homobasidiomycetes. In the cladogram of WOLTERS (1987) it is also grouped marginally to cluster 5 within a misinterpreted assemblage of so-called "Doliporobasidiomycetes". In total, the different evaluations yield similar results. However, this is in obvious contrast to convincing systematic conclusions based on morphological, ultrastructural, and ontogenetic data.

A cladistic analysis of WOLTERS (1987) confirms type A/type B dichotomy as proposed by GOTTSCHALK & BLANZ (1985). The cladogram is considered to illustrate not only evolutionary steps of the 5S rRNA molecule, but also organismic evolution. Type B is named "Doliporobasidiomycetes", misinterpeting such taxa as *Ustilago maydis* and *U. hordei* which have irregular, non doliporoid septa, and *Exobasidium vaccinii* and *Microstroma juglandis* with definitely simple septal pores.

Classifications derived from 5S rRNA properties indicate an isolated position of *A. pulcherrimum* within the Basidiomycetes. Simple septal pores with associated electron dense bodies, and yeasts capable of endospore formation are in accordance with a new interpretation. SPBs with discs are also distinctive for basic groups of the Heterobasidiomycetes. *Atractiella* and *Phleogena* spp. may

represent the core of the Atractiellales s.str. as derived mainly from ultrastructural data. This is in good agreement with 5S rRNA homologies. The holobasidiate *Pachnocybe ferruginea* appears to be correctly placed together with other simple pored, predominantly phragmobasidiate Heterobasidiomycetes, including *Septobasidium carestianum* (GOTTSCHALK, 1985). *Chionosphaera apobasidialis* is misplaced compared with positions derived from all other important characteristics.

11. Ecology and distribution

Unfortunately our knowledge on ecological properties of atractielloid fungi is extremely poor. Nevertheless, it is an interesting fact that nearly all *Agaricostilbum* specimens known, are collected from palms. However, BRADY et al. (1984) report one collection of *A. pulcherrimum* on *Xanthorrhoea* sp. from Australia. In their host list also *Pandanus fascicularis* from India is mentioned. OBERWINKLER et al. (1982), dealing with the palmivorous Graphiolaceae gave a short outline on Basidiomycetes restricted to palmaceous substrates. One result of COX's (1976) studies concerns the association of *Chionosphaera apobasidialis* with contaminating hyphomycetes, *Chloridium minutum* and *Cladosporium herbarum* that influence fruiting of the species. According to COX (1976): "It seems more likely that *Chionosphaera* is a contact mycoparasite which obtains from other fungi the stimulants needed to produce basidiocarps." The specimens studied by COX (1976) were collected from corticate branches of *Carpinus caroliniana* and *Quercus stellata*. OBERWINKLER & BANDONI (1982.a) examined material from *Carpinus americana, C. caroliniana, Castanea dentata,* and *Quercus stellata*. It is unclear whether *Ch. apobasidialis* is restricted to bark of amentiferous trees of the Betulales and Fagales, as might be suggested from the present collections. The species has not yet been reported from outside of North America.

Most herbarium specimens labeled as *Stilbum vulgare* and examined by OBERWINKLER & BANDONI (1982.a) from North America were *Chionosphaera apobasidialis*. MARTIN (1952) who listed the species in the "North Central Tremellales", noted that he had no record of its occurrence in the north central region. Under favorable conditions, *St. vulgare* can grow in large populations, which were found on dead inflorescences of *Archontophoenix cunninghamiana* (Arecaceae) in Australia. The substrates on which *St. vulgare* has been collected are rather heterogeneous. It is not possible to interpret these data ecologically. More field observations are needed for better understanding of the ecological niches in which this species lives.

Because of the uncertainty of species delimitation and identification, no reliable information on the geographic distribution of *Phleogena faginea* can be given at present. Literature reports provide scanty information on the ecology of *Ph. faginea*. A rather detailed description of the habitat was given by TALLASCH (in TALLASCH & JAHN, 1970). Besides growing on standing *Fagus* and *Carpinus, Ph. faginea* was also found on logs of *Fagus* lying on the ground (TALLASCH & JAHN, 1970). Specimens from Australia (FO 32014, 32088, 32259) were growing in essentially comparable localities, standing and fallen logs of angiosperm trees, on bark and rather hard, mainly undecayed wood. MÖLLER (1895) who collected *Phleogena* (identified by him as *Pilacre petersii*) in Blumenau, South Brasil, mentioned that it was growing on dry and very hard branches, and another gathering on construction wood of *Cedrela* sp. Thus, it appears that *Phleogena* species prefer angiosperm substrates of hard wood and bark on standing trees or only partly decayed lying logs. Reports from coniferous wood of *Picea abies* (WOJEWODA, 1977) have to be confirmed. It also seems that this species predominantly develops fruiting bodies during the cold season. Both, special habitat and time of fructification may be reasons for rare collections.

12. Systematics, phylogeny and taxonomic conclusions

The order Atractiellales was introduced (OBERWINKLER & BANDONI, 1982.a) to accommodate those species which differ markedly from *Auricularia,* the type genus of the Auriculariales, and from other auricularioid taxa with actively abstricted basidiospores. The *Auricularia-Hirneola* group is distinguished by its peculiar basidiocarps, which are unlike those of other genera currently treated in the Auriculariales, by the curved microconidia formed by germinating basidiospores, and by dolipores with parenthesomes.

On the basis of the above discussions we propose the following taxonomic rearrangement for atractielloid Heterobasidiomycetes:

Agaricostilbales: Agaricostilbaceae.
Atractiellales: Hoehnelomycetaceae, Phleogenaceae, *Helicogloea, Saccoblastia.*
Incertae sedis: Chionosphaeraceae; Pachnocybaceae; Atractogloeaceae.

Agaricostilbum species as circumscribed by the stalked-capitate, arid basidiocarps, gasteroid, auricularioid basidia capable of multiple spore production, simple septal pores with associated electron dense bodies, disc-like interphase-prophase SPBs, yeasts with endospores, and an aberrant 5S rRNA nucleotide sequence appear to

be unique in the Basidiomycetes. Therefore, a new order and a new family are proposed to accomodate the genus.

Agaricostilbales OBERWINKLER & BAUER, ordo nov.

Heterobasidiomycetes fructificationibus stillatis vel stipitiformiter capitatis, ex hyphis distinctis hyalinisque compositis. Septa hypharum simpliciter perforata. Cystidia nulla. Basidia longa, mature transverse septata, Gasteromycetum modo basidiosporis non eiectis, repetite oreundis. Basidiosporae hyalinae, tunicis non amyloideis, cellulas singulas germinando producunt.

Typus ordinis: Agaricostilbaceae OBERW. & BAUER, opus ipsum.

Agaricostilbaceae OBERWINKLER & BAUER, fam. nov.

Agaricostilbalium familia unica ordinis proprietatibus analogis.

Typus familiae: *Agaricostilbum* WRIGHT, emend. WRIGHT, BANDONI & OBERW., Mycologia 73: 885, 1981.

Agaricostilbum was originally described as a genus of the Deuteromycetes (WRIGHT, 1970). After a detailed investigation, WRIGHT et al. (1981), and OBERWINKLER & BANDONI (1982.a) came to the conclusion that the taxon can be assigned with certainty to the Heterobasidiomycetes. The genus *Amerobotryum* was introduced by SUBRAMANIAN & NATARAJAN (1975) for a synnematous fungus on *Cocos nucifera* (Arecaceae) from India. The authors (SUBRAMANIAN & NATARAJAN, 1977) later put *Amerobotryum indicum,* the type and only species, under synonymy with *Agaricostilbum palmicolum.* BRADY et al. (1984) studied synnematous fungi and found that *Isaria pulcherrima,* described by BERKELEY & BROOME (1873) from Ceylon, is conspecific with *Agaricostilbum palmicolum.* According to these authors also *Isaria cocoa* must be treated as a later synonym. Further, they found that the holotype of *Isaria palmae* is "...in poor condition and therefore its taxonomic status is uncertain". The name *Stevensomyces palmae* (MORRIS & FINLEY, 1965) was considered to be of doubtful application.

Agaricostilbales OBERWINKLER & BAUER, ordo nov.

The Atractiellales in an emended circumscription comprise auricularioid Heterobasidiomycetes with resupinate and stalked-capitate fructifications. Basidia produce sterigmata and ballistobasidiospores or they are gasteroid, developing sessile spores which are passively released. The septal pore is simple, but associated with vesicle-like globules, each having an electron dense peripheral layer and an electron translucent inner part. Membrane complexes, called

"symplechosomes", are regularly present in well-developed, cytoplasmatic cells. The interphase-prophase SPBs have discs.

Hoehnelomycetaceae

Atractiella and *Agaricostilbum* species share basidiocarp gross morphology, hyphal structure, septation, and arrangement. They differ in basidiospore ontogeny and germination, and in a wide range of various substrates. Each basidial cell produces a single blastogenous basidiospore in species of *Atractiella;* multiple budding that occurs in *Agaricostilbum* species has never been observed in species of *Atractiella.*

The genus *Atractiella* was introduced by SACCARDO (1886) to accommodate a single species, *A. brunaudiana.* He questioned ".., in capitulum expansis ibique basidia (?).." , but treated the genus in the amerosporous Hyalostilbeae of the Fungi Imperfecti. Shortly later, SCHROETER (1887) proposed the genus *Pilacrella* with a single species, *Pilacrella solani.* Finally, WEESE (1920) erected the genus *Hoehnelomyces* for a Javanian species, *H. javanicus.* Both *Pilacrella* and *Hoehnelomyces* have to be regarded as synonyms of *Atractiella.* According to DONK (1958), the correct name of this species is *Hoehnelomyces macrosporus* (PENZ. & SACC.) BOEDIJN (= *Sphaeronemella macrospora* PENZ. & SACC.)." – Though the type specimen of *S. macrospora* is in poor condition, a careful microscopic study revealed sufficient morphological details for a definite generic position in *Atractiella.* Moreover, the species is extremely similar to *A. brunaudiana* in basidiocarp morphology, structure, and dimensions of marginal hyphae, basidia and basidiospores. A specific difference may be given in shape of basidiospores, those of *A. brunaudiana* often being slightly constricted to pyriform, while spores of *A. macrospora* are predominantly ellipsoid to inconspicuously navicular. Both, *Sphaeronemella macrospora* and *Hoehnelomyces javanicus* are based on specimens from Java. BOEDIJN (in DONK, 1958) and DONK (1958) did not mention whether authentic material of both taxa was studied by them or not. An *Atractiella* sp., located in the Herbarium Bogoriense could be studied. Though not an authentic material, this collection strongly supports the conclusion that *Hoehnelomyces* is a synonym of *Atractiella.* OBERWINKLER & BANDONI (1982.a) examined the type of *Pilacrella delectans* MÖLLER. This species obviously belongs in the genus *Atractiella* because of the similarity of all important morphological characteristics with those of *A. brunaudiana.* On the other hand, *A. delectans* is easily distinguishable from the generic type and from *A. macrospora.* The marginal hairs are thin-walled, and the basidiospores distinctly smaller, perhaps due to shrinkage caused by preservation in a fixative. The species shares these characteristics with the other *Atractiella* taxa.

Phleogenaceae

Phleogena faginea is certainly the best known member of the auricularioid, gasteroid Heterobasidiomycetes with stalked-capitate basidiocarps. Though similar in outer appearance to other species in the Atractiellales, the morphology of *Phleogena* species is unparalleled to some degree: (1) Basidiocarps are composed of clamped hyphae with (2) simple septal pores; (3) many hyphae are brown colored; (4) hyphidia, strongly branched and protruding beyond the basidia to form a peridial layer; (5) basidiospores are thick-walled and brown; (6) they germinate with hyphae, and (7) yeast stages have never been observed. (8) Efibulate hyphae, originating from germinating spores are capable of producing an anamorph stage with sympodial conidiophores and blastogenously developed conidia.

Phleogena faginea has clamped hyphae, a unique characteristic in all stalked-capitate, gasteroid-auricularioid taxa. However, the pustulate *Atractogloea stillata* also has fibulate hyphae. That species does not either have colored hyphae and basidiospores nor hyphidia. Consequently also a peridial layer is lacking. Moreover, in *A. stillata* basidiospore development is a multiple budding process, and the spores are likewise germinate by budding. Therefore, *Atractogloea* and *Phleogena* are best separated in two distinct taxa. One of the taxonomically most important characteristics is the peridial layer formed of strongly ramified, entwined and fibulate hyphidia which protrude beyond the basidial layer. Such a morphology is unique in the auricularioid, gasteroid groups. Hyphidia are known from species of *Agaricostilbum* and *Atractiella*. But these are scattered and are structurally and functionally unlike those of *Phleogena*.

Direct germination of basidiospores by hyphae, and the lack of a yeast state are most remarkable. In all other taxa of the auricularioid, gasteroid Heterobasidiomycetes yeast stages or blastogenously formed microconidia are known. Though yeasts and secondary spores are lacking in *Phleogena*, the heterobasidiomycetous nature cannot be questioned. Both, the auricularioid basidium and the simple septal pore are heterobasidiomycetous features.

Brefeld (1888) was the first who found the auricularioid basidium in *Phleogena:* "In der Formausbildung der Basidien stimmen die Auricularieen mit der Familie der Pilacreen überein, beide haben quergetheilte 4zellige und 4sporige Basidien." Consequently he included the "Pilacreen" besides the "Auricularieen" and "Tremellineen" in a taxon that was called by him "Protobasidiomyceten" (Brefeld, 1888). Later he (Brefeld, 1889) noticed the similarity between *Phleogena* and *Tulostoma* concerning the

angiocarpous, stalked-capitate, gasteroid basidiocarp and the resemblance of the basidia. This similarity was first observed by SCHROETER (1877) who used TULASNE's descriptions and illustrations for a comparison with *Tulostoma*. SHEAR & DODGE (1925), accepting that interpretation, called *Phleogena* a "Protogasteromycete, whose nearest known relative among the puff balls appears to be *Tulostoma*". Not only the outer appearance of the basidiocarp but also basidial morphology are surprisingly similar. However, transverse septate meiosporangia never have been reported from *Tulostoma* species. Another difference seems to be basidiospore development on short sterigmata. For comparison we studied the ultrastructure of the hyphal septa and found dolipores with perforated parenthesomes, indicating a homobasidiomycetous relationship for *Tulostoma brumale*. For *Ph. faginea*, OBERWINKLER & BANDONI (1982.a) illustrated simple pores. Further investigations confirmed these findings. Commonly the pore is occluded by electron dense material, and small, dark-colored, and globose compartments are closely associated with the pore. This pore type is best comparable to that of *Atractiella* spp. *Helicogloea lagerheimii* and *Saccoblastia* sp. have an identical septal pore ultrastructure. These taxa also share the symplechosomes.

Chionosphaeraceae

The Chionosphaeraceae is represented by two species, *Stilbum vulgare* and *Chionosphaera apobasidialis*, each belonging to a monotypic genus. Both differ strongly in basidial morphology, *Stilbum* being phragmobasidiate and *Chionosphaera* holobasidiate. OBERWINKLER & BANDONI (1982.a) assigned the two taxa to one family because of a variety of taxonomically important similarities. These are: (1) Gross morphology of basidiocarps stalked-capitate; (2) hyphae efibulate, simple pored, hyaline; (3) hyphal arrangement compactly parallel in the stipe and densely ramified in the subhymenium; (4) cystidia and hyphidia lacking; (5) basidiospores borne on short, straight sterigmata; (6) basidiospore subelliptical, thick, rough-walled, hyaline; (7) yeast stage present. In addition, (8) disc-like SPBs occur in interphase-prophase of both taxa.

These characteristics are taxonomically suitable. Nevertheless, the question arises about the taxonomic importance of the phragmo- and holobasidial meiosporangium. In the present examples a careful morphological examination yields further arguments for closely related taxa. The basidia are subcylindric with slightly to conspicuously broadened apical parts; there, the basidiospores are produced on short basidial outgrowths. In *Ch. apobasidialis*, the sterigmata are arranged as a crown at the top of the basidium; in *St. vulgare*

usually two sterigmata are formed, each on a single basidial cell and both separated by a transverse septum. Commonly the upper parts of the basidia are conspicuously bent, thus exposing both spores to the surface. Occasionally, two sterigmata, originating from one basidial cell, could be found. Such peculiarities and irreguliarities may indicate that both basidial types are not only comparable morphologically, but may even be closely related phylogenetically. That interpretation presupposes that the phragmobasidial meiosporangium evolved into a holobasidiate one, or vice versa. The first explanation is favored here and mainly derived from a comparison of septal structures in tremellaceous basidia, in which several examples of partial septations have been found.

Ch. apobasidialis shares most of the taxonomically important characteristics with other gastroid, auricularioid taxa. These are (1) stilboid basidiocarps; (2) efibulate and simple pored hyphae; (3) gasteroid basidia with passively released basidiospores; and (4) often an anamorph stage of haploid yeasts. Filobasidiaceous species possess dolipores without parenthesomes, while in *Ch. apobasidialis* only simple septal pores could be found. Differences in basidiocarp morphology should not be valued too highly taxonomically. However, it has to be remembered that basidia in *Filobasidium* arise from a loose network of hyphae which do not really form a proper resupinate basidiocarp. In pure culture of *Ch. apobasidialis,* sporophores were obviously developed, though basidiocarps were not formed (Cox, 1976). Essentially two characteristics led Oberwinkler & Bandoni (1982.a) to propose a heterobasidiomycetous relationship for *Ch. apobasidialis,* the septal pore type, and basidiospore germination by budding. Such characteristics can also be used for Filobasidiaceae to place them in the Heterobasidiomycetes. Therefore, again, the proposal of Cox (1976) to place the Filobasidiaceae to the Aphyllophorales is rejected. Also the position of *Ch. apobasidialis* in 5S rRNA dendograms, based on Walker's (1984) data is contradictory to a systematic evaluation.

St. vulgare shares with most other gasteroid, auricularioid species (1) the synnematous, stilboid basidiocarp, (2) the efibulate hyphae, and (3) the simple septal pore. An important feature is the total lack of hyphidia, thus yielding hymenia composed solely of basidia. Also, sterile basidiocarp surfaces are not specially structured, and therefore appear to be smooth.

Pachnocybaceae Oberwinkler & Bauer, fam. nov.

Heterobasidiomycetes carposomatis stilboideis basidiisque aseptatis, sterigmatibus brevibus inde sporae plus minusve sedentes et mature non eiectae sunt. Basidiosporae brunneae, crassi-

tunicatae, tunicis levibus, non amyloideis, hyphis brevibus conidiis nascentibus vel cellulis singulis, allantoideis germinant. Septa hypharum simpliciter perforata sunt, dolipori absunt. Corpus nucleo associatum tempore inter divisionem nuclei parte media discis marginalibus binatim compositum est.

Typus familiae: *Pachnocybe* BERK., in SMITH's English Flora, 5(2): 333–335, 1836.

Pachnocybe is a genus commonly assigned to the Fungi Imperfecti (ELLIS, 1971). CARMICHAEL & al. (1980) suggested an ascomycetous relationship and referred to illustrations of PADEN (1977). However, OBERWINKLER & BANDONI (1982.a) who studied the morphology of *P. ferruginea,* inclusive of the septal pore ultrastructure, found strong evidences for basidiomycetous relationships. The authors decidedly did not include that taxon in the Atractiellales. 5S rRNA dendrograms, as discussed above, cluster *P. ferruginea* with *Septobasidium carestianum,* and *Helicobasidium purpureum.* Interphase and prophase SPBs with discs appear to be in accordance with such groupings.

Atractogloeaceae OBERWINKLER & BAUER, fam. nov.

Heterobasidiomycetes meiosporangiis transverse septatis sine sterigmatibus, Gasteromycetum modo basidiosporis non eiectis, repetite oreundis. Basidiosporae hyalinae, tunicis levibus, non amyloideis, cellulas singulas germinando producunt. Septa hypharum simpliciter perforata sunt. Corpus nucleo associatum tempore inter divisionem nuclei diglobosum est.

Typus familiae: *Atractogloea* OBERW. & BANDONI, Mycologia 74: 634, 1982.

Atractogloea stillata deviates from all other presently known species of the Atractiellales by the sessile basidiocarps and the fibulate hyphae. It lacks hyphidia and marginal hairs in or around the hymenium. Hymenia with such sterile terminal cells are found in members of the Chionosphaeraceae, fungi which differ in basidial morphology. Clamps are also lacking in many gasteroid, auricularioid species. *A. stillata* is rather exceptional in having clamps, which are found also in the *Phleogena faginea,* and which are common in the gasteroid Ustilaginales species. A unique combination of important characters, justifying the proposal of an own family for this taxon, are: (1) auricularioid basidia with multiple basidiospore production; (2) simple septal pores with slightly swelling pore margins; (3) diglobular SPBs, and (4) a yeast stage resulting from basidiospore budding.

Acknowledgments

We are grateful to Dr. R.J. BANDONI for critically reading the manuscript. We thank Miss S. SÜSSBRICH for skillful technical assistance with portions of TEM studies. We also thank the Directors of BPI, DAOM, HBG, M, NY, PAD, and PC, for loans of specimens of their institutions. Financial support from the "Deutsche Forschungsgemeinschaft" is gratefully acknowledged.

References

BANDONI, R.J. (1984). The Tremellales and Auriculariales: an alternative classification. – Trans. mycol. Soc. Japan 25: 489–530.

— & A.A. BISALPUTRA (1971). Budding and fine structure of *Tremella mesenterica* haplonts. – Can. J. Bot. 49: 27–30.

BAUER, R. (1987). Uredinales – Germination of basidiospores and pycnospores. – Stud. Myc. 30: 111–125.

BECKWITH, A.M. (1929). *Pilacre faginea* proves to be a heterothallic fungus. – Bull. Torrey Bot. Cl. 56: 359–360.

BERBEE, M.L. & K. WELLS (1988). Ultrastructural studies of mitosis and the septal pore apparatus in *Tremella globospora*. – Mycologia 80: 479–492.

BERKLEY, M.C. & C.E. BROOME (1873). Enumeration of the fungi of Ceylon. Part 2, containing the remainder of the Hymenomycetes, with the remaining established tribes of fungi. – Jour. Linn. Soc. 14: 29–140.

BLANZ, P. & M. GOTTSCHALK (1984). Comparison of 5S ribosomal sequences from smut fungi. – Syst. Appl. Microbiol. 5: 518–526.

— & — (1986). Systematic position of *Septobasidium, Graphiola* and other Basidiomycetes as deduced from their 5S ribosomal RNA nucleotide sequences. – Syst. Appl. Microbiol. 8: 121–127.

— & M. UNSELD (1987). Ribosomal RNA as a taxonomic tool in mycology. – Stud. Myc. 30: 247–258.

BOIDIN, J., F. CANDOUSSAU & P. LANQUETIN (1979). Premières récoltes d'*Hoehnelomyces* (Phragmobasidiomycètes, Auriculariales) en Europe; culture, cycle. – Beih. Sydowia Ann. Myc. Ser. II 8: 71–75.

BOURETT, T.M. & D.J. MCLAUGHLIN (1985). Mitosis and septum formation in the basidiomycete *Helicobasidium mompa*. – Can. J. Bot. 64: 130–145.

BRADY, B.L., B.C. SUTTON & R.A. SAMSON (1984). *Agaricostilbum pulcherrimum* (BERK. & BROOME) comb. nov. – Trans. Br. mycol. Soc. 83: 540–541.

BREFELD, O. (1888). Protobasidiomyceten. – Unters. Gesammtgeb. Mykol. VII, Verl. A. Felix, Leipzig.

— (1889). Basidiomyceten III. – Unters. Gesammtgeb. Mykol. VIII. Verl. A. Felix, Leipzig.

— (1908). Die Kultur der Brandpilze. – Unters. Gesammtgeb. Mykol. IX. Verl. A. Felix, Leipzig.

CARMICHAEL, J.W., W.B. KENDRICK, I.L. CONNERS & L. SIGLER (1980). Genera of Hyphomycetes. – Univ. Alberta Press.

COX, D.E. (1976). A new homobasidiomycete with anomalous basidia. – Mycologia 68: 481–510.

DONK, M.A. (1958). The generic names proposed for Hymenomycetes - VIII. Auriculariaceae, Septobasidiaceae, Tremellaceae, Dacrymycetaceae. – Taxon 7: 164–178, 193–207, 236–250.

DYKSTRA, M.J. (1974). Some ultrastructural features in the genus *Septobasidium*. – Can. J. Bot. 52: 971–972.

ELLIS, M.B. (1971). Dematiaceous Hyphomycetes. – Commonwealth Myc. Inst., Kew.

GOTTSCHALK, M. (1985). Untersuchungen zur Phylogenie der Basidiomyceten anhand des Vergleichs der Nucleotidsequenzen. – Diss. Univ. Tübingen.

— & P. BLANZ (1984). Highly conserved 5S ribosomal RNA sequences in four rust fungi and atypical 5S rRNA secondary structure in *Microstroma juglandis*. – Nucl. Acids Res. 12: 3951–3958.
— & — (1985). Untersuchungen an 5S ribosomalen Ribonukleinsäuren als Beitrag zur Klärung von Systematik und Phylogenie der Basidiomyceten. – Z. Mykol. 51: 205–243.
HEATH, I.B. (1981). Nucleus-associated organelles in fungi. – Int. Rev. Cyt. 69: 191–221.
— & M.C. HEATH, (1976). Ultrastructure of mitosis in the cowpea rust fungus *Uromyces phaseoli* var. *vigneae*. – J. Cell Biol. 70: 592–607.
HORI, H. & S. OSAWA (1987). Origin and evolution of organisms as deduced from 5S ribosomal RNA sequences. – Mol. Biol. Evol. 4: 445–472.
HUGHES, S.J. (1958). Revisiones Hyphomycetum aliquot cum appendice de nominibus rejiciendis. – Can. J. Bot. 36: 727–836.
HUYSMANS, E., E. DAMS, A. VANDENBERGHE & R. DE WACHTER (1983). The nucleotide sequences of the 5S rRNAs of four mushrooms and their use in studying the phylogenetic position of Basidiomycetes among the eukaryotes. – Nucleic Acids Res. 11: 2871–2880.
— & R. DE WACHTER (1986). The distribution of 5S ribosomal RNA sequences in phenetic hyperspace. Implications for eubacterial, eukaryotic, archaebacterial and early biotic evolution. – Endocyt. C. Res. 3: 133–155.
JUEL, H.O. (1898). *Stilbum vulgare* TODE. Ein bisher verkannter Basidiomycet. – Bih. Kongl. Svenska Vetensk.-Akad. Handl. 24: 1–15.
KENDRICK, B. & R. WATLING (1979). Mitospores in Basidiomycetes. In: KENDRICK, B. The whole fungus. Vol 2. – Nat. Mus. Nat. Sci. Canada & Kanaskis Foundation. pp. 473–545.
KHAN, S.R., & J.W. KIMBROUGH (1980). Ultrastructure and the taxonomy of *Eocronartium*. – Can. J. Bot. 58: 642–647.
KIMURA, M. (1968). Evolutionary rate at the molecular level. – Nature 217: 624–626.
— (1969). The rate of molecular evolution considered from the standpoint of population genetics. – Proc. Natl. Am. Sci. 63: 1181–1188.
— (1980). A simple method for estimating evolutionary rates of base substitutions through comparative studies of nucleotide sequences. – J. Mol. Evol. 16: 111–120.
— (1983). The neutral theory of molecular evolution. – Cambridge Univ. Press, Cambridge.
KING, J.L. & T.H. JUKES (1969). Non-Darwinian evolution. – Science 164: 788–798.
KROPP, B.R. & M.E. CORDEN (1986). Morphology and taxonomy of *Pachnocybe ferruginea*. – Mycologia 78: 334–342.
LANGLEY, C.H. & W.M. FITCH (1974). An examination of the constancy of the rate of molecular evolution. – J. Mol. Evol. 3: 161–177.
LIU, W. & R.N. NAZAR (1983). Structure of a ribosomal 5S RNA from a mushroom, *Coprinus cinereus*. – Biochem. Biophys. Res. Comm. 116: 148–153.
MARQUARDT, H. (1962). Der Feinbau von Hefezellen im Elektronenmikrsokop. I. Mitt.: *Rhodotorula rubra*. – Z. Naturf. 17b: 295–298.
MARTIN, G.W. (1952). Revision of the North Central Tremellales. – State Univ. Iowa Stud. Nat. Hist. 19: 1–122.
MCCULLY, E.K. & C.F. ROBINOW (1972.a). Mitosis in heterobasidiomycetous yeasts. I. *Leucosporidium scottii (Candida scottii)*. – J. Cell Sci. 10: 857–881.
— & — (1972.b). Mitosis in heterobasidiomycetous yeasts. II. *Rhodosporidium* sp. *(Rhodotorula glutinis)* and *Aessosporon salmonicolor (Sporobolomyces salmonicolor)*. – J. Cell Sci. 11: 1–31.
MCLAUGHLIN, D.J. (1979). Ultrastructure of the hymenium of *Auricularia polytricha*. – Mushsroom Sci. 10: 219–229.
— (1980). Ultrastructure of the metabasidium of *Auricularia fuscosuccinea*. – Amer. J. Bot. 67: 1225–1235.

Möller, A. (1895). Protobasidiomyceten. – Bot. Mitt. Trop. 8: 1–152.
Moore, R.T. (1978). Taxonomic significance of septal ultrastructure with particular reference to the jelly fungi. – Mycologia 70: 1007–1024.
Morris, E.F. & D.E. Finley (1965). A new genus based on *Isaria palmae*. – Mycologia 57: 483–485.
Nishikawa, K. & S. Takemura (1974). Nucleotide sequences of 5S rRNA from *Torulopsis utilis*. – FEBS Lett. 40: 106–109.
Oberwinkler, F. & R.J. Bandoni (1982.a). A taxonomic survey of the gasteroid, auricularioid Heterobasidiomycetes. – Can. J. Bot. 60: 1726–1750.
— & — (1982.b): *Atractogloea:* a new genus in the Hoehnelomycetaceae (Heterobasidiomycetes). – Mycologia 74: 634–639.
— , — , P. Blanz, & L. Kisimova-Horovitz (1983). *Cystofilobasidium:* a new genus in the Filobasidiaceae. – System. Appl. Microbiol. 4: 114–122.
— , — , — , G. Deml & L. Kisimova-Horovitz (1982). Graphiolales: Basidiomycetes parasitic on palms. – Pl. Syst. Evol. 140: 251–277.
O'Donnell, K. & D.J. McLaughlin (1981.a). Ultrastructure of meiosis in the hollyhock rust fungus *Puccinia malvacearum*. I. Prophase I – Prometaphase I. – Protoplasma 108: 225–244.
— & — (1981.b). Ultrastructure of meiosis in the hollyhock rust fungus *Puccinia malvacearum*. II. Metaphase I – Telophase I. – Protoplasma 108: 245–263.
— & — (1981.c). Ultrastructure of meiosis in the hollyhock rust fungus *Puccinia malvacearum*. III. Interphase I – Interphase II. – Protoplasma 108: 265–288.
— & — (1984.a). Ultrastructure of meiosis in *Ustilago maydis*. – Mycologia 76: 468–485.
— & — (1984.b). Postmeiotic mitosis, basidiospore development, and septation in *Ustilago maydis*. – Mycologia 76: 486–502.
Paden, J.W. (1978). Morphology, growth in culture, and conidium formation in *Poronia pileiformis*. – Can. J. Bot. 56: 1774–1776.
Patton, A.M. & R. Marchant (1978). A mathematical analysis of dolipore/parenthesome structure in Basidiomycetes. – J. Gen. Microbiol. 109: 335–349.
Prusso, D.C. & K. Wells (1967). *Sporobolomyces roseus*. I. Ultrastructure. – Mycologia 59: 337–348.
Reynolds, E.S. (1963). The use of lead citrate at high pH as an electron opaque stain in electron microscopy. – J. Cell Biol. 17: 208–212.
Saccardo, P.A. (1886). Sylloge Fungorum. 4: 578.
Schroeter, J. (1887). Die Pilze Schlesiens. – In: Cohn, F. Kryptogamenflora Schlesiens 3: 257–384.
Sebald, F.R. (1977). Feinstrukturstudien zur Ontogenie von Arten der Uredinales und verwandter Basidiomyceten. – Diss. Univ. Tübingen.
Shear, R.L. & B.O. Dodge (1925). The life history of *Pilacra faginea* (Fr.) R.Br. – J. Agric. Res. (Wash. D.C.) 30: 407–417.
Smith, D.G. & R.G. Coss (1984). Calibrating the molecular clock: estimates of ground markers. – Mol. Biol. Evol. 1: 249–259.
Spurr, A.R. (1969). A low viscosity epoxid embedding medium for electron microscopy. – J. Ultrastr. Res. 26: 31–43.
Subramanian, C.V. & K. Natarajan (1975). Two new Hyphomycetes from India. – Mycologia 67: 1211–1217.
— & — (1977). On the genera *Agaricostilbum* and *Amerobotryum*. – Mycologia 69: 1224–1226.
Tallasch, H. & H. Jahn (1970). *Phleogena faginea* (Fr.) Link im Naturschutzgebiet "Hasbruch" bei Bremen. – Westfäl. Pilzbr. 8: 31–35.
Taylor, J.W. & K. Wells (1979). A light and electron microscopic study of mitosis in *Bullera alba* and the histochemistry of some cytoplasmic substances. – Protoplasma 98: 31–62.

THYAGARAJAN, T.R., S.F. CONTI & H.B. NAYLOR (1962). Electron microscopy of *Rhodotorula glutinis*. – J. Bacteriol. 83: 381–394.

TU, C.C., & J.W. KIMBROUGH (1978). Systematics and phylogeny of fungi in the *Rhizoctonia* complex. – Bot. Gaz. 139: 454–466.

WALKER, W.F. (1984). 5S rRNA sequences from Atractiellales, and basidiomycetous yeasts and fungi imperfecti. – Syst. Appl. Microbiol. 5: 352–359.

— & W.F. DOOLITTLE (1982). Redividing the Basidiomycetes on the basis of 5S rRNA sequences. – Nature 299: 723–724.

— & — (1983). 5S rRNA sequences from eight Basidiomycetes and fungi imperfecti. – Nucl. Acids Res. 11: 7625–7630.

WEESE, J. (1920). Beitrag zur Morphologie und Systematik einiger Auriculariineengattungen. – Ber. Deutsch. Bot. Ges. 37: 512–519.

WELLS, K. (1977). Meiotic and mitotic divisions in the Basidiomycotina. – In: ROST, T.L. & E.M. GIFFORD, Jr. (eds.). Mechanisms and control of cell division. Stroudsburg, Pennsylvania. Dowden, Hutschinson & Russ, Inc. pp. 337–374.

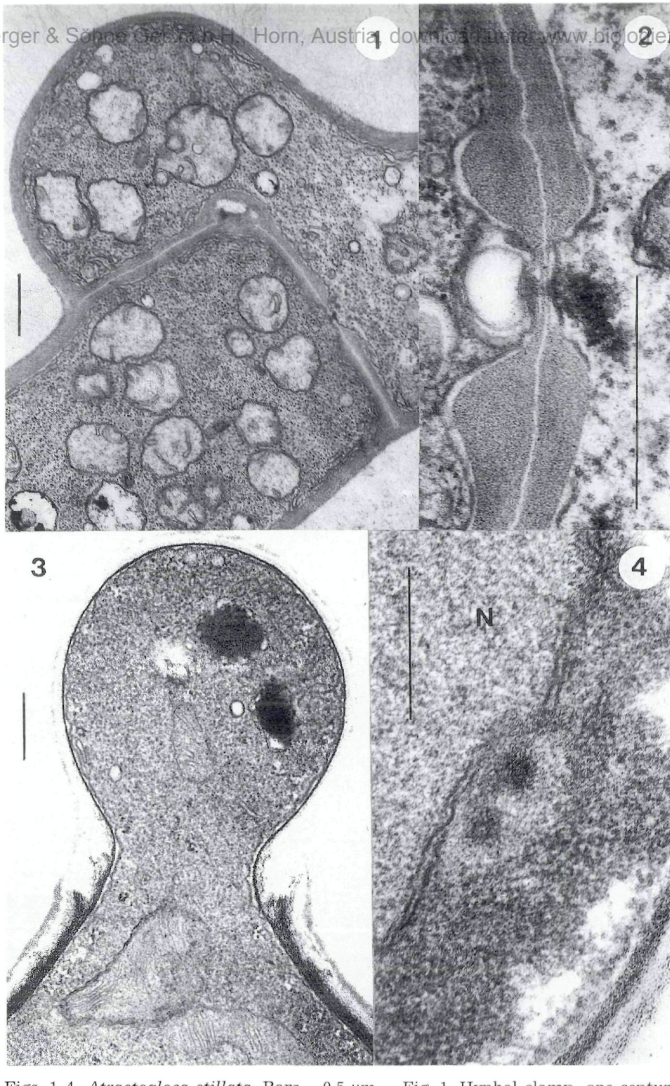
WOJEWODA, W. (1977). Grzyby (Mycota). – Flora Polska 8: 1–334.

WOLTERS, J. (1987). Phylogenie der Organismen und Zellorganellen – kladistische Strukturanalysen ribosomaler RNAs. – Diss., Freie Universität Berlin.

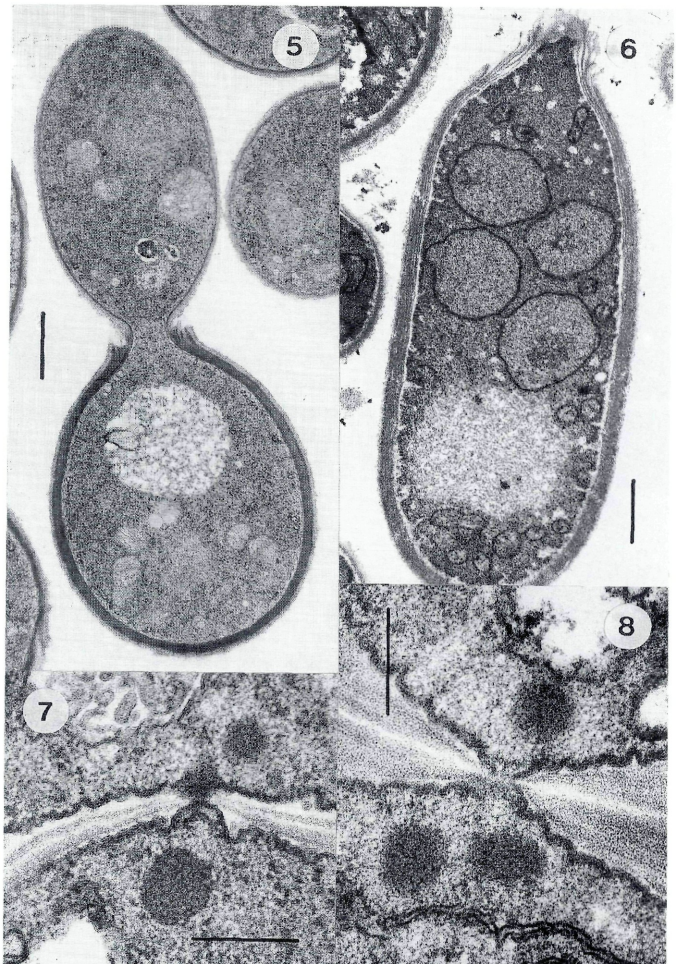
— & V.A. ERDMANN (1988). Compilation of 5S rRNA gene sequences. – Nucl. Acids Res. (Suppl.) 16: 1–69.

WRIGHT, J.E. (1970). *Agaricostilbum*, a new genus of Deuteromycetes on palm spathes of Argentina. – Mycologia 62: 679–682.

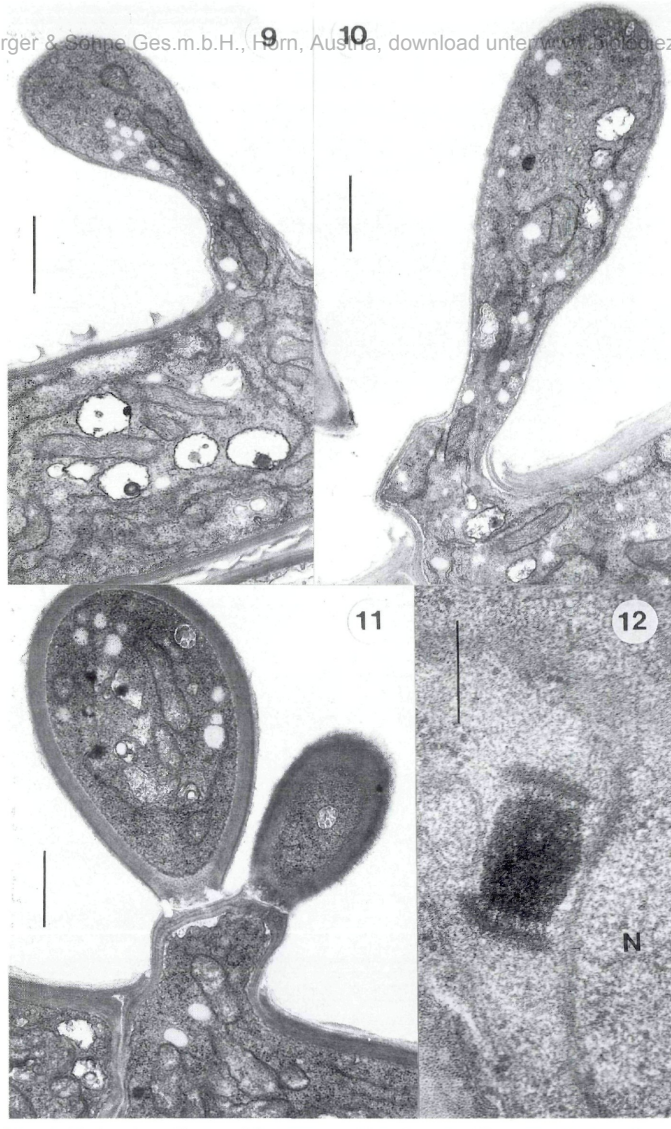
— , R.J. BANDONI & F. OBERWINKLER (1981). *Agaricostilbum:* an auricularioid basidiomycete. – Mycologia 73: 880–886.



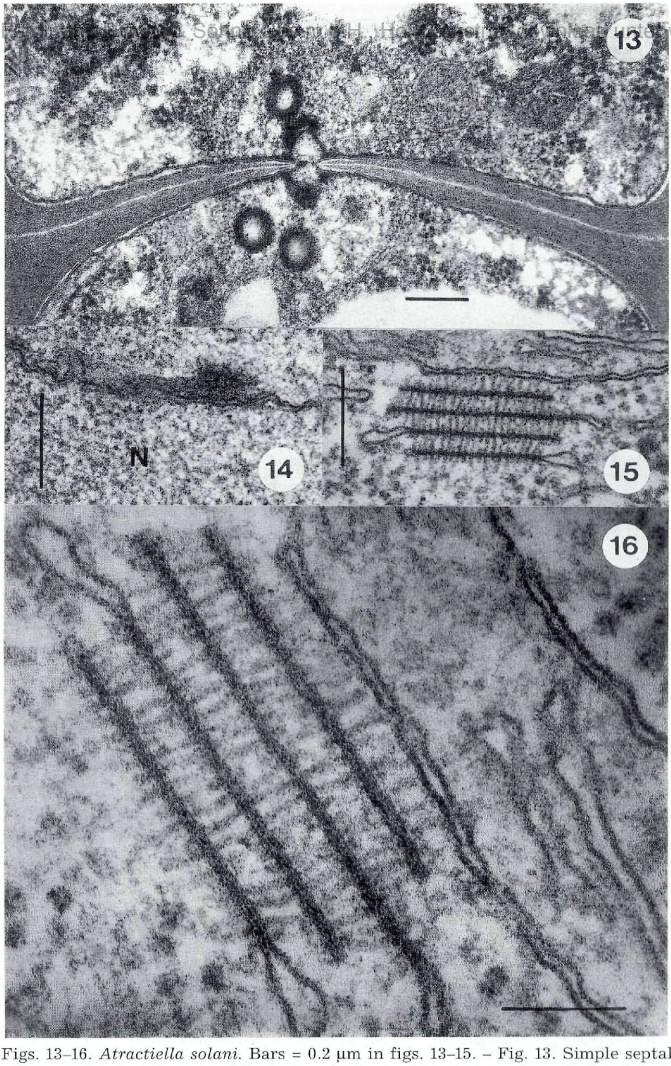
Figs. 1–4. *Atractogloea stillata*. Bars = 0.5 µm. – Fig. 1. Hyphal clamp, one septum showing simple pore. – Fig. 2. Detail of septal pore; note septal swelling close to pore opening. – Fig. 3. Yeast budding, showing scars of mother cell. – Fig. 4. Diglobular mitotic interphase SPB adjacent to nuclear envelope. N = Nucleus.



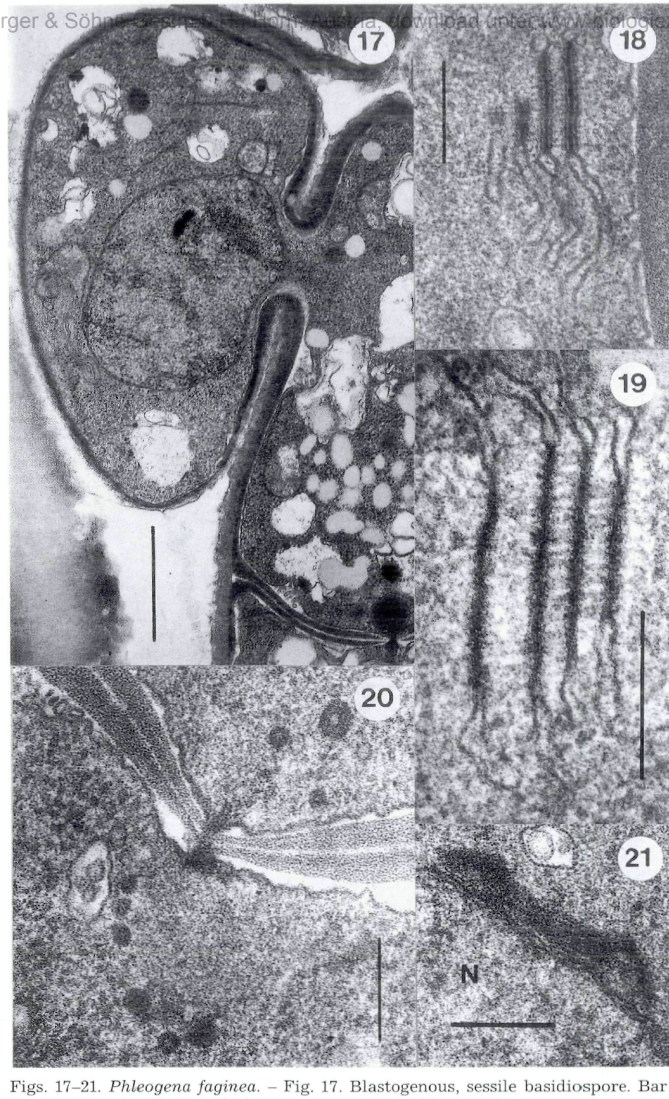
Figs. 5–8. *Agaricostilbum pulcherrimum.* – Fig. 5. Yeast budding; note multilayered scar of mother cell. Bar = 1 µm. – Fig. 6. Yeast cell with 4 nuclei; note budding locus. Bar = 1 µm. – Figs. 7, 8. Simple septal pores associated with electron dense bodies. Bars = 0.2 µm.



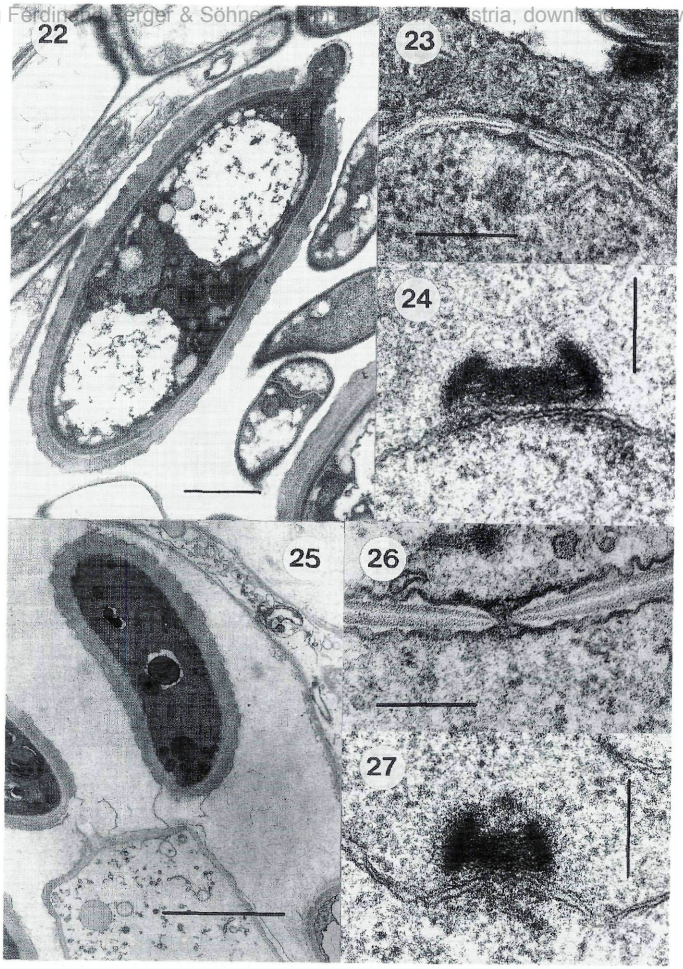
Figs. 9–12. *Agaricostilbum pulcherrimum*. – Figs. 9–11. Basidiospore ontogeny. Bars = 1 µm. – Figs. 9, 10. Young basidia. – Fig. 11. Two successive basidiospores developing from from the basidial cell extension. – Fig. 12. Section of prophase I SPB, showing midpiece and two marginal discs oriented vertical to nuclear envelope. N = nucleus.



Figs. 13–16. *Atractiella solani*. Bars = 0.2 µm in figs. 13–15. – Fig. 13. Simple septal pore associated with vesicle-like bodies. – Fig. 14. Mitotic interphase SPB in longitudinal section, showing midpiece and lateral discs. – N = Nucleus. – Figs. 15, 16. Symplechosomes: dictyosome-like membrane complexes interconnected with electron dense bars. Note flaring of double membranes at periphery outside of interconnected areas. Bar in fig. 16 = 0.1 µm.

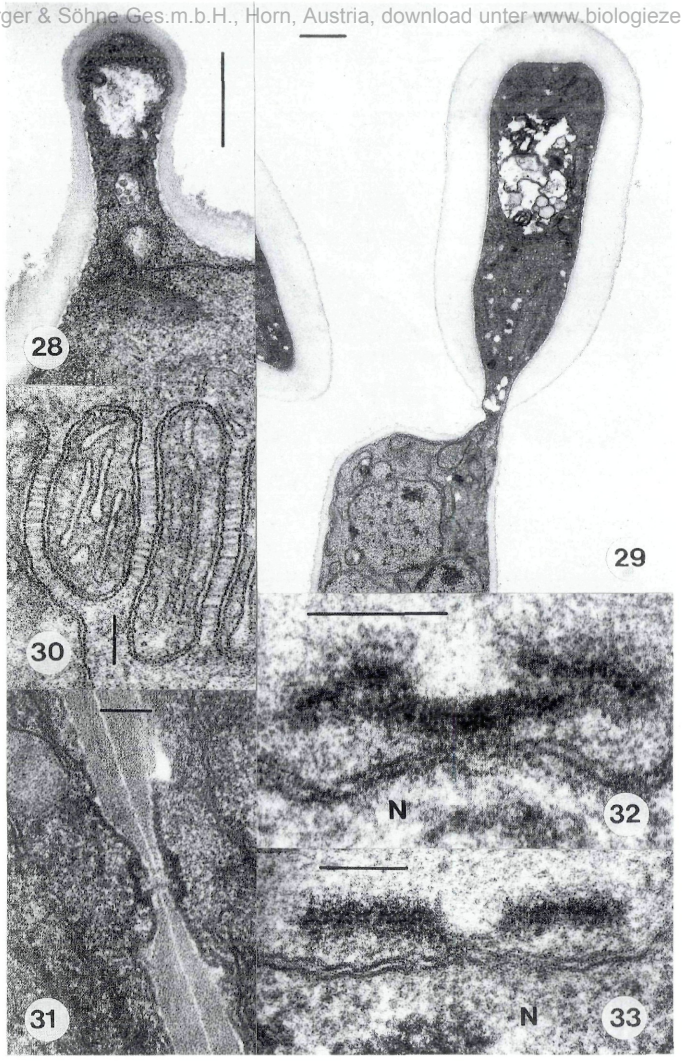


Figs. 17–21. *Phleogena faginea*. – Fig. 17. Blastogenous, sessile basidiospore. Bar = 1 µm. – Figs. 18, 19. Symplechosomes: dictyosome-like membrane complexes interconnected with electron dense bars. Note flaring of double membranes at periphery outside of interconnected areas. – Fig. 20. Simple septal pore; note vesicle-like bodies at some distance from pore opening. – Fig. 21. Longitudinal section of prophase I SPB, showing conspicuous midpiece and obliquely inserted, marginal discs. Bars in figs. 18–21 = 0.2 µm.



Figs. 22–24. *Stilbum vulgare*. – Fig. 22. Germinating basidiospore; note rough outer cell wall layer. Bar = 1 µm. – Fig. 23. Simple septal pore occluded by electron dense material. Bar = 0.2 µm. – Fig. 24. Longitudinal section of prophase I SPB, showing midpiece and lateral discs. Bar = 0.2 µm.

Figs. 25–27. *Chionosphaera apobasidialis*. – Fig. 25. Apex of old basidium with two basidiospores still attached. Note thick, rough spore wall. Bar = 1.6 µm. – Fig. 26. Simple septal pore. Bar = 0.2 µm. – Fig. 27. Longitudinal section of prophase I SPB, showing small midpiece and conspicuous, electron dense lateral discs. Bar = 0.2 µm.



Figs. 28–33. *Pachnocybe ferruginea*. – Figs. 28, 29. Basidiospore development. – Fig. 28. Initial stage of spore ontogeny. Bar = 0.5 µm. – Fig. 29. Basidiospore, symmetrically attached to short sterigma. Bar = 1 µm. – Fig. 30. Mitochondria interconnected by electron dense bars. Bar = 0.1 µm. – Fig. 31. Simple septal pore. Bar = 0.1 µm. – Figs. 32, 33. Longitudinal sections of SPBs. Bars = 0.1 µm. N = nucleus. – Fig. 32. Prophase I SPB. – Fig. 33. Interphase II SPB.