

Heat-tolerant fungi and applied research: more data on pseudothermophilic taxa

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Abstract – Information on presumed marked heat tolerance levels of about 20 fungal taxa, referred to as pseudothermophilic fungi, is critically reviewed. This assessment also includes individual nomenclatural status and correct taxonomic position. For several, the assumption conveyed by their specific epithet *thermophila* (also *thermophilus*, *thermoideum* or *thermosus*) appeared misleading since the fungi under consideration proved to be strict thermotolerant organisms. This situation concerns *Salilagenidium thermophilum* (oomycete), the zygomycetes *Absidia idahoensis* var. *thermophila*, *Mucor thermophilus*, *Rhizopus pseudochinensis* var. *thermosus* (now a synonym of *R. microsporus* var. *chinensis*) and *R. thermosus* (now a synonym of *R. oryzae*). Similar cases are the two ascomycetes *Achaetomium thermophilum* (a present synonym of *A. macrosporum*) and *Sordaria thermophila* and the following anamorphic fungi: *Calcarisporiella thermophila*, *Gilmaniella thermophila* (regarded as a synonym of *G. macrospora*), *Zalerion thermophylli* (a synonym of *Z. maritima*) and *Geotrichum candidum* var. *thermoideum* (of uncertain taxonomic position).

Few other taxa were erroneously qualified as possessing thermophilic attributes: *Rhizopus homothallicus* and the anamorphic fungi *Stachybotrys ramosa* (presently renamed *S. thermotolerans*), *Bactrodesmium fusiforme* (generic affinity regarded as doubtful), *Trichocladium heterosporum* (also excluded from the genus) and the type species of *Phaeodactylella*, *P. lignicola*, of unclear taxonomic position. *Aspergillus thermomutatus*, the anamorph of *Neosartorya pseudofischeri*, is a strict thermotolerant. The lichenized ascomycete *Acarospora thermophila* has a species epithet referring to the marked xerothermic conditions of its original locality. Finally *Nodulisporium thermoroseum* is a 'ghost name'. Additional cases of similar pseudothermophilic fungi of other systematic groups will be reviewed in subsequent notes.

INTRODUCTION

The ability to grow at high temperatures is an uncommon feature amongst fungi (Dix & Webster, 1995). Heat tolerant fungi may be either obligate thermophiles or simple thermotolerants. Following Cooney & Emerson's (1964) definitions of these subgroups, a thermophilic fungus develops optimum growth at a temperature of about 50°C and minimum growth at or above 20°C; a thermotolerant fungus is able to develop well below 20°C with a maximum growth temperature value at about 50°C.

The segregation of heat tolerant fungi into thermophilic and thermotolerants *sensu* Cooney & Emerson has not always been consistent in

modern literature on pure and applied research. The outcome is that several fungi able to develop at 15°C were repeatedly referred to as possessing thermophilic attributes (Johri *et al.*, 1999; Maheshwari *et al.*, 2000; Mehrotra, 1985). A clear distinction between both subgroups of heat tolerant fungi is however essential to optimize investigations of temperature-dependent physiological processes.

A modern revision of the systematics of obligate thermophiles paved the way for a clear separation of fungi able to develop at high temperatures (Mouchacca, 1997, 1999). Mouchacca (2000a) critically reviewed heat tolerance levels of fungal names occasionally or commonly qualified as thermophiles mostly in the literature on applied research; the review also considered their nomenclatural status and a relevant update of a taxonomic nature. The assessment of 130 names of such pseudothermophilic fungi produced a list of 30 taxonomically correct binomials of strictly thermotolerant taxa. Further interest in thermotolerant fungi produced a list of an additional fifty species for which simple heat tolerance levels have also been disclosed, mostly in recent times (Mouchacca, in press).

The material of the present contribution gradually accumulated in the course of our monograph on thermophilic fungi. About 20 pseudothermophilic taxa are here considered on the basis of their reportedly marked to less marked heat tolerance levels; for some, such an ability appears to be implied in their particular specific or varietal epithets. This document is an addition to the former publication on thermotolerant species erroneously reported as possessing thermophilic attributes (Mouchacca, 2000 a). Taxa here surveyed belong to the following systematic groups: Oomycetes, Zygomycetes, Ascomycetes and anamorphic fungi. Similar cases of Holobasidiomycetous fungi shall be dealt with in a separate note.

LIST OF TREATED SPECIES

Taxa treated are introduced according to broad taxonomic categories. Within these, genera are listed alphabetically. For each taxon, the full bibliographic reference of the original binomial is given. For invalid names the relevant article of the International Code of Botanical Nomenclature is specified (Greuter *et al.*, 2000). All efforts were undertaken to re-appraise the taxonomic status of the pseudothermophilic taxa considered. For names having undergone taxonomic changes the latest taxonomically accepted binomial is provided along with the corresponding original reference. Additional synonyms are often reported and the source of the relevant taxonomic document is specified.

Available information on heat tolerance levels is also specified. In the case of species or varietal epithets appearing misleading with respect to this ecological feature, reasons for this are underlined based on published data of ecological nature.

The following symbols and abbreviations are used in the text. ? basionym; =: synonym and ICBN: International Code of Botanical Nomenclature. For the cited culture collections the following acronyms were used. ATCC: American Type Culture Collections, USA; CBS: Centraalbureau voor Schimmelfcultures, The Netherlands; IMI: International Mycological Institute (now CAB International), United Kingdom. Finally author's of fungal names were abbreviated following Kirk & Ansell (2003, electronic version).

OOMYCETE***Salilagenidium thermophilum* (K. Nakam., Miho Nakam., Hatai & Zafran) M.W.**

Dick – *Straminipilous Fungi. Systematics of the peronosporomycetes including accounts of the marine straminipilous protists, the plamodiophorids and similar organisms*, Doordrecht, Netherlands, etc: Kluwer Academic Publishers: 315. 2001.

≡ *Lagenidium thermophilum* K. Nakam., Miho Nakam., Hatai & Zafran – *Mycoscience* 36: 400. 1995.

This oomycete was found to infect the eggs and larvae of the mangrove crab, *Scylla serrata* Forsskäl, in Bali, Indonesia. Strains proved to represent a new species of *Lagenidium* Schenk having a unique discharge process. The reported growth temperatures range from 15-45°C with an optimum between 30 and 40°C. It follows that the fungus is simply a thermotolerant organism.

The taxonomic position of known *Lagenidium* species was recently reappraised by Dick (2001) in his general revision of the Straminipilous Fungi. Species of *Lagenidium* are parasitic in marine Crustacea and Mollusca and can sometimes be grown in the laboratory. Presently they comprise two teleomorphic and three holoanamorphic members. Dick provided evidence that both subgroups are congeneric and would thus better be accommodated in the new genus *Salilagenidium* M.W. Dick (etymology: *Lagenidium*-like fungi from saline habitats).

ZYGOMYCETES***Absidia idahoensis* var. *thermophila* G.Q. Chen & R.Y. Zheng – *Mycotaxon* 69: 174. 1998.**

This new variety of *Absidia idahoensis* Hesselt., M.K. Mahoney & S.W. Peterson was introduced for a soil-borne Chinese strain still able to develop growth at 48°C (Chen & Zheng, 1998) instead of the maximum growth temperature value of 37°C reported for the species (Hesseltine *et al.*, 1990). The new variety is thus a thermotolerant organism in Cooney & Emerson's sense.

Among known species of *Absidia* Tiegh., only *A. corymbifera* (Cohn) Sacc. & Trotter is markedly thermotolerant: optimal development is disclosed at 35°C with a maximum at 48-52°C (see Domsch *et al.*, 1980). These values match cardinal growth temperatures reported for the variety *thermophila*. A comparison of representative cultures of *A. corymbifera*, *A. idahoensis* and the new variety should thus be undertaken to elucidate their respective taxonomic positions.

Absidia corymbifera is known as the most common causal agent of mucormycosis of the lungs, nasal sinuses, cornea and other organs in man and warm-blooded animals, and also of mycotic abortion in cattle (Ellis D.H., 1998). It was isolated from a variety of substrata including soils of different types (Domsch *et al.*, 1980).

***Mucor thermophilus* R. Prakash & A.K. Sarbhoy – *Zentralblatt für Mikrobiologie* 148: 531. 1993; *nom. inval.*, Arts. 37.1 & 37.5 ICBN.**

According to Prakash & Sarbhoy (1993) 'the new species is similar to *Mucor piriformis* A. Fish. in certain characters. However, it differs from this in not bearing two types of sporangiophores, which are sometimes branched. The sporangia are black at maturity rather than brownish grey. It differs from *Mucor piriformis* in that sporangia may reach 350 µm in length. In the present species, sporangiospores are hyaline and larger than in *Mucor piriformis*, extending up to

12 µm. The species in question is able to grow and sporulate at 30°C and above (37°C) whereas the other two species failed to grow at 30°C. These differences are, therefore, sufficient enough for creating a new species. The specific epithet has been given on the thermotolerant nature of the species. The culture has been isolated from fruits of *Carica papaya* L., Delhi, and is deposited in the Indian Type Culture Collection as a cultotype (*Mucor thermophilus*) ITCC No. 3364'.

Cardinal growth temperatures of *Mucor thermophilus* were not determined by the Indian authors. Until these data become available the fungus should better be regarded as a thermotolerant in Cooney and Emerson' sense. Thermotolerant species of *Mucor* Fresen. *sensu lato* that can develop above 40°C are now being continuously described (Schipper & Samson, 1994). A modern taxonomic account of the genus is presently hardly needed.

***Rhizopus homothallicus* Hesselt. & J.J. Ellis** – *Mycologia* 53: 419. 1961; Schipper & Stalpers – *Studies in Mycology, Baarn* 25: 33. 1984.

This thermotolerant species of *Rhizopus* Ehrenb. was again recently featured as a thermophilic fungus by Mateos Diaz *et al.* (2005) in the course of their search for thermostable lipases suitable for biocatalysis applications. The relevant strain was isolated from a coconut sample collected in the Mexican tropics. Optimal lipase production occurred at 40°C under conditions of solid state fermentation; this optimum was achieved at 30°C in conditions of liquid state fermentation.

The species still shows good growth at 46°C but none at 50°C (Hesseltine & Ellis J.J., 1961). The recent relevant description provided by Schipper & Stalpers (1984) was based on colonies of the type strain incubated at 30°C; but again the minimum and optimal growth values were not defined. The ecology of this Zygomycete awaits to be documented.

Classification and identification of *Rhizopus* species has commonly largely been based on morphology. Schipper (1984) divided the genus into three groups: the *R. stolonifer* group, the *R. oryzae* group and the *R. microsporus* group according to characteristics of the sporangial apparatus and growth temperatures. *Rhizopus homothallicus* is the sole member of the *R. microsporus*-group to produce homothallic zygospores at 30°C on yeast extract agar. Members of this group show visible growth at 45°C but such is not the case for the *Rhizopus stolonifer*-group and *R. oryzae* Went & Prins. Geerl. Based on the ability to develop at 45°C, Ellis D.H. (1998) regarded all members of the *R. microsporus*-group as thermophiles. However, this concept of thermophily among fungi was subsequently refuted (Mouchacca, 2000 a).

Recently Liou *et al.* (2007) conducted a phylogenetic analysis involving strains of all *Rhizopus* species. The analysis focused on the D1/D2 region of LSU rDNA sequences. The results yielded a phylogram with rather four well-supported clades. Clade A of the phylogram comprised *R. azygosporus*, *R. caespitosus*, *R. homothallicus*, *R. microsporus*, and *R. schipperae*. The sporangial apparatus and growth temperatures of these species are thus in accordance with diagnostic characters of the *R. microsporus* group as formerly delimited by Schipper (1984).

***Rhizopus microsporus* var. *chinensis* (Saito) Schipper & Stalpers** – *Studies in Mycology, Baarn* 25: 31. 1984.

≡ *Rhizopus chinensis* Saito – *Zentralblatt für Bakteriologie, Parasitenkunde, Infektionskrankheiten und Hygiene*. Abteilung 2, 13: 156. 1904.

= *Rhizopus pseudochinensis* var. *thermosus* Y. Takeda – *Journal of the agricultural chemical Society of Japan* 11: 845. 1935; *nom. inval.*, Art. 36.1 ICBN.

Two other synonyms, namely *Rhizopus bovinus* J.F.H. Beyma and *Rhizopus chinensis* var. *liquefaciens* Y. Takeda, are also provided by Schipper & Stalpers (1984).

Rhizopus pseudochinensis M. Yamaz. [*Journal of the Scientific Agricultural Society of Tokyo* 193: 996. 1918] was regarded by Schipper (1984) as a synonym of *R. oryzae* Went & Prins. Geerl. displaying maximum colony growth at 45°C. Later on Prakash & Sarbhoy (1993) introduced the combination *Rhizopus microsporus* var. *pseudochinensis* (M. Yamaz.) R. Prakash & A.K. Sarbhoy because 'the smooth-walled sporangia and their size as well as that of the sporangiospores should be given more emphasis'. They argued that Schipper (1984) favoured the synonymy on 'the basis of sporangial size and nature of spores'. However, this synonymy also integrates the mating behaviour of the type strain of *Rhizopus pseudochinensis*.

Schipper & Stalpers (1984) proposed var. *chinensis* because all strains labelled *Rhizopus chinensis* were able to mate with tester strains of *R. microsporus* var. *microsporus*. Positive mating among tested strains is regarded as conclusive evidence of taxonomic affiliation at the species level in zygomycetous taxa. The two varieties differ, however, in that the sporangiospores are distinctly angular-ellipsoidal and striate, up to 6.5 µm, in var. *microsporus* but subglobose to slightly angular, up to 7.5 µm diam., and rather homogenous in var. *chinensis*.

Representative strains of var. *chinensis* still grow well at 45°C but sporangia do not mature; no growth occurs at 50°C. Mehrotra & Basu (1980) isolated a strain referable to *Rhizopus chinensis* from Indian sawdust and regarded it as thermophilic. This record was erroneously reported later on as *Rhizomucor chainensis* by Subrahmanyam (1999: 32).

Rhizopus pseudochinensis var. *thermosus*, type strain CBS 394.34, differs by its moderate growth at 50°C; the columellae are conical-ellipsoidal with sporangiospores being subglobose and 6-9 µm in diam; in older slant cultures azygospores occur (Schipper & Stalpers, 1984). CBS 394.34 and all available strains of *Rhizopus chinensis* mated, however, with either of the tester strains of *R. microsporus* var. *microsporus* CBS 699.68 (+) and CBS 700.68 (-). The distinction of the var. *thermosus* on the slightly higher maximum temperature is accordingly not warranted.

Data of a recent phylogenetic analysis of *Rhizopus* strains conducted by Liou *et al.* (2007) concluded the composition of the *R. microsporus* group as defined by morphological characteristics was relatively well supported when based on LSU rDNA partial sequences. The group is part of Clade A of the resulting phylogram also comprising *R. azygosporus*, *R. caespitosus*, *R. homothallicus* and *R. schipperae*. Furthermore and based on results of matings tests, Schipper & Stalpers (1984) have reduced to distinct varieties many species of the *R. microsporus* group. This decision was subsequently substantiated by DNA complementary studies undertaken by Ellis J.J. (1986). However, the new phylogram places *R. azygosporus* in the *R. microsporus* group, indicating the taxonomic status of *R. azygosporus* needs further investigation.

***Rhizopus oryzae* Went & Prins. Geerl.** – *Verhandelingen der Koninklijke Nederlandse Akademie van Wetenschappen Afd. Naturkunde*, Sect. 2, 4: 16. 1895; Schipper – *Studies in Mycology, Baarn* 25: 13. 1984.

? *Rhizopus arrhizus* A. Fish., in *Rabenhorst – Kryptogamenflora* 1: 233. 1892.

= *Mucor arrhizus* (A. Fish.) Hagem – *Untersuchungen über norwegische Mucorineen*: 37. 1907, 1908.

= *Rhizopus thermosus* Yosh. Yamam. – *Journal of the Society of Agricultural Forestry, Sapporo* 17: 493. 1925.

For further synonyms, see Schipper (1984).

Up to 1984, there was a general agreement that the identity of *Rhizopus arrhizus* was somewhat doubtful and the species is perhaps best considered as based on an atypically developed strain of *R. oryzae* (Schipper, 1984; Scholer, 1970). This consensus favoured the selection of the specific epithet *oryzae* for the relevant zygomycete despite the fact the binomial *Rhizopus oryzae* had been introduced after the name *R. arrhizus*. Schipper (1984) also suggested that all strains of the *R. oryzae* group are synonymous with *R. oryzae*. Such included *Rhizopus thermosus* (Schipper, 1984).

Strains referable to *Rhizopus oryzae* grow in the temperature range 15-45°C, with an optimum around 27°C, a minimum at 6°C and a maximum at 45°C. Colonies are very fast growing at 27°C, reaching 5-8 mm in height. Key features include good growth at 40°C, sporangiophores often more than 1 mm in height, rhizoids with secondary branching and sporangia between 100 and 240 µm in diam. (see Domsch *et al.*, 1980). *Rhizopus thermosus* was proposed because it showed some growth at 45°C. A study of the type species CBS 405.51 (ATCC 11002, Japan, K. Kominami) as well as a careful analysis of the original description showed that just another *R. oryzae* was concerned (Schipper, 1984).

In 1985, Ellis J.J. proposed however to segregate the *R. oryzae* group into two taxa: *R. arrhizus* var. *arrhizus* and *R. arrhizus* var. *delemar*. *Amylomyces rouxii* was then reduced to *R. arrhizus* var. *rouxii* based on their DNA relatedness. However following Liou *et al.* (2007) the clade B of their newly established phylogram for species of *Rhizopus* includes *R. oryzae* and *A. rouxii*. But then the relevant strains of *R. oryzae* fall into two groups. These results thus support the taxonomic status of *R. oryzae* and *A. rouxii* as delimited by Ellis J.J. (1985). However, in this same Clade B, *A. rouxii* falls into the two groups of *R. oryzae* indicating it might be polyphyletic. Thus for resolving the taxonomic concept in the *R. oryzae* and *A. rouxii* complex, more isolates and studies have to be included.

Most strains of *Rhizopus oryzae* have been isolated as active agents in the production of oriental foods or alcoholic beverages in Indonesia, China and Japan. This thermotolerant zygomycete is, however, considered as the most common causative agent of zygomycosis, accounting for some 60% of the reported culture-positive cases, and nearly 90 % of those of the rhinocerebral form of infection. It also produces the ergot alkaloid agroclavine which is toxic to humans and animals (Ellis D.H., 1998).

Finally the two remaining clades of the newly established *Rhizopus* phylogram (Liou *et al.* 2007) concern the *R. stolonifer* group and the taxa with recurved sporangiophores. Clade C comprises *R. stolonifer* and *R. sexualis* and contain two subclades. One includes strains of *R. stolonifer* var. *stolonifer*, the other contains strains of *R. sexualis* var. *sexualis* and *R. sexualis* var. *americanus* with high support values. *R. sexualis* var. *americanus* has smaller sporangia, causing its recognition as a distinct variety. Clade D comprises the taxa with recurved sporangiophores and is well supported; most of these strains are currently classified as varieties of *R. stolonifer* by mating experiments (Schipper 1984).

ASCOMYCETES

Achaetomium macrosporum J.N.Rai & Wadhvani & J.P. Tewari – *Indian Phytopathology* 23: 54. 1970.

- = *Achaetomiella macrospora* (J.N. Rai, Wadhvani & J.P. Tewari) v. Arx – *Proceedings, Koninklijke Nederlandse Akademie van Wetenschappen. Series C*, 76: 292. 1973.
- = *Achaetomium fusisporum* J.N. Rai & H.J. Chowdhery [as ‘*fusisporus*’] – *Journal of the Indian botanical Society* 52: 310. 1973, 1974.
- = *Achaetomium thermophilum* M. Basu – *Current Science* 51: 524. 1982; ? *nom. inval.*, Art. 9.5; synonymy *vide* Cannon (1986).

The original living strain of *Achaetomium thermophilum* was isolated from leaf litter in Bhattni, Uttar Pradesh, India. It differed from *Achaetomium macrosporum* in being ‘thermophilic’, although no minimum growth temperature was indicated. Cannon (1986) examined a culture (IMI 292262) derived from the holotype. Growth and sporulation proved to be satisfactory at 25°C, indicating that the fungus is rather thermotolerant. Ascospores were also found to be uniporate rather than biporate as stressed in the protologue and thus matching those of *Achaetomium macrosporum*. Based on these observations, Cannon (1986) concluded that the ascomycete proposed by M. Basu is conspecific with *Achaetomium macrosporum*.

Species of *Achaetomium* J.N. Rai & J.P. Tewari – type species *A. globosum* J.N. Rai & J.P. Tewari – are known to be good thermotolerants (von Arx *et al.*, 1988). The concept of the genus is, however, still under debate. Cannon (1986) transferred the two Indian species introduced after *Achaetomium macrosporum*, namely *A. luteum* J.N. Rai & J.P. Tewari and *A. strumarium* J.N. Rai *et al.*, to *Chaetomium* Kuntze. He only retained in the genus *Achaetomium*, the type species, *A. macrosporum* and *A. marinum* H.J. Chowdhery & J.N. Rai.

Von Arx *et al.* (1988) disagreed with Cannon’s generic concept and with the disposition of described species. They only retained the three original Indian species and excluded *Achaetomium macrosporum* on the basis of it being possibly similar to *Chaetomium vitellinum* A. Carter or *C. megasporum* Sörgel ex Seth. This latter opinion was subsequently substantiated by Rodriguez *et al.* (2004), who introduced *Achaetomium umbonatum* K. Rodr., Stchigel & Guarro; they also transferred *Chaetomium irregulare* Sörgel to *Achaetomium*.

***Neosartorya pseudofischeri* S.W. Peterson** – *Mycological Research* 96: 549. 1992.
anamorph: *Aspergillus thermomutatus* (Paden) S.W. Peterson – *Mycological Research* 96: 549. 1992.

≡ *Aspergillus fischeri* var. *thermomutatus* Paden – *Mycopathologia Mycologia Applicata* 36: 161. 1968.

The variety *thermomutatus* was originally isolated from mouldy cardboard at the University of Victoria, British Columbia, Canada. It was regarded to differ from previously known varieties of *Aspergillus fischeri* Wehmer in having many vesicles set at an angle to the conidiophore when cultures were incubated at 22°C; cleistothecia, asci and ascospores develop at this temperature. The vesicles are, however, upright when cultures are incubated at 37°C, hence the specific epithet. The ascospores were regarded as being practically identical to those of *Neosartorya fischeri* (Wehmer) Malloch & Cain, having a reticulate ornamentation on the ascospore walls (Paden, 1968).

A second strain of the variety was obtained in the course of an autopsy of lesions of human neck vertebrae at the U.S. Centers for Disease Control, Atlanta, Georgia, USA; the fungal lesion was however not the immediate cause of death. Following Peterson (1992), closer examination of ascospores of either strains revealed the presence of ascospore wall ornaments shaped as triangular flaps, rather than the reticulate ridges of the original description.

Neosartorya pseudofischeri does not fit into *N. fischeri* because it lacks anastomosing ridges on the ascospore wall (Peterson, 1992). Colonies on malt agar incubated at 25°C produced abundant ascomata and limited numbers of conidial heads, so that the colony appearance is dominated by the ascomata. This incubation temperature did not however induce a high incidence of conidiophores sharply bent below the vesicle. Conidia and ascomata were produced at 25 and 37°C, with better production of conidia and fewer ascomata at the latter temperature. Other cardinal growth temperatures were not assessed for this thermotolerant fungus.

Strains of *Aspergillus fumigatus* Fresen. can be clearly differentiated from *Neosartorya pseudofischeri* by the production of ascomata in culture (Petersen, 1992). This statement was, however, recently questioned by Balajee *et al.* (2005) based on their re-examination of three clinical strains maintained as *Aspergillus fumigatus*: FH240, FH242 and FH274. Molecular data suggested these isolates belong to *Neosartorya pseudofischeri*: FH274 and FH240 were 100% homologous while FH242 was 99% homologous to the sequence of *N. pseudofischeri*. Strains FH240 and FH242 failed however to produce the confirmatory asci and ascocarps on several media at 25°C and 37°C, and were thus classified as *Aspergillus thermomutatus*; strain FH274 developed the characteristic ascospores of *N. pseudofischeri*. All three isolates grew profusely at 45°C and exhibited limited growth at 48°C.

According to Balajee *et al.* (2005), it 'thus appears that traditional phenotypic methods alone may be inadequate for fungal speciation, and that an integration of molecular speciation methods with available classical techniques appears to be warranted to accurately characterize fungi, especially ascomycetes'. This provocative statement does not, however, provide an answer to the question of how a cleistothecial strain of *Neosartorya* Malloch & Cain was filed as *Aspergillus fumigatus*.

***Sordaria thermophila* Fields** – *Mycologia* 60: 1117. 1968.

The original isolate was derived from cow dung collected in Big Bend National Park, Texas (USA), and incubated in a moist chamber at the University of Texas. The isolate proved to be a heterothallic member of the genus *Sordaria* Ces. & De Not. approximating *S. heterothallis* Fields & Maniotis (1963) in most gross morphological features, but deviating by the smaller size of its perithecia, asci and ascospores. Single spore cultures of the new isolate, when crossed with *Sordaria heterothallis*, resulted in the production of perithecia with aborted asci. Similar results from many such crosses indicated the new isolate to be closely related to *Sordaria heterothallis*, yet sufficiently genetically divergent to prevent interfertility. No reactions were found with other species of the same genus. According to Fields (1968) the marked morphological differences and interfertility with other species merit the erection of a new species.

Fields (1968) underlined the selected 'specific epithet refers to a high temperature requirement for ascospore germination. Ascospores of the new species germinated less than 1% on media containing sodium acetate. With an additional treatment of 40-45°C for 8-12 h, germination was increased to 40%. Ascospore germination of other species of *Sordaria* is stimulated by media containing acetate, but no other species of the genus responded to heat treatment'. In the published note, however, no data are given on the *in vitro* linear variation of growth with temperature. Also the conditions at which moist chambers were incubated are not specified. The thermophilic nature of this ascomycete thus cannot be ascertained; it could at least be regarded as a thermotolerant organism.

Guarro & von Arx (1987) re-examined all cultures of *Sordaria* species maintained in the CBS Culture Collection (excluding *Sordaria thermophila*) and compiled a checklist of all known *Sordaria*. They restricted the genus to species with ostiolate ascomata, cylindrical asci and aseptate, elongate, cylindrical, ellipsoidal, or fusiform ascospores with a distinct mucous sheath. *Sordaria thermophila* was retained in the genus and regarded as a 'heterothallic relative of *Sordaria fimicola* (Rob.) Ces. & De Not. with thermophilic requirements'. They did not, however, examine the relevant holotype, a dried culture: Fields 110: Herbarium, New York Botanical Garden. Further investigations are thus needed to circumscribe this ascomycete, which has apparently not been reported since its description (Bell, 2005).

Species of *Sordaria* with ascospores ovate to broadly ellipsoidal, smooth and lacking a mucous sheath were then classified in a separate genus *Asordaria* Arx *et al.* (von Arx *et al.*, 1987). Eriksson & Hawksworth (1988: 61, note 559) regarded these morphological differences as insufficient to merit the recognition of a separate genus; they proposed synonymy under *Sordaria* pending the discovery of further supporting characters.

LICHENISED ASCOMYCETE

***Acarospora thermophila* Herre** – *Botanical Gazette* 55: 394. 1913.

According to Herre (1913) 'the lichen is common on rocks everywhere in the desert about Reno, Nevada (USA), at an altitude of 1200-1500 m; not rare on Mt Rose at 2400 m and above, and abundant in the Sierras along the Truckee River near the Nevada-California state line at an altitude of 2000 m. In a preceding paper [*Botanical Gazette* 51: 286-297. 1911] this plant was mistakenly called by me *Acarospora thamnina*. It is certainly the most successful xerophyte of the Nevada desert, growing in the driest places, where it is exposed to the most intense light and heat. The apothecia are rarely developed, but the scales are commonly covered with a parasitic fungus so that they appear fertile, but their true condition is readily shown by careful sectioning'. The development of this species of *Acarospora* A. Massal. under such pronounced xerothermic conditions definitely accounts for the selection of the epithet *thermophila*. But this epithet most likely implies the exaggerated thermotolerant abilities of the organism.

In the same note Herre (1913) also provided a description of *Acarospora thamnina* (Tuck.) Herre [= *Lecanora thamnina* Tuck. – *Genera Lichenum*: 120. 1872; = *Lecanora cervina* b. *thamnina* Tuck. – *Synopsis. North American Lichens* 1: 202. 1882]. *Acarospora thermophila* was subsequently regarded as matching *A. thamnina* by Magnusson (1929), a taxonomic decision that has apparently not gained wide acceptance (Esslinger, 2006).

ANAMORPHIC FUNGI

***Bactrodesmium fusiforme* Udaiyan** [as '*fusiformis*'] – *Journal of Economic and Taxonomic Botany* 15: 631. 1991, 1992; *nom. inval.*, Art. 37.1 ICBN.

Udaiyan (1991) described several new species of Indian hyphomycetes from the water-cooling towers of electrical power stations at Madras. The original strains developed on pine (*Pinus patula*) and beech (*Fagus rhamnifolia*) test blocks maintained for a certain time in contact with flowing water at ca. 42°C. After a prolonged incubation period, test blocks are removed and developing fungi recovered following the isolation techniques suggested by Butcher (1971,

1972) and Eaton & Jones (1971); the latter incubated decaying blocks at room temperature on common laboratory media to isolate pure cultures of wood-degrading fungi. The Indian author most probably applied this same procedure, as he omitted details on the recovery technique. No holotype was designated by him for the new names introduced. Udaiyan (1991) only stressed that 'type cultures have been deposited in the Botany Department, Centre for Advanced Studies in Botany, Madras University, Madras'. Repeated requests for examining some of these type cultures not deposited elsewhere proved unsuccessful.

The original strain of *Bactrodesmium fusiforme* was isolated from beech test blocks placed both at the inlet point of the cooling tower and at the collecting lagoon of the Basin Bridge Power Generating Station. According to Udaiyan (1991) 'the fungus required a high temperature of 47°C to grow on PDA medium and even at this temperature only vegetative growth was obtained. The fungus may probably require a temperature higher than this for sporulation and that high temperature could be available in the deteriorated wood blocks at both the situations'. The obvious question is thus: under which cultural conditions was the author able to prepare the provided protologue and the relevant iconography of this new dematiaceous sporodochial species? The observation that only vegetative growth was obtained at 47°C might also simply indicate that this is the maximum growth temperature for the present dematiaceous hyphomycete.

The new *Bactrodesmium* Cooke species was not qualified as a thermophile although the published comment on its growth temperature relationships would suggest such an attribute. Its development on wood test blocks continuously washed by warm water may be an indication this fungus is rather a thermotolerant. However, only *in vitro* cultures at fixed incubation temperatures would provide valid data on its heat-tolerance.

In addition, the provided description stresses that the 'conidiogenous cells are monoblastic, integrated, terminal, determinate and cylindrical'. The accompanying iconography, however, clearly depicts a few conidiophores with determinate growth, some showing elongation by subapical proliferation and, most important, others with percurrent proliferations of the annellophore type. Furthermore, in the protologue of the second new *Bactrodesmium* introduced in the same note, *B. indicum* Udaiyan [as '*indica*'], the author underlined 'the presence of long, fasciculate, macronematous conidiophores and the formation of successive conidia on percurrently proliferating conidiophores, the conidia bearing two dark-bands distinguishes this species from the rest'. However, in the provided *camera lucida* drawings of *Bactrodesmium indicum* the conidia are drawn as being distinctly 5-7 septate. These unclear protologues suggest that the author was not dealing with pure cultures of a single fungus.

According to Ellis M.B. (1971), species of *Bactrodesmium* are characterised by 'conidiogenous cells being monoblastic, integrated, terminal, determinate and cylindrical'. As the original material could not be examined despite several requests, the exact identity of *Bactrodesmium fusiformis* remains unassessed.

***Calcarisporiella thermophila* (H.C. Evans) de Hoog** – *Studies in Mycology, Baarn* 7: 68. 1974.

≡ *Calcarisporium thermophilum* H.C. Evans [as '*thermophile*'] – *Transactions of the British Mycological Society* 57: 247. 1971.

The original living culture was recovered from coal spoil tips at Staffordshire, England. According to Evans (1971b), the minimum growth temperature is 16°C, with an optimum at 40°C and a maximum at 50°C.

Calcarisporium thermophilum should thus be considered a thermotolerant in Cooney & Emerson's sense.

Calcarisporium Preuss is characterized by the arrangement of the conidiogenous cells in verticils on erect conidiophores, but they may also be produced singly on micronematous conidiophores. Strains of *Calcarisporium thermophilum* have not shown the distinct conidiophore stage on standard media and true verticils are not evident (Evans, 1971a). The fungus also differs by the distinctive shape of the conidiogenous cells and conidiiferous rachids, two features preventing its maintenance in this genus. This accounts for the establishment of the mucedinaceous genus *Calcarisporiella* de Hoog for this fungus.

Calcarisporiella thermophila morphologically resembles some species of *Sporothrix* Hektoen & Perkins ex Nicot & Mariat, but differs by its widely undulating, fragile hyphae, the shape of the conidiogenous cells, the broad conidium-bearing denticles and the shape and size of the conidia. The genus *Calcarisporiella* is apparently still monospecific (Mouchacca, 1997).

***Geotrichum candidum* var. *thermoideum* Qureschi & Mirza** – *Biologia, Lahore* 27: 144. 1981.

The authentic strain was obtained from camel dung in Pakistan. The holotype could not be examined by van Oorschot & de Hoog (1984) who regarded the variety as a possible synonym of *Arthrographis sulfurea* (Grev.:Fr.) Stalpers & van Oorschot [in Stalpers – *Studies in Mycology, Baarn* 24: 87. 1984], a mesophilic hyphomycete. In the original diagnosis no attention was paid to the presence or absence of connectives between the conidia along the fertile hyphae (van Oorschot & de Hoog, 1984). Similar attempts by the present author to obtain the relevant type material have remained unsuccessful.

Geotrichum candidum Link:Fr. is the anamorph of *Galactomyces geotrichum* (Butler & Peterson) Redhead & Malloch and is known under diverse names (de Hoog *et al.*, 1986). In *Geotrichum* Link species the wide fertile hyphae disarticulate retrogressively, initially into rectangular cells which later break up into short cylindrical to cubic cells (in *Geotrichum candidum*), then inflating slightly in basipetal sequence. Representative strains do not grow above 40°C, with optimal development at about 25°C. It follows that var. *thermoideum* has no taxonomic affiliations with the species. The selected epithet most probably refers to the high temperature conditions under which the dung was incubated.

***Gilmaniella macrospora* Mustafa** [as Moustafa] – *Persoonia* 8: 332. 1975.
= *Gilmaniella thermophila* Qureshi & J.H. Mirza – *Biologia, Lahore* 29: 341. 1983; synonymy *vide* Mouchacca (2000a).

The original living strain of *Gilmaniella thermophila* was derived from goat dung in Pakistan. In the protologue, it is simply stated that the fungus 'showed nearly all the characteristics of *Gilmaniella humicola* G.L. Barron, genus type species, from which it was clearly distinct in its ability to grow at a wider range of temperatures of 37-50°C, a minimum of 27-30°C and a maximum of 52-55°C, the fungus is described here as a new thermophilic species'. Attempts to obtain authentic material including a probable living strain proved unsuccessful. Neither has any isotype been deposited in any major international herbarium. The reported morphological and ecological features of the fungus cannot thus be checked.

Gilmaniella macrospora was first encountered while examining the mycoflora of salt-marsh soils of Kuwait, but its thermotolerance was not then investigated (Moustafa, 1975). It was also subsequently recovered, although infrequently, from Iraqi soils examined by Abdullah & Al-Bader (1990). The Iraqi strains developed optimal growth at 40°C (50 mm in 4 days); no growth occurred at 50°C while at 25°C linear growth attained 15 mm in 4 days. The specific epithet

refers to the globose conidia being larger than those of *Gilmaniella humicola*, the type species, *i.e.*: 14-18 vs 7-10 μm for the latter. The brief diagnosis and the sketchy iconography provided for *Gilmaniella thermophila* clearly approximate the published characters of *G. macrospora*. For this reason, both taxa are regarded as conspecific.

Gilmaniella thermophila was overlooked by Sivanesan and Sutton (1985) while describing *G. punctiformis* and by Moustafa and Ezz-Eldin (1989) in their incorporation of *G. multiporosa* from Egyptian soils in North Sinai. The recent description of *Gilmaniella bambusae* (Umali *et al.*, 1998) brings to six the number of known species.

***Nodulisporium thermoroseum* Subrahm.; *nom. nud.*, Art. 32.1c ICBN.**

The name was first reported in a memoir prepared for a D.Sc. degree and was considered to represent a thermophilic fungus (Subrahmanyam, 1984). The original strain developed on moistened cellophane paper (Subrahmanyam, 1999: 23). As no subsequent publication validating this binomial could yet be traced, this must be considered a 'ghost name' (Mouchacca, 1997, 1999). Renuka *et al.* (1986) reported the presence of phenolic compounds and citric and lactic acids in the culture filtrates for the name *Nodulisporium thermoroseum*. Attempts to examine the strain under consideration proved unsuccessful.

Many conidial states of the ascomycetous genus *Hypoxylon* Bull.:Fr. could be accommodated in the genus *Nodulisporium* Preuss and in several other genera of hyphomycetes. A document clarifying the taxonomy of these conidial states await to be produced (Jong & Rogers, 1972).

***Phaeodactylella lignicola* Udaiyan – *Journal of Economic and Taxonomic Botany* 15: 631. 1991, 1992; *nom. inval.*, Art. 37.1 ICBN.**

This is the type species of *Phaeodactylella* Udaiyan (Udaiyan, 1991). The genus was introduced to accommodate a hyphomycete isolated from beech test blocks placed at the inlet point of the cooling tower and at the collecting lagoon of the Basin Bridge Power Generating Station in Madras. Following the protologue 'the colonies on PDA and malt agar disclose slow growth at laboratory conditions of light and temperature, attaining 1.3 cm diam. in 13 d; colonies addressed with little aerial mycelium and on PDA and MA gray to blackish gray. But at the high temperature of 42°C, the growth is accelerated attaining 4 cm in 13 d. The optimum temperature for this fungus was recorded at 45°C'. The fungus was not however qualified as a thermophile, and no further growth temperature data were determined.

This dematiaceous hyphomycete was characterised by 'solitary, septate, acrogenous conidia that are gangliar in origin, and hence, it is closer to the genus *Dactylella* Grove. Conidia are reported to be of two types: long, fusiform, pale to mid-brown, smooth, 2-5 septate, and distinctly short, clavate, 1-3 septate conidia. An interesting character of the fungus is the formation of 'trapping loops reminding the nematophagous fungi' (Udaiyan, 1991).

Presently no nematode trapping fungus with dark mycelium has yet been described (Rubner, 1996). The reported (and depicted) trapping loops that 'reminds the nematophagous fungi but its function is not known since no nematodes were seen in the loops' closely approximate simple hyphal loops commonly developed by some hyphomycetes in culture. Also, the second type of conidia, which are pyriform to obpyriform, 0-3 septate, dark brown to black, with terminal cells also dark brown to black, are suggestive of conidia of some *Trichocladium* Harz species (Goh & Hyde, 1999). It follows that the exact taxonomic position of this thermotolerant hyphomycete awaits a careful study of authentic material, although this has remained unavailable despite several requests.

***Stachybotrys thermotolerans* McKenzie**, in Pinruan, McKenzie, Jones & Hyde – *Fungal Biodiversity* 17: 149. 2004; *nom. nov.*

= *Stachybotrys ramosa* Udaiyan – *Journal of Economic and Taxonomic Botany* 15: 641. 1991, 1992; *nom. illegit.* Art. 53.1 ICBN, being a homonym of *Stachybotrys ramosa* Dorai & Vittal – *Transactions of the British mycological Society* 87: 642. 1986, 1987.

The fungus was isolated from pine test blocks placed at the inlet spray nozzle of the cooling tower of the power generating station of the Madras Fertilizers plant. It was stated to be 'a thermophile-optimum 43°C'. Also the 'size, shape and non-verruculose nature of the smooth conidia, 8.5-15.5 × 3.5-5 µm, makes this species distinct from known species of *Stachybotrys* Corda' (Udaiyan, 1991). As no authentic material could be located, its identity and heat tolerance levels can not properly be assessed.

The Indian author is apparently unaware of Cooney & Emerson's definitions of heat tolerant fungi (Cooney & Emerson 1964) since no relevant cardinal growth temperatures were determined to substantiate the thermophilic nature of this dematiaceous hyphomycete. The proposed taxon should better be regarded as a thermotolerant species and this indication accounts for the new specific epithet *thermotolerans* selected by Pinruan *et al.* (2004). The latter authors were also unable to locate the original material.

The conidia were depicted by Udaiyan (1991) as ellipsoidal, truncate at the base, straight, dark brown, smooth, 8.5-15.5 × 3.5-5 µm. They are reminiscent of conidia of *Stachybotrys dichroa* Grove, which are ellipsoidal or cylindrical and rounded at the ends, often obliquely attenuated at the base, olivaceous brown to almost black, verrucose when mature, 8-14 × 4-6 µm (Ellis M.B., 1971).

***Trichocladium heterosporum* Udaiyan** [as '*heterospora*'] – *Journal of Economic and Taxonomic Botany* 15: 644. 1991, 1992; *nom. inval.*, Art. 37.1 ICBN.

The original strain developed on pine and beech test blocks maintained at the inlet spray nozzle and in the collecting lagoon of the cooling tower of the Basin Bridge Power Generating Station, Madras. Following the protologue 'the fungus required a temperature of 42°C for growth and sporulation on PDA and malt agar and an optimum temperature at 47°C. The presence of two types of conidia and the thermophilic nature are unique features not seen in other species of *Trichocladium*. Hence, it is proposed as a new species'.

Trichocladium Harz, based on the lectotype species *T. asperum* Harz, includes dematiaceous species of hyphomycetes producing solitary, thick-walled, more or less pyriform to clavate phragmoconidia on micronematous or semi-macronematous, mononematous conidiophores (Goh & Hyde, 1999). The production of two types of conidia thus excludes *Trichocladium heterosporum* from the genus. The conidia are reportedly subglobose to obpyriform, 2-septate, not constricted at the septa, and measure 13.5-15.5 × 6.5-10.5 µm, thus being reminiscent of species of *Bactrodesmium* Corda having a central dark band; some conidia of the provided iconography have a large central dark band.

No cardinal growth temperatures were reported for this fungus. Also, it was not specified under which cultural conditions the provided description was established. *Trichocladium heterosporum* should thus better be considered for the moment as a thermotolerant organism. The exact taxonomic position and the proper level of thermotolerance of this hyphomycete await localisation of an eventual authentic strain.

***Zalerion maritima* (Linder) Anastasiou** – *Canadian Journal of Botany* 41: 1136. 1963.

≡ *Helicoma maritimum* Linder, in Lindner – *Farlowia* 1: 405. 1944.

= *Zalerion thermophylli* Udaiyan – *Journal of Economic and Taxonomic Botany* 15: 644. 1991, 1992; *nom. inval.*, Art. 37.1 ICBN.

For other synonymies, see Ellis M.B. (1976).

The original strain of *Zalerion thermophylli* developed on beech wood test blocks placed at the inlet point of the cooling tower and at the collecting lagoon of Basin Bridge Power Generating Station. Following Udaiyan (1991) the 'present species resembles *Zalerion arboricola* Buczaki but differs from it in the conidial measurements, mode of coiling, unbranched and smooth nature and the high temperature requirement for growth (42°C) warrants its placement in the new species'. In the protologue no indication is however given on the laboratory conditions that enabled the description of the fungus, nor were any cardinal growth temperature values assessed. The specific epithet *thermophylli* might have thus been simply suggested by the high temperature of the warm water circulating in the cooling tower.

The dematiaceous hyphomycetous genus *Zalerion* Moore & Meyers was established for a widely distributed mesophilic fungus trapped on wood blocks immersed in sea-water, *Z. maritima* (Linder) Anastasiou, earlier described under several names (Ellis M.B., 1976). It produces conidia characterized by being helioid, solitary, multiseptate, constricted at the septa, pale to mid brown and smooth; the coiled hyphae is 4-11 µm thick and the conidia is 15-40 µm in diam. The protologue of *Zalerion thermophylli* stresses that the solitary conidia are helioid, multiseptate, constricted at the septa, mid to dark brown, smooth, coiled regularly or irregularly often in several planes, simple, 13.5-25.5 × 8.5-10.5 µm. These characters and the accompanying iconography closely matches the description of the type species rather than that of *Zalerion arboricola* (Mouchacca, 2000a).

Bills *et al.* (1999) re-examined all available strains of *Zalerion arboricola* and of other members of the genus. Their observations confirmed the latter is morphologically and phylogenetically heterogeneous. Furthermore, one strain reported as *Zalerion arboricola* proved so distinct that it warranted the establishment of the new genus *Glarea* Bills & F. Peláez, type species *G. lozoyensis* Bills & F. Peláez. These authors, however, did not take *Zalerion thermophylli* into consideration.

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