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Published in:
Studies in Mycology

Link to article, DOI:
10.3114/sim.2011.69.04

Publication date:
2011

Document Version
Publisher's PDF, also known as Version of record

Citation (APA):
Samson, R. A., Peterson, S. W., Frisvad, J. C., & Varga, J. (2011). New species in Aspergillus section Terrei. Studies in Mycology, (69), 39-55. https://doi.org/10.3114/sim.2011.69.04

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New species in *Aspergillus* section Terrei

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Abstract: Section Terrei of *Aspergillus* was studied using a polyphasic approach including sequence analysis of parts of the β-tubulin and calmodulin genes and the ITS region, macro- and micromorphological analyses and examination of extracellular profiles to describe three new species in this section. Based on phylogenetic analysis of calmodulin and β-tubulin sequences seven lineages were observed among isolates that have previously been treated as *A. terreus* and its subspecies by Raper & Fennell (1965) and others. *Aspergillus alabamensis*, *A. terreus* var. *flocosus*, *A. terreus* var. *aficanus*, *A. terreus* var. *aureus*, *A. hortai* and *A. terreus* NRRL 4017 all represent distinct lineages from the *A. terreus* clade. Among them, *A. terreus* var. *flocosus*, *A. terreus* NRRL 4017 and *A. terreus* var. *aureus* could also be distinguished from *A. terreus* by using ITS sequence data. New names are proposed for *A. terreus* var. *flocosus*, *A. terreus* var. *aficanus*, *A. terreus* var. *aureus*, while *Aspergillus hortai* is recognised at species level. *Aspergillus terreus* NRRL 4017 is described as the new species *A. pseudoterreus*. Also included in section *Terrei* are some species formerly placed in sections *Flavipes* and *Versicolores*. A clade including the type isolate of *A. niveus* (CBS 115.27) constitutes a lineage closely related to *A. carneus*. *Fannelia nivea*, the hypothesised teleomorph is not related to this clade. *Aspergillus allahabadii*, *A. niveus* var. *indicus*, and two species originally placed in section *Versicolores*, *A. ambiguous* and *A. microcysticus*, also form well-defined lineages on all trees. Species in *Aspergillus section Terrei* are producers of a diverse array of secondary metabolites. However, many of the species in the section produce different combinations of the following metabolites: acetylaranotin, asperphenamate, aspochalamins, aspulvinones, asteltoxin, asterriquinones, aszonalenins, atrovenetins, butyroloactones, citreoviridins, citrinins, deacatinis, fulvic acid, geodins, gregatinis, mevinolins, senarhytpine, teric acid (only the precursor 3,6-dihydroxydiquinone found), terreins, terrequineins, teretins and teritems. The cholesterol-lowering agent mevinolin was found in *A. terreus* and *A. neoafricanus* only. The hepatotoxic extracellular chitin was found in eight species: *A. alabamensis*, *A. allahabadii*, *A. carneus*, *A. flocosus*, *A. hortai*, *A. neoafricanus*, *A. niveus* and *A. pseudoterreus*. The neurotoxic extracellular citreoviridin was found in five species: *A. neoafricanus*, *A. aureoterreus*, *A. pseudoterreus*, *A. terreus* and *A. niveus*. Teritems, tremorgenic extratoxins, were found in some strains of *A. alabamensis* and *A. terreus*.

Key words: Ascomycetes, *Aspergillus* section Terrei, β-tubulin, calmodulin, citreoviridin, *Eurotiales*, extrotoxins, ITS, polyphasic taxonomy.

Taxonomic novelties: *Aspergillus aureoterreus* stat. et nom. nov., *Aspergillus flocosus* comb. et stat. nov., *Aspergillus neoafricanus* stat. et nom. nov., *Aspergillus neoafricanus* var. nov., *Aspergillus pseudeuterreus* sp. nov.

INTRODUCTION

*Aspergillus* section *Terrei* (Gams et al. 1985; *A. terreus* species group according to Raper & Fennell 1965) includes species with columnar conidial heads in shades of buff to brown. The most important species of this section is *A. terreus*, which is an ubiquitous fungus in our environment. Strains of this cosmopolitan species are frequently isolated from desert and grassland soils and compost heaps, and as contaminants of plant products like stored corn, barley and peanuts (Kozakiewicz 1989). *Aspergillus terreus* is an economically important species from a number of aspects. *Aspergillus terreus* isolates are used in the fermentation industry for the production of itaconic acid and citric acid and for enzyme production (Bigelis & Arora 1992, Lowe 1992). *Aspergillus terreus* isolates produce a range of secondary metabolites, some of which have properties valuable for mankind, including lovastatin, a cholesterol lowering drug (Alberts et al. 1980), the antitumor metabolites terrein (Arakawa et al. 2008, Demasi et al. 2010), quadrone (Carton et al. 1978) and asterriquinone (Kaji et al. 1998), acetylichenosterase inhibitors like territrem B (Chen et al. 1999) and terreulactone, butyroloactones (Schimmel et al. 1998), and cyclosporine A (Sallam et al. 2003). Antiviral compounds such as acecaliaranotin has also been reported from *Aspergillus terreus* (Miller et al. 1968, Kamata et al. 1983). Other secondary metabolites reported to be produced by *A. terreus* isolates are considered as mycotoxins, including citrerin (Franck & Gehrken, 1980), patulin (Kent & Heatley 1945, Draughon & Ayres 1980, Reddy & Reddy 1988), cinirin (Sankawa et al. 1983), emodin (Fuji & Heatley 1982), terretoxin (Springer et al. 1979, Li et al. 2005), geodin (Kiriyama et al. 1977, Rannest et al. 2011), teritems (Ling et al. 1979), gliotoxin (Lewis et al. 2005, Kupfahl et al. 2008), and cytochalasin E (Fujishima et al. 1979). *Aspergillus terreus* is also an important human pathogen, and often causes disseminated infection with increased lethality compared to other *Aspergillus* spp. (Tracy et al. 1983, Iwen et al. 1998, Lass-Florl et al. 2000, Walsh et al. 2003, Baddley et al. 2003, Steinbach et al. 2004, Balajee 2009). Recent data indicate that the accessory conidia produced by *A. terreus* can induce elevated inflammatory responses in a pulmonary model of aspergillosis (Deak et al. 2009, 2011). The importance of *A. terreus* to human health and industry is underlined by the fact that annotation of the full genome sequence of *A. terreus* isolate NIH 2624 is in progress (Birren et al. 2004, http://fungi.ensembl.org/Aspergillus_terreus/Info/Index), while whole-genome shotgun sequencing of isolate ATCC 20542 has also been carried out (Askenazi et al. 2003). Additionally, transcriptional and metabolite profiles (Askenazi et al. 2003) and the extracellular proteome of *A. terreus* have also been examined recently (Han et al. 2010).
Aspergillus terreus was the only species assigned to the A. terreus species group by Raper & Fennell (1965). Molecular studies have since indicated that this section should be expanded to include a number of other species (Peterson 2000, 2008, Varga et al. 2005). Besides A. terreus and its varieties, section Terrei also includes A. niveus (teleomorph: Fennellia nivea), A. carneus, A. niveus var. indicus, A. allahabadi, A. ambiguus and A. microcysticus (Peterson 2000, 2008, Varga et al. 2005). The first three species have previously been placed in section Flavipedes and the last three species were placed in section Versicolors (Raper & Fennell 1965, Samson 1979). Aspergillus niveus has been reported to cause pulmonary aspergillosis (Auberger et al. 2008), and recent data indicate that several isolates previously assigned to A. terreus, including clinical isolates causing aspergillosis, actually represent a separate species, A. alabamensis (Balajee et al. 2009). The last authors also indicated that A. terreus var. aureus should be recognised as distinct species, but they did not provide a formal description (Balajee et al. 2009).

In this study, we examined available isolates, which morphologically belong to section Terrei, to clarify the taxonomic

| Taxon | Strain No. | Origin |
|-------|------------|--------|
| A. neoaficannus | CBS 130.55T = NRRL 2399 | Aspergillus terreus var. africanus; soil, Tafo, Ghana |
| | NRRL 4609 | Aspergillus terreus var. africanus; soil, Panama |
| | IBT 13121 | Aspergillus terreus var. africanus; soil, Japan |
| A. alabamensis | IBT 12702 | Soil, New Mexico |
| | WB 1920 = IBT 22563 | Soil, Cuba |
| | DTO 15-F8 = IBT 29084 | Soil, Argentina |
| | DTO 15-F9 = IBT 29086 | Soil, Argentina |
| | UAB 18 | Sputum, Alabama, USA |
| | UAB 15 | Sputum, Alabama, USA |
| | UAB 20T | Wound, Alabama, USA |
| | NRRL 29810 = IBT 29081 | Soil, Florida, USA |
| A. allahabadi | CBS 164.63T = NRRL 4539 | Soil, India |
| | CCRC 32133 = IBT 21128 | Soil, Taipei, Taiwan |
| A. ambiguus | CBS 117.58T = NRRL 4737 | Savannah soil, Somalia |
| A. aureoterreus | CBS 265.81 | Wheat flour, India |
| | CBS 503.65T = NRRL 1923 | Soil, Texas, USA |
| A. carneus | CBS 494.65T = NRRL 527 = IBT 13986 | Culture contaminant, District of Columbia, USA |
| | NRRL 1928 | Soil, Kansas, USA |
| | NRRL 298 = IBT 22569 | Soil, Kansas, USA |
| A. flocosus | CBS 116.37T = WB 4872 = IBT 22556 | Aspergillus terreus var. flocosus; Waste cloth, Wuchang, China, isolated by Y.K. Shih as No. A 369 |
| A. hortai | CBS 124230T = NRRL 274 = IBT 26384 | Clinical isolate, from ear, Brazil |
| | IBT 16744 | Soil, Galapagos Islands |
| | IBT 16745 | Soil, Galapagos Islands |
| | IBT 6271 | Soil, Florida, USA |
| A. microcysticus | CBS 120.58T = NRRL 4749 | Savannah soil, Somalia |
| A. neoindicus | CBS 444.75T = NRRL 6134 | Aspergillus niveus var. indicus; Soil, Maharashtra, India |
| A. neoneiveus | CBS 261.73T = NRRL 5299 | Forest soil, Thailand |
| | CBS 262.73 = NRRL 5502 | Forest soil, Thailand |
| | CBS 114.33 = NRRL 515 | P. Biourge |
| | CBS 471.91 = NRRL 1955 | Soil, Ontario, Canada |
| A. niveus | CBS 115.27T = NRRL 5505 | A. Blochwitz |
| | NRRL 4751T | Fennellia nivea var. bifida; unknown |
| A. pseudoterreus | CBS 123890 = NRRL 4017 | Soil, Argentina |
| A. terreus | IBT 26915 | Capanbara droppings, Gamboa, Panama |
| | CBS 601.65T = NRRL 255 | Soil, Conneccticut, USA |
| | NRRL 260 | Soil, College Station, Texas, USA |
| | NRRL 1913 | Lung of pocket mouse, Arizona, USA |
| | IBT 6450 | Corn, India |
| | IBT 14590 = UAMH 4733 | Soil, Golf course, Japan |
| | IBT 24859 | Saltern, Slovenia |
| | NRRL 680 = CBS 594.65 = IBT 6252 | Soil, G. Ledingham |
| | CBS 117.37 = WB 4873 | Aspergillus terreus var. subflocosus; Air, Wuchang, China |
status of this section. We used the polyphasic approach including sequence analysis of parts of the β-tubulin and calmodulin genes and the ITS region, macro- and micromorphological analyses and examination of extrolite profiles of the isolates to clarify their taxonomic identity.

MATERIALS AND METHODS

Isolates

The fungi used in this study are listed in Table 1.

Morphological analysis

For macromorphological observations, Czapek Yeast Autolysate (CYA), Malt Extract (MEA) Agar, Yeast Extract Sucrose Agar (YES), Creatine Agar (CREA), and Oatmeal Agar (OA) were used (Samson et al. 2010). The isolates were inoculated at three points on each plate of each medium and incubated at 25 °C and 37 °C in the dark for 7 d. For micromorphological observations, microscopic mounts were made in lactic acid from MEA colonies and a drop of alcohol was added to remove air bubbles and excess conidia.

Extrolite analysis

The isolates were grown on CYA and YES at 25 °C for 7 d. Extrolites were extracted after incubation. Five plugs of each agar medium were taken and pooled together into same vial for extraction with 0.75 mL of a mixture of ethyl acetate / dichloromethane / methanol (3:2:1) (v/v/v) with 1 % (v/v) formic acid. The extracts were filtered and analysed by HPLC using alkylphenone retention indices and diode array UV-VIS detection as described by Frisvad & Thrane (1987, 1993), with minor modifications as described by Smedsgaard (1997). The column used was a 50 x 2 mm Luna C-18 (II) reversed phase column (Phenomenex, CA, USA) fitted with a 2 x 2 mm guard column.

Genotypic analysis

The cultures used for the molecular studies were grown on malt peptone (MP) broth using 1 % (w/v) malt extract (Oxoid) and 0.1 % (w/v) bacto peptone (Difco), 2 mL of medium in 15 mL tubes. The cultures were incubated at 25 °C for 7 d. DNA was extracted from the cells using the Masterpure™ yeast DNA purification kit (Epicentre Biotechnol.) according to the instructions of the manufacturer. The ITS region and parts of the β-tubulin and calmodulin genes were amplified and sequenced as described previously (Varga et al. 2007a–c). Sequences have been deposited in GenBank under accession numbers FJ491703–FJ491731, and FJ531192–FJ531243.

Data analysis

The sequence data was optimised using the software package Seqman from DNASTar Inc. Sequence alignments were performed by MEGA v. 4.0 (Tamura et al. 2007) and improved manually. For parsimony analysis, the PAUP v. 4.0 software was used (Swofford 2002). Alignment gaps were treated as a fifth character state and all characters were unordered and of equal weight. Maximum parsimony analysis was performed for all data sets using the heuristic search option with 100 random taxa additions and tree bisection and reconstruction (TBR) as the branch-swapping algorithm. Branches of zero length were collapsed and all multiple, equally parsimonious trees were saved. The robustness of the trees obtained was evaluated by 1 000 bootstrap replications (Hillis & Bull 1993). Fennellia flavipes NRRL 5504 was chosen as outgroup in these analyses on the basis of prior studies (Peterson 2008).

RESULTS AND DISCUSSION

Phylogeny

Of the aligned β-tubulin sequences, a portion of 569 positions including 244 parsimony informative characters was selected for the analysis; MP analysis of the sequence data resulted in 46 679 similar, equally most parsimonious trees (tree length = 569 steps, consistency index = 0.7232, retention index = 0.9204), one of which is shown in Fig. 1. The calmodulin data set consisted of 764 characters including 299 parsimony informative sites; MP analysis resulted in 12 most parsimonious trees (length = 806, consistency index = 0.7122, retention index = 0.8704), one of which is presented in Fig. 2. The ITS data set consisted of 499 characters including 55 parsimony informative sites; MP analysis resulted in 36 equally most parsimonious trees (length = 102, consistency index = 0.8529, retention index = 0.9600), one of which is presented in Fig. 3.

Seven lineages were observed among isolates that have previously been treated as A. terreus and its subspecies by Raper & Fennell (1965) and others. Aspergillus alabamensis, A. terreus var. floccosus, A. terreus var. africans, A. terreus var. aureus (A. aureoterreus according to Balajee et al. 2009), A. hortai and A. terreus NRRL 4017 all represent distinct lineages from the A. terreus clade based on phylogenetic analysis of calmodulin and β-tubulin sequences (Figs 1, 2). Among them, A. terreus var. floccosus, A. terreus var. africans, A. terreus var. aureus (A. aureoterreus) could also be distinguished from A. terreus by using ITS sequence data (Fig. 3). The A. terreus clade includes some other isolates which form well-defined subclades on the trees based on both β-tubulin and calmodulin sequence data. Further studies are needed to clarify if they represent separate species.

Also included in section Terrei are some species formerly placed in sections Flavipedes and Versicolores. A clade including the type isolate of A. niveus (CBS 115.27) constitutes a lineage closely related to A. carneus. Fennellia nivea, the hypothesised teleomorph is not related to this clade. Aspergillus alahabadii, A. niveus var. indicus, and two species originally placed in section Versicolores, A. ambiguus and A. microcysticus also form well-defined lineages on all trees (Figs 1–3).

Extrolites

Species in Aspergillus section Terrei are producers of a diverse array of secondary metabolites (Table 2). However, many of the species in the section produce different combinations of the following metabolites: acetylaranotin, asperphenamate, aspochalamins, aspulvinones, asteltoxin, asterric acid, asterriquinones, aszonalenins, atrovenetins, butyrolactones, citreoisocoumarins, citreoviridins, citrinins, decaturins, fulvic acid, geodins, gregatins, mevinolins, serantrypinone, terreic acid (only the precursor 3,6-dihydroxytoluquinone found), terreins,
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A. hortai IBT 6271
A. hortai IBT 16744
A. hortai IBT 16745
A. hortai IBT 26384
A. hortai NRRL 274
A. neoafrcianus NRRL 2399
A. neoafrcianus NRRL 4609
A. neoafrcianus IBT 13121
A. pseudoterreus NRRL 4017
A. alabamensis DTO 15-F9
A. alabamensis UAB 18
A. alabamensis DTO 15-F8
A. alabamensis UAB 15
A. alabamensis UAB 20
A. terreus IBT 12702
A. terreus IBT 22563
A. terreus NRRL 29810
A. floccosus CBS 116.37
A. floccosus IBT 22556
A. aureoterreus CBS 265.81
A. aureoterreus NRRL 1923
A. neoindicus CBS 444.75
A. neoindicus NRRL 6134
A. allahabadi NRRL 4539
A. carneus NRRL 1928
A. carneus NRRL 298
A. carneus CBS 494.65
A. niveus CBS 471.91
A. niveus NRRL 1955
A. carneus CBS 111.49
A. niveus CBS 115.27
A. niveus NRRL 4751
A. niveus NRRL 5505
A. niveus NRRL 515
A. niveus NRRL 114.33
A. ambiguus NRRL 4737
A. microcysticus NRRL 4749

F. flavipes NRRL 5504

Fig. 1. The single MP tree obtained based on phylogenetic analysis of β-tubulin sequence data of Aspergillus section Terrei. Numbers above branches are bootstrap values. Only values above 70 % are indicated. F. = Fennellia.
Fig. 2. One of the MP trees obtained based on phylogenetic analysis of calmodulin sequence data of Aspergillus section Terrei. Numbers above branches are bootstrap values. Only values above 70 % are indicated. F. = Fennellia.
Fig. 3. One of the MP trees obtained based on phylogenetic analysis of ITS sequence data of Aspergillus section Terrei. Numbers above branches are bootstrap values. Only values above 70% are indicated. F. = Fennellia.
terrequinones, terretonins and territrems. The cholesterol-lowering agent mevinolin was found in A. terreus and A. neoaficanaus only. The hepatotoxic extrolite citreoviridin was found in eight species: A. alabamensis, A. allahabadi, A. carneus, A. floccosus, A. hortai, A. microcysticus, A. niveus and A. pseudoterreus. The neurotoxic extrolite citreoviridin was found in five species: A. neoaficanaus, A. auroterreus, A. pseudoterreus, A. terreus and Fennellia neonivea. Territrems, tremorgenic extrolites, were found in some strains of A. alabamensis and A. terreus.

Species descriptions

**Aspergillus aureoterreus** Samson, S.W. Peterson, Frisvad & Varga, stat. et nom. nov. MycoBank MB560392. Fig. 5. Basionym: Aspergillus terreus Thom var. aureus Thom & Raper – In A Manual of the Aspergilli: 198, 1945.

Type of *Aspergillus terreus* var. *aureus* from soil, Texas, USA.

This variety was proposed by Thom & Raper (1945) based on the slow growing colonies, which are bright yellow. It produces conidiofhores tardily. As with the variety *africanus*, the ex-type isolate can be clearly distinguished based on our phylogenetic analysis. Therefore we propose to raise the taxon to species level.

**Aspergillus floccosus** Samson, S.W. Peterson, Frisvad & Varga, comb. et stat. nov. MycoBank MB560393. Fig. 6.

Type of *Aspergillus terreus* var. *floccosus* from waste cloth, Wuchang, China, isolated by Y.K. Shih as No. A 369.

The ex-type culture shows white floccose colonies with some hyaline exudate on Czapek yeast agar. On MEA colonies are white with a light brown centre. Isolates of *A. terreus* may vary in colony morphology sometimes showing floccose colonies. The ex-type isolate can be clearly distinguished based on our phylogenetic analysis. Therefore we propose to raise the taxon to species level.

**Aspergillus terreus** var. *floccosus* from waste cloth, Wuchang, China, isolated by Y.K. Shih as No. A 369.

Terrestrial species with strongly floccose colonies and conidial heads which show hyaline exudate on Czapek yeast agar. On MEA colonies are white with a light brown centre. Isolates of *A. terreus* may vary in colony morphology sometimes showing floccose colonies. The ex-type isolate can be clearly distinguished based on our phylogenetic analysis. Therefore we propose to raise the taxon to species level.

**Aspergillus terreus** var. *floccosus* from waste cloth, Wuchang, China, isolated by Y.K. Shih as No. A 369.

The ex-type culture shows white floccose colonies with some hyaline exudate on Czapek yeast agar. On MEA colonies are white with a light brown centre. Isolates of *A. terreus* may vary in colony morphology sometimes showing floccose colonies. The ex-type isolate can be clearly distinguished based on our phylogenetic analysis. Therefore we propose to raise the taxon to species level.

**Aspergillus terreus** var. *floccosus* from waste cloth, Wuchang, China, isolated by Y.K. Shih as No. A 369.

The ex-type culture shows white floccose colonies with some hyaline exudate on Czapek yeast agar. On MEA colonies are white with a light brown centre. Isolates of *A. terreus* may vary in colony morphology sometimes showing floccose colonies. The ex-type isolate can be clearly distinguished based on our phylogenetic analysis. Therefore we propose to raise the taxon to species level.
Fig. 4. Aspergillus aureoterreus. A–C. Colonies incubated at 25 °C for 7 d, A. CYA, B. MEA, C. CREA, D–I. Conidiophores and conidia. Scale bars = 10 μm.
Fig. 5. *Aspergillus floccosus*. A–C. Colonies incubated at 25 °C for 7 d, A. CYA, B. MEA, C. CREA, D–I. Conidiophores and conidia. Scale bars = 10 μm.
Fig. 6. Aspergillus hortai. A–C. Colonies incubated at 25 °C for 7 d, A. CYA, B. MEA, C. CREA, D–I. Conidiophores and conidia. Scale bars = 10 μm.
Fig. 7. Aspergillus neoafricanus. A–C. Colonies incubated at 25 °C for 7 d, A. CYA, B. MEA, C. CREA, D–I. Conidiophores and conidia. Scale bars = 10 μm.
Fig. 8. Aspergillus neoindicus. A–C. Colonies incubated at 25 °C for 7 d. A. CYA, B. MEA, C. CRE. D–I. Conidiophores and conidia. Scale bars = 10 μm.
Aspergillus neoniveus sp. nov. A–B. Colonies incubated at 25 °C for 7 d, A. CYA, B. MEA, C. Crusts of Hülle cells, D–E, G–I. Conidiophores and conidia. F. Hülle cells. Scale bars = 10 μm, except F = 100 μm.

Fig. 9. Aspergillus neoniveus sp. nov. A–B. Colonies incubated at 25 °C for 7 d, A. CYA, B. MEA, C. Crusts of Hülle cells, D–E, G–I. Conidiophores and conidia. F. Hülle cells. Scale bars = 10 μm, except F = 100 μm.
Fig. 10. Aspergillus pseudoterreus. A–C. Colonies incubated at 25 °C for 7 d, A. CYA, B. MEA, C. CREA, D–I. Conidiophores and conidia. Scale bars = 10 μm.
they examined the ex-type culture and believed that it belonged to A. flavipes. Interestingly they did not treat it as a synonym of this species, but left the name under A. terreus.

**Aspergillus hortai** (Langeron) Dodge, in Medical Mycology: 628. 1935. Fig. 7. 
**Basionym:** Sterigmatocystis hortai Langeron, Bulletin Société de Pathologie Exotique 15: 383. 1922.

Type of Sterigmatocystis hortai as clinical isolate, from ear, Rio de Janeiro, Brazil.

Langeron (1922) described this fungus from a human ear in Rio de Janeiro. Dodge (1935) noticed the resemblance of Langeron’s fungus and transferred it to *Aspergillus*. However, Raper & Fennell (1965) considered it as a synonym of *Aspergillus terreus*, but our phylogenetic analysis clearly shows that it is distinct from *A. terreus* and therefore we accept it as a distinct species. *Aspergillus hortai* is known from the ex-type isolate and soil isolates from the Galapagos Islands and Florida (USA). The species show a strong morphological resemblance to *A. terreus*, but have a distinct extrolites profile.

**Aspergillus neafricanus** Samson, S.W. Peterson, Frisvad & Varga, comb. et stat. nov. MycoBank MB560391. Fig. 4. 
**Basionym:** Aspergillus terreus Thom var. africanus Fennell and Raper, Mycologia 47: 86. 1955.

Type of *Aspergillus terreus* var. africanus from soil, Tafo, Ghana.

The ex-type strain of *Aspergillus terreus* var. africanus is grouped in a distinct lineage from *Aspergillus terreus* and therefore we propose to raise the taxon to species level. Raper & Fennell (1965) considered this as a variety because they observed slow growing colonies on Czapek agar bright yellow vegetative mycelium. CBS 130.55 = NRRL 2399 derived from the type is now more or less floccose, with a yellow centre. Sporulation with yellow brown conidia occurs at the edges of the colonies. The degenerated condition of the culture also explains why we did not observe the sclerotium-like structures on malt extract agar + 20 % sucrose, which were reported by Raper & Fennell (1965).

**Aspergillus neoindicus** Samson, S.W. Peterson, Frisvad & Varga, stat. et nom. nov. MycoBank MB560394. Fig. 8. 
**Basionym:** Aspergillus niveus var. indicus Lal & Sarbhoy, Indian Phytopathology 25: 309. 1973.

Type of *Aspergillus niveus* var. indicus from soil, Maharastra, India.

This species was described as a variety from soil in Maharashtra by Lal & Sarbhoy (1973) and was considered by Samson (1979) as a synonym of A. flavipes. The species is phylogenetically distinct from A. flavipes, and is characterised by yellow green mycelial tufts. On OA a dark green pigment is diffusing into agar. The discrete masses of ellipsoidal and elongate Hülle cells described by Lal & Sarbhoy (1973) and Samson (1979) have not been observed in our current study.

**Aspergillus neoniveus** Samson, S.W. Peterson, Frisvad & Varga, nom. nov. MycoBank MB5603945. Fig. 9. 
**Basionym:** Emericella nivea Wiley & Simmons, Mycologia 65: 934. 1973 (non Aspergillus niveus Blochwitz, 1929).

≡ *Fennellia nivea* (Wiley & Simmons) Samson, Stud. Mycol. 18: 5. 1979

Type from forest soil in Thailand.

Samson (1979) considered *Emericella nivea* distinct from the other *Emericella* species by the hyaline to pale yellow ascospores and the anamorph belonging to the A. flavipes group. The species is similar to *Fennellia flavipes* Wiley & Simmons and could be classified as the second species of *Fennellia*. However our phylogenetic analysis and those by Peterson (2008) and Peterson *et al*. (2008) showed that the isolates of *Emericella nivea* clustered separately from the isolates of *Aspergillus niveus* and hence it represents a different taxon. Following the need for an orderly transition to a single-name nomenclatural system (Hawksworth *et al*. 2011) we have chosen to name this species in *Aspergillus* and not in its teleomorph genus, *Fennellia*.

**Aspergillus pseudoterreus** S. W. Peterson, Samson & Varga, sp. nov. MycoBank MB560396. Fig. 10.

Colonies on CYA orange brown with yellow tufts reaching a diameter of 4 cm. On MEA colonies are bright yellow with a clear orange edge. In older cultures of up to 14 d conidiophores are bundled in loose synnemata. Conidiophores typically biseriate, producing columnar heads. Vesicles globose 16–23 μm diam, stipitatus levibus, hyalinis, 4.5–7 μm latis. Conidios levibus, hyalinis, globosis vel ellipsoidis, 1.5–2.2 × 1.8–2.5 μm.

Typus: from soil Argentina (CBS H-20631 – holotypus, culture ex-type NRRL 4017).

Colonies on CYA orange brown with yellow tufts reaching a diameter of 4 cm. On MEA colonies are bright yellow with a clear orange edge. In older cultures of up to 14 d conidiophores are bundled in loose synnemata. Conidiophores typically biseriate, producing columnar heads. Vesicles globose 16–23 μm, stipe smooth, hyaline, 4.5–7 μm, conidia smooth, hyaline, globose to ellipsoidal, 1.5–2.2 × 1.8–2.5 μm.

This species was isolated from soil in Argentina and is characterised by a pronounced synnematal growth on MEA. The colony colour is reddish brown with biseriate conidiophores producing globose to ellipsoidal conidia.

**ACKNOWLEDGEMENTS**

We thank Uwe Braun for the Latin diagnosis and advice on nomenclatural issues.

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