

A new species of *Melanochaeta* from Kenya

George K. Mugambi*^{1,2,3} and Sabine M. Huhndorf²

¹ Dept. of Biological Sciences, University of Illinois at Chicago, 845 W. Taylor St (MC 066), Chicago, IL 60607, USA

² The Field Museum, Dept. of Botany, 1400 S. Lake Shore Drive, Chicago, Illinois 60605-2496, USA

³ National Museums of Kenya, Botany Dept. P.O. Box 45166, 00100, Nairobi, Kenya

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Melanochaeta taitensis is described as a new species from Kenya. It is distinguished from the other known species of *Melanochaeta* by having large, 3-septate, strongly curved ascospores. Phylogenetic analyses of nuclear 28S large subunit (LSU) sequences including the new species and Kenyan collections of *Melanochaeta hemipsila* support its separation from other species in the genus.

Keywords: 28S nrLSU, anamorph, molecular phylogeny, *Sporoschisma*.

The genus *Melanochaeta* E. Müll., Harr & Sulmont was erected by Müller *et al.* (1969) to accommodate lignicolous species that have superficial perithecial ascomata with capitate setae arising from the entire perithecial surface. Similar setae associated with conidiomata are also found on the substrate. According to Index Fungorum (<http://www.indexfungorum.org>), four names are included in the genus: *M. hemipsila* (Berk. & Broome) Müll.; Harr & Sulmont; *M. aoteoroae* (S. Hughes) Müll.; Harr & Sulmont, *M. garethjonesii* Sivi-chai & Hywel-Jones and *M. daemonoropis* J. Fröhl. & K. D. Hyde. Prior to being placed in *Melanochaeta*, the first two species were placed in various genera including *Lasiosphaeria* Ces. & de Not. and *Chaetosphaeria* Tul. & C. Tul.

During field surveys of the wood-inhabiting pyrenomycetous fungi in Kenya, two species of *Melanochaeta* were collected. One collection matches *M. hemipsila* and the other is morphologically distinct: molecular analysis using partial sequence of nuclear large subunit ribosomal DNA (28S nrLSU DNA) supports its separation from the other species in the genus. The new species is described and illustrated.

* Corresponding author: G.K. Mugambi; email: gmugam1@uic.edu

Materials and Methods

The fungus was collected from Taita hills forest in Kenya during April, 2005. The ascomata were dried at low electric heat and stored in paper bags until further processed at the Field Museum, Chicago, USA. The ascomata were squash-mounted in water and images of micromorphological structures were captured with a Dage DC-330 video system mounted on a Zeiss Axioskop microscope. For molecular analysis, total DNA was extracted using the DNeasy Plant Mini Kit (Qiagen, Hilden, Germany) following the instructions of the manufacturer. Nuclear LSU DNA was amplified using LROR, LR6 and LR3 primers (Vilgalys & Hester 1990). The final volume of the PCR reactions was 25 μ L and contained 2.5 μ L buffer, 2.5 μ L dNTP mix, 1 μ L of each primer (10 μ M), 5 μ L of BSA, 1.5 μ L taq, 2 μ L genomic DNA extract and 9.5 μ L deionized water. The annealing temperature was set at 50 °C and PCR reaction allowed to run for 34 cycles. The fragments were sequenced using the Big Dye Terminator reaction kit (ABI PRISM, Applied Biosystems, Forster City, USA). Sequencing was performed using the same set of primers as PCR. The specimens sequenced in this study and their locality data are given below. The other sequences used in the analysis were obtained from GenBank (<http://www.ncbi.nlm.nih.gov>).

Sequences were aligned using ClustalW (Thompson *et al.* 1994) and further corrected manually. Alignments were analyzed using program PAUP*version 4.0b10 (Swofford 2003). Maximum Parsimony was carried out using heuristic search for 1000 replicates with TBR branch swapping and MulTree option in effect. For assessing branch support Bootstrapping (Felsenstein 1985) was performed based on 1000 replicates with the same settings as the heuristic search.

Results

Four new nuLSU DNA sequences and 18 sequences from the GenBank (Fig. 1) were aligned to produce a matrix of 1049 unambiguously aligned nucleotide positions. Maximum Parsimony analysis recovered four most parsimonious trees (all with CI 0.664, RI 0.772, and HI 0.336) and one is shown (Fig. 1). All of the *Melanochaeta* species form a monophyletic group with 100 % bootstrap support (Fig. 1). The four collections of *M. hemipsila* form a strongly supported clade (91 %) and form a sister group to our new species although this relationship is not strongly supported.

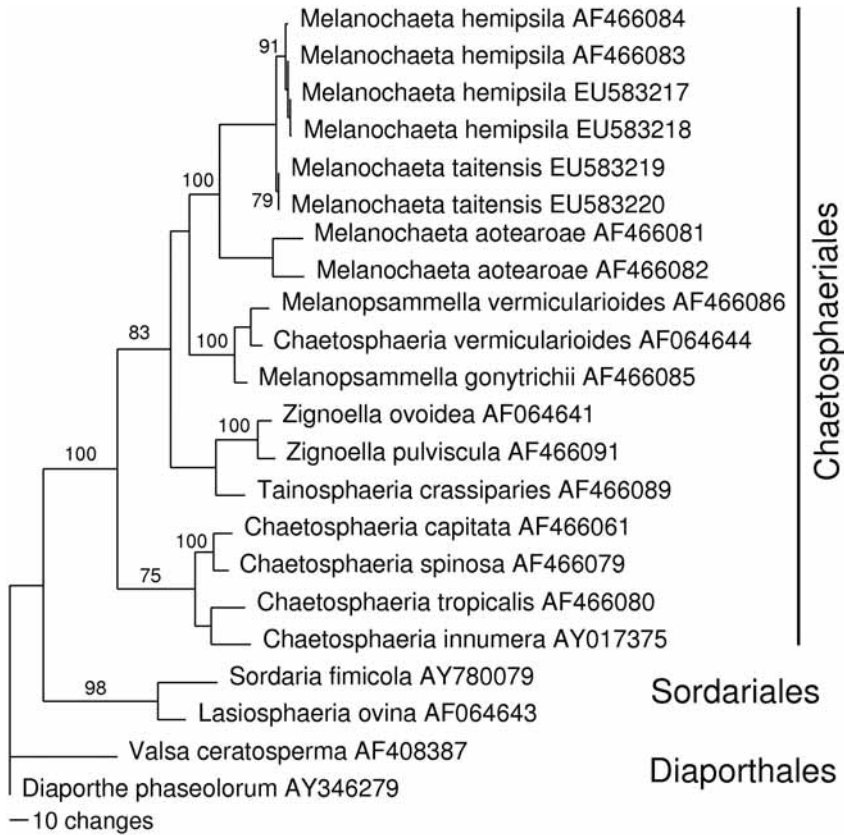


Fig. 1. One of the four most-parsimonious trees based on the LSU sequence data set of 22 taxa. Numbers on the branches indicate bootstrap support based on 1000 replicates.

***Melanochaeta taitensis* Mugambi & Huhndorf sp. nov.** Figs. 2–11.

Anamorph. – *Sporoschisma* Berk. & Broome

Mycobank no.: MB512334

Ascomata ampulliformia vel obpyriformia, brunnea, tunica alba, 350–410 µm diam., 340–390 µm alta, separata, superficialia, cum setis sparsisbrunneis, apicibus capitatis hyalinis. Asci cylindrico-clavati, 158–200 × 10–15 µm, unitunicati, annuli apicales tenues, octospori. Ascosporae fusiformes, (19-) 23–30 (-33) × (5.5-) 6.7–7.7 (-8.6) µm, 3-septatae, cellulis medis brunneis et cellulis terminalibus hyalinis.

Perithecia ampulliform to obpyriform, not collapsing when dry, glaucous with cottony, white tomentum, dark brown to black raised papillae, 350–410 µm diameter, 340–390 µm high, superficial, occurring solitary to aggregated, anchored to the substrate by an indistinct basal stroma, with scattered setae on the ascomata and the substratum. Setae erect, dark brown, with swollen, hyaline to light

brown, capitate apex. Paraphyses hyaline, cylindrical, non-branched and longer than asci. Asci unitunicate, cylindrical to narrowly clavate, 158–200 x 10–15 μm (n = 15), apex with prominent non-amyloid ring, with eight partially biseriolate ascospores. Ascospores fusoid, strongly curved, 3-septate, brown, often with paler end cells, (19-) 23–30 (-33) x (5.5-) 6.7–7.7 (-8.6) μm (n = 36). Conidiomata growing on the substrate among the ascomata, dark brown, cylindrical, simple, phialidic. Conidia produced in chains, cylindrical, 5-septate, dark brown with smaller hyaline end cells.

Etymology. - Refers to the type locality.

Habitat. - On decorticated wood.

Known distribution. - Known only from the type location in Kenya.

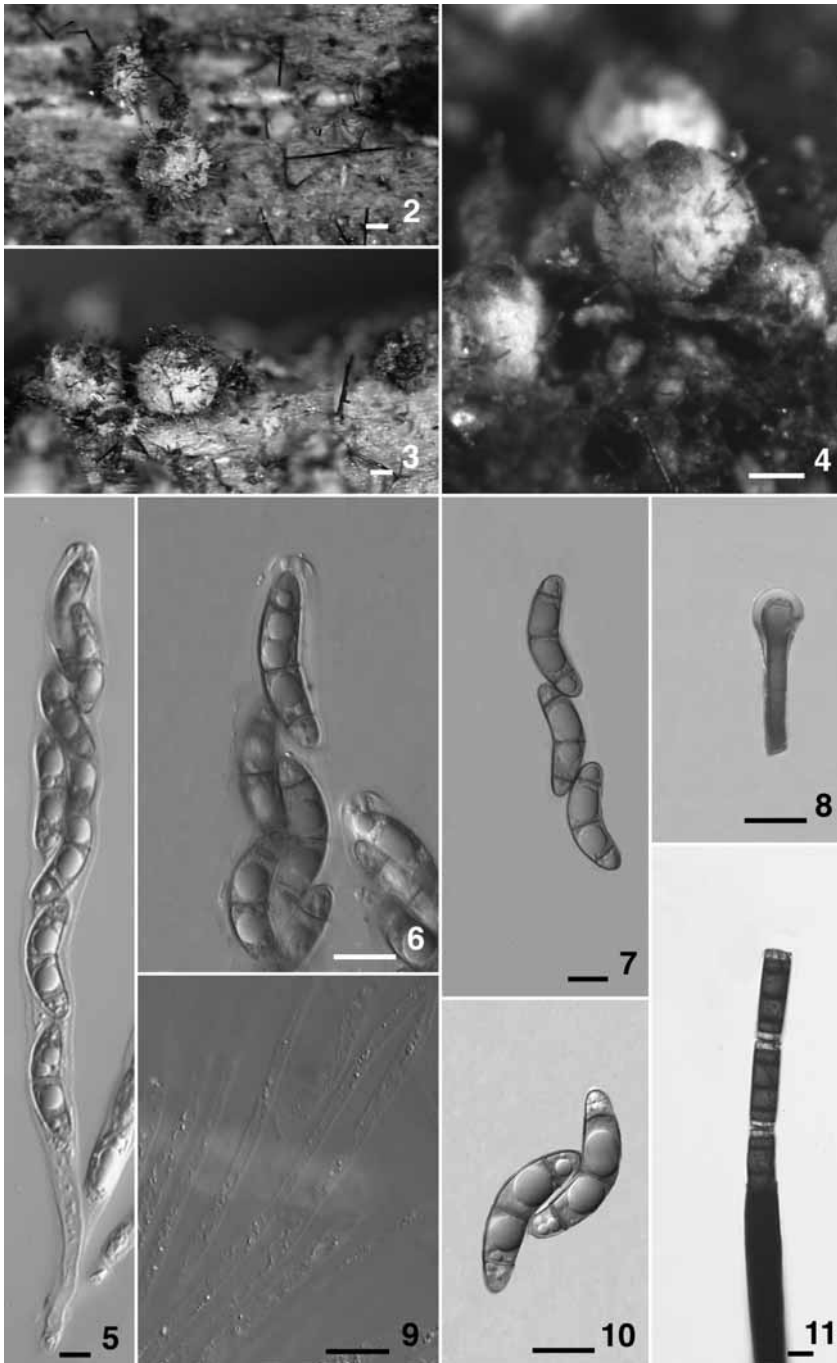
Material examined. - KENYA, Coast Province, Taita-Taveta District, Taita Hills, Ngangao forest reserve, S 3° 22.301', E 38° 20.446', 1800 m, 25 Apr 2005, on woody branch 2 cm diameter, Mugambi 156N (GenBank EU583220) (EA holotype; F isotype, designated here); Mugambi 150N (GenBank EU583219).

Melanochaeta hemipsila (Berk. & Broome) Müll., Harr & Sulmont, *Revue Mycol.*, Paris 33: 377 (1969) [1968].

Material examined. - KENYA. Rift Valley Province: Kajiado District, Institute of Primate Research-Ololua Forest Reserve, S 1° 24.682, E 36° 38.182, ca. 1858 m, 25 May 2005, on woody branch about 3 cm diameter, Mugambi 1112 (GenBank EU583217); 13 Dec 2006, Mugambi 1247 (GenBank EU583218).

Discussion

Molecular and morphological characters strongly support the placement of *M. taitensis* within the genus *Melanochaeta*. Its morphology is distinct from taxa already recognized in this genus and we therefore describe it as new species. *Melanochaeta taitensis* differs from *M. hemipsila* in both ascus and ascospore morphology. The ascospores are much larger and five-septate in *M. hemipsila* while they are three-septate in *M. taitensis*. Both species, however, have similar anamorph morphologies, with cylindrical, five-septate conidia, with hyaline end cells. *Melanochaeta taitensis* can be distinguished from *M. aotearoae* by both ascospore and conidial morphology. The conidia of *M. taitensis* are five-septate with hyaline end cells while in *M. aotearoae* they are one to two-septate. Despite both species having three-septate ascospores, they are strongly curved in *M. taitensis* and fusiform and straight in *M. aotearoae*. On the other hand, *M. garethjonesii* differs from *M. taitensis* by having predominantly four-septate ascospores that are slightly constricted at



Figs. 2–11. *Melanochaeta taitensis* (from Mugambi 156N). 2–4. Perithecia on substratum. 5. Ascus. 6. Ascus apex with ring. 7. Ascospores. 8. Capitate seta. 9. Paraphyses. 10. Ascospores. 11. Conidioma with conidia. **Bars:** 2–4 = 100 μ m; 5–11 = 10 μ m.

the septa and conidia that are predominantly one-septate (Sivichai et al. 2000). *Melanochaeta daemonoropsis* differs in its concolorous ascospores and smaller ascomata (Fröhlich & Hyde 2000).

Ascomycete collecting in Africa in the past, and wood-inhabiting, pyrenomycetous specimens in particular, have been generally overlooked. Kenya has a poor baseline for known ascomycetes. Even the selected and limited collecting performed for this study uncovered taxa new to science and reveals that a rich diversity of fungi is there to be found.

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Autor(en)/Author(s): Mugambi George K., Huhndorf Sabine M.

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