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## A new fungicolous species of *Titaea* and new reports of *Bahusaganda indica* and *Exosporium ampullaceum* (hyphomycetes) from tropical rainforests in Panama

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A new species of *Titaea* overgrowing an unidentified black, thyriothecioid ascomycete on leaves of *Ocotea* sp. (*Lauraceae*) was found in a tropical rainforest in Chiriquí Province of Panama. The conidia of *Titaea tetrabrachiata* sp. nov. develop four tetrahedrally radiating, septate arms and three subglobose cells between the arms arising from the central cell of each conidium. *Bahusaganda indica* and *Exosporium ampullaceum*, both found on dead herbaceous stems, are reported for the first time from Panama.

**Key words:** anamorphic fungi, Ascomycota, mycoparasitic fungi, neotropics

### Introduction

During a study of the diversity of fungi on dead and living plants in a neotropical rainforest, several species of hyphomycetes were found in Panama for the first time, among them, an undescribed species of *Titaea* Sacc.

Sutton (1984) accepted four species of *Titaea* among the hitherto ca. 15 species within the genus described in detail by Hansford (1946), Boedijn (1951), Damon (1952), and Ciferri (1955). A further three species were added by Matsushima and Matsushima (1996) and Peláez *et al.* (1999). The concept for this genus is based on conidiophore and conidium morphology. Conidiophores are hyaline and apically bear intercalary or terminal, ellipsoidal to cylindrical cells each having one or a few broad, unpigmented conidiogenous loci. The hyaline conidia arise solitarily from each conidiogenous locus and consist of a basal cell that gives rise to two or more arms and one or more globose to ellipsoidal cells. Ecologically, the occurrence

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on foliicolous ascomycetes seems to be typical for several species of *Titaea* (Sutton, 1984).

Species of *Titaea* and similar hyphomycetes are considered to be anamorphs of species in the ascomycete genera *Paranectriella* (Henn. ex Sacc. & D. Sacc.) Höhn. and *Puttemansia* Henn. (Hansford, 1946; Pirozynski, 1977; Rossman, 1987). These genera were originally placed in the *Hypocreales* because of their light-coloured perithecioid ascocarps seated on stromata, but later transferred to the *Tubeufiaceae*, *Pleosporales*, because of the bitunicate asci (Rossman, 1987).

## Materials and methods

Living or recently detached leaves and an unidentified species of *Ocotea* sp. (*Lauraceae*) and dead herbaceous stems were collected in rainforests in the high mountains (approx. 2000-2500 m) of Parque Nacional Volcán Barú and Parque Internacional de la Amistad in Chiriquí Province, Panama (February-March 2003). Material was air-dried and scanned with a dissecting microscope. Fungi were mounted in 5% KOH and/or cotton blue in lactic acid and examined with a light microscope. Because of the scarcity of the material in some specimens, it was not possible to obtain enough data for a statistical treatment of conidiophore and conidium measurements without destroying the entire specimen. Where it was possible, conidium measurements were calculated as mean values  $\pm$  standard deviations and with extreme values given in brackets. Free-hand drawings were made using a 1000x magnification and scaled paper. Specimens of the fungi were deposited in the Herbarium of the University of Panama (PMA).

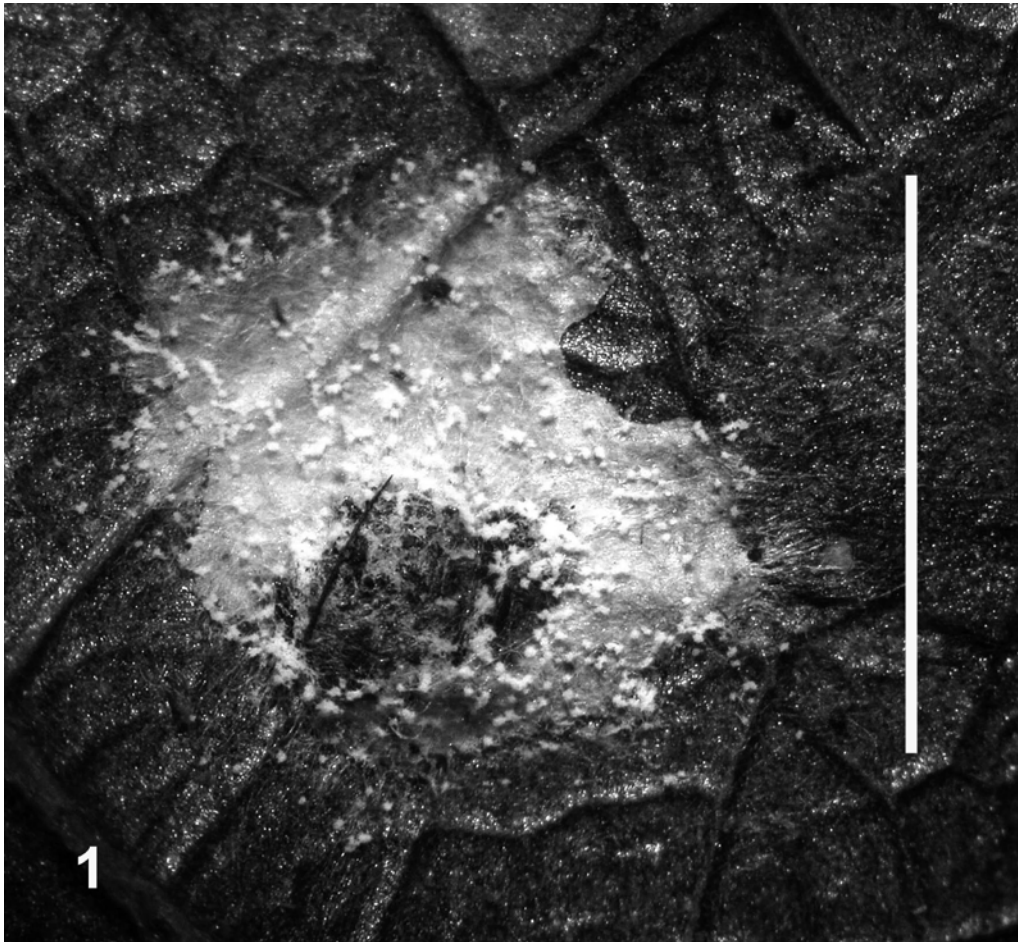
## Results

***Titaea tetrabrachiata* R. Kirschner, sp. nov.** (Figs. 1-2)

*Etymology*: from Greek, tetra-four, brachion-arm, “with four arms”.

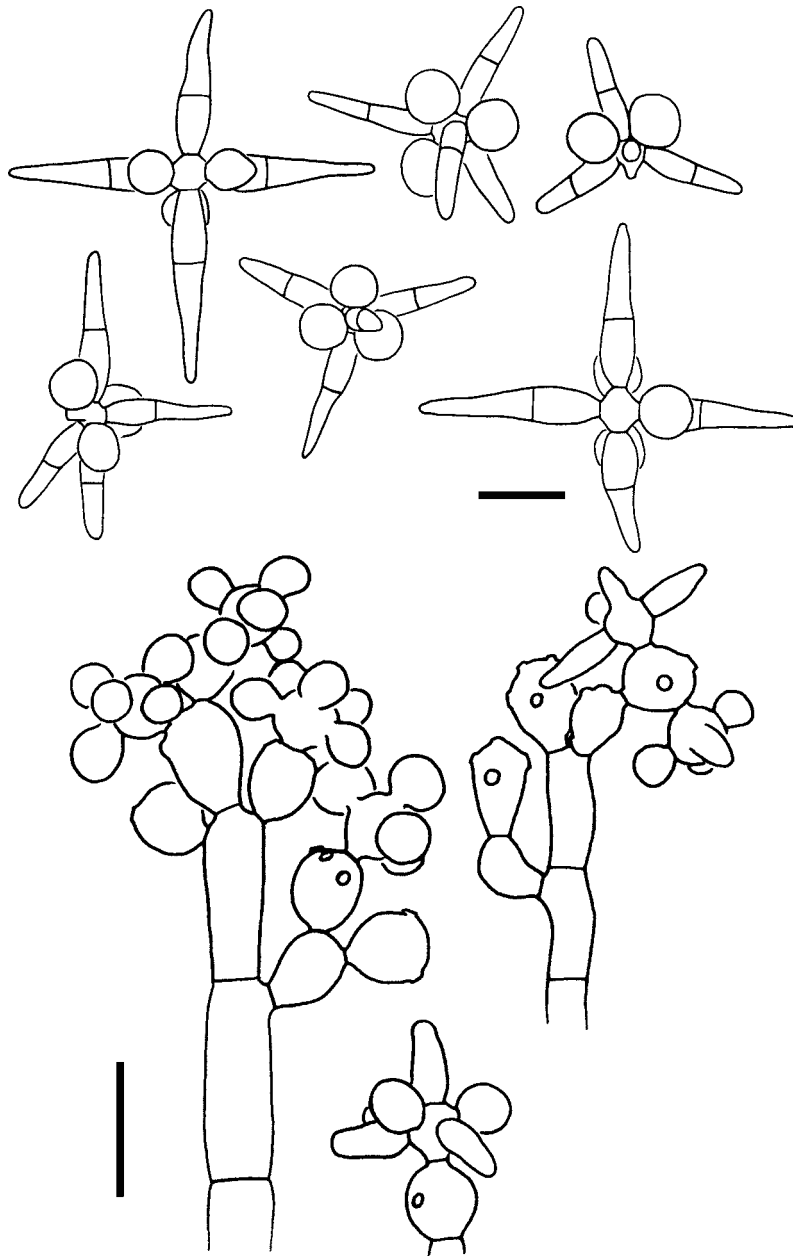
*Mycelium* super fructificationibus ascomycetum, densum, cremeum, ad 2.5 mm diam. *Conidiophora* mononematosa, hyalina, laevia, longitudo stipitis excedens 100  $\mu$ m, latitudo 3-5  $\mu$ m, apicaliter ramificata, ramis 5-7  $\times$  4-5  $\mu$ m, cellulis conidiogenis similibus, intercalaribus vel terminalibus, cicatricibus hyalinis, 1  $\times$  1  $\mu$ m. *Conidia* ex corpusculo centrali unicellulari (3-4  $\mu$ m diam.), 3 cellulis subglobosis (5-6  $\mu$ m diam.) et 4 brachiis 1-septatis (raro 0-septatis) composita (11-22  $\times$  3-4  $\mu$ m).

*Colonies* hypophyllous, overgrowing or laterally growing out from underneath the black, superficial, shield-like ascomata of an unidentified thyrtothecioid ascomycete, covering an area of up to ca. 2.5 mm diam., mycelium forming a dense, cream coloured mat, covered by scattered white



**Fig. 1.** *Titaea tetrabrachiata* (from holotype). Colony and tufts of conidiophores and conidia seen under the dissecting microscope. Bar = approx. 2 mm.

heads of conidiophores and conidia. *Conidiophores* superficial, mononematous, smooth, hyaline, ca. 100  $\mu\text{m}$  long, 3-5  $\mu\text{m}$  wide, apically forming a head composed of swollen cells measuring 5-7  $\times$  4-5  $\mu\text{m}$  that themselves produce intercalary and terminal conidiogenous cells of the same shape and size. Conidiogenous loci 1  $\times$  1  $\mu\text{m}$  diam., slightly protruding, hyaline, and thin-walled after conidium dehiscence. *Conidia* often stuck together on the tip of the conidiophore forming a dry head visible with a dissecting microscope; hyaline, smooth, composed of a central cell with the main body 3-4  $\mu\text{m}$  diam. and a basal hilum measuring 1-2  $\times$  1  $\mu\text{m}$ , four arms tetrahedrally radiating from this central cell; arms straight or slightly curved, 11-22  $\mu\text{m}$  long, 3-4  $\mu\text{m}$  wide at the base and tapering to 1-1.5  $\mu\text{m}$  at the tip;



**Fig. 2.** *Titaea tetrabrachiata* (from holotype). Conidiophore heads with developing conidia, detached conidia. Bars = 10  $\mu$ m.

mostly with a septum (rarely a single arm without a septum and then shorter); three bladder-like, subglobose cells 5-6  $\mu\text{m}$  diam. inserted between the arms; whole conidia approx. 34-44  $\mu\text{m}$  from tip to tip.

*Habitat:* On black, superficial, shield-like ascomata of an unidentified thyriothecioid ascomycete on living or recently detached leaves of *Ocotea* sp. (*Lauraceae*).

*Known distribution:* Panama.

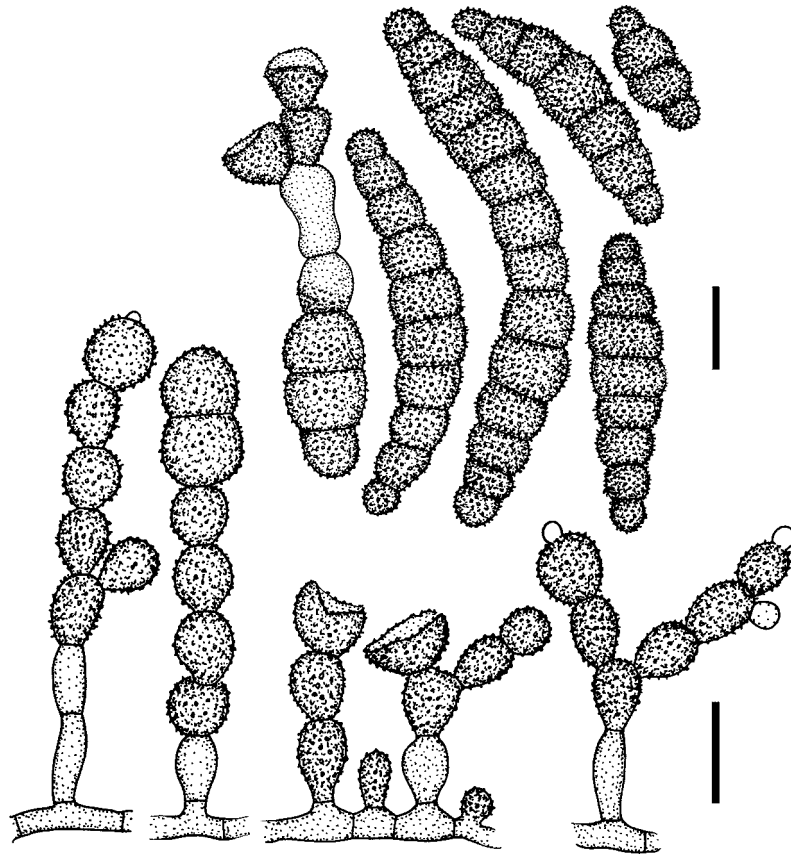
*Material examined:* PANAMA, Chiriquí, Parque Nacional Volcán Barú, Sendero de los Quetzales, ca. 2.000-2.400 m, on black thyriothecioid ascomata on living or recently fallen leaves of *Ocotea* sp. (*Lauraceae*), 15 February 2003, R. Kirschner & M. Piepenbring 1587-A, (PMA; **holotype designated here**).

*Notes:* The hyaline, intercalary and terminal conidiogenous cells with several protruding conidiogenous scars agree with the concept of the genus (Sutton, 1984). The additional, bladder-like cells, however, do not form the distal cell of the main axis of the conidium in *T. tetrabrachiata*, nor arise from the lower side of the arms, but laterally from the basal conidium cell, which is in contrast to the concept proposed by Sutton (1984). Describing *T. complexa*, Matsushima and Matsushima (1996) apparently also did not consider this difference significant on the generic level, because the bladder-like cells of *T. complexa* do not arise from the arms either but laterally from the basal cell of the conidium as in *T. tetrabrachiata*. We follow the wider genus concept applied by Matsushima and Matsushima (1996), particularly with respect to the identical morphology of the conidiogenous cells.

The conidia of *T. tetrabrachiata* are most similar to those of *T. clarkeae*, *T. complexa*, *T. costaricana*, and *T. triradiata*, because of the three-dimensional arrangement of the arms and the presence of hyaline, smooth, bladder-like or subglobose cells. In contrast to *T. tetrabrachiata*, however, arms are not septate in *T. complexa*, *T. costaricana*, and *T. triradiata*. In *T. clarkeae* the central body of the conidium produces only up to three septate arms and only a single subglobose cell (Sutton, 1984). The arms of the conidia of *T. complexa* are short, not exceeding 12  $\mu\text{m}$ , and accompanied by 3-5 subglobose cells (Matsushima and Matsushima, 1996). *Titaea costaricana*, excluded from the genus by Sutton (1984) because of sessile conidium production, differs by narrower arms not thicker than 1  $\mu\text{m}$  at the base and the presence of mostly five subglobose cells per conidium (Boedijn, 1951). The conidia of *T. triradiata*, a species of doubtful status (Sutton, 1984), possess three aseptate arms and four globose to cylindrical cells (Hansford, 1946).

**Key to species of *Titaea* based on conidial characteristics derived from the literature (including some species with a doubtful position within the genus). Parts of conidia differentiated in a basal cell, septate or aseptate arms, and subglobose to ellipsoidal cells situated on the basal cell or on the arms.**

1. Conidial arms arranged in one plane .....2
1. Conidial arms arranged three-dimensionally .....8
  
2. At least one conidial arm directed downwards,  $\pm$  parallel to the basal cell .....3
2. Conidial arms not directed downwards .....4
  
3. Conidia with one arm directed downwards ..... *T. callispora* Sacc.
3. Conidia with two arms directed downwards ..... *T. miconiae* (F. Stevens) Damon
  
4. Conidial arms not septate .....5
4. At least one conidial arm septate .....7
  
5. Conidia with more than one subglobose to ellipsoidal cell .....6
5. Conidia not with more than one subglobose to ellipsoidal cell .....  
..... *T. volucriata* K. Matsush. & Matsush.
  
6. Arms not directly arising from the basal cell . *T. formosa* Peláez, R.F. Castañeda & Arenal
6. Arms directly arising from the basal cell ..... *T. hemileiae* Hansf.
  
7. Conidia with more than 1 subglobose to ellipsoidal cells ..... *T. doidgeae* Hansf.
7. Conidia with only one subglobose to ellipsoidal cell ..... *T. clarkeae* Ellis & Everh.
  
8. Conidial arms 0.5-1  $\mu$ m wide ..... *T. costaricana* (Syd.) Boedijn
8. Conidial arms wider than 1  $\mu$ m .....9
  
9. Conidial arms septate .....10
9. Conidial arms not septate .....11
  
10. Conidia with only one subglobose to ellipsoidal cell ..... *T. clarkeae* Ellis & Everh.
10. Conidia with more than one subglobose to ellipsoidal cells. *T. tetrabrachiata* R. Kirschner
  
11. Conidia without subglobose to ellipsoidal cells ..... *T. pes-avis* Cif.
11. Conidia with subglobose to ellipsoidal cells .....12
  
12. Subglobose cells with spines ..... *T. toddaliae* Hansf.
12. Subglobose to ellipsoidal cells smooth .....13
  
13. Conidia with four arms ..... *T. complexa* K. Matsush. & Matsush.
13. Conidia with three arms ..... *T. triradiata* Hansf.



**Fig. 3.** *Bahusaganda indica*. Conidiophores (bar = 10 µm) and conidia (bar = 20 µm).

***Bahusaganda indica*** (Subram.) Subram.

(Fig. 3)

*Colonies* dark brown, pulverulent, up to 1.6 mm wide and confluent up to several cm long. *Hyphae* superficial, smooth, light brown, 2-3 µm diam. *Conidiophores* composed of 0-2 basal elongate, smooth cells and a torulose, simple or sparsely branched chain of subglobose to ellipsoidal, strongly echinulate cells, some of them collapsing, becoming dark brown and cupulate after conidium dehiscence, conidiophores rarely with interspersed smooth, elongate cells, medium to dark brown, 20-50 × 3-6 µm. *Conidia* solitary, brown, echinulate with spines 0.5-1 µm long, torulose, with 3-16 constricted septa, (33-)55-112(-143) µm long (n = 30), 13-17 µm at the broadest part, narrowing to the terminal cells (7-10 µm at their broadest part), individual cells 7-15 µm long.

*Habitat*: On dead herbaceous stem.

*Known distribution*: India (Subramanian, 1958; Subramanian and Srivastava, 1994), Panama (new record).

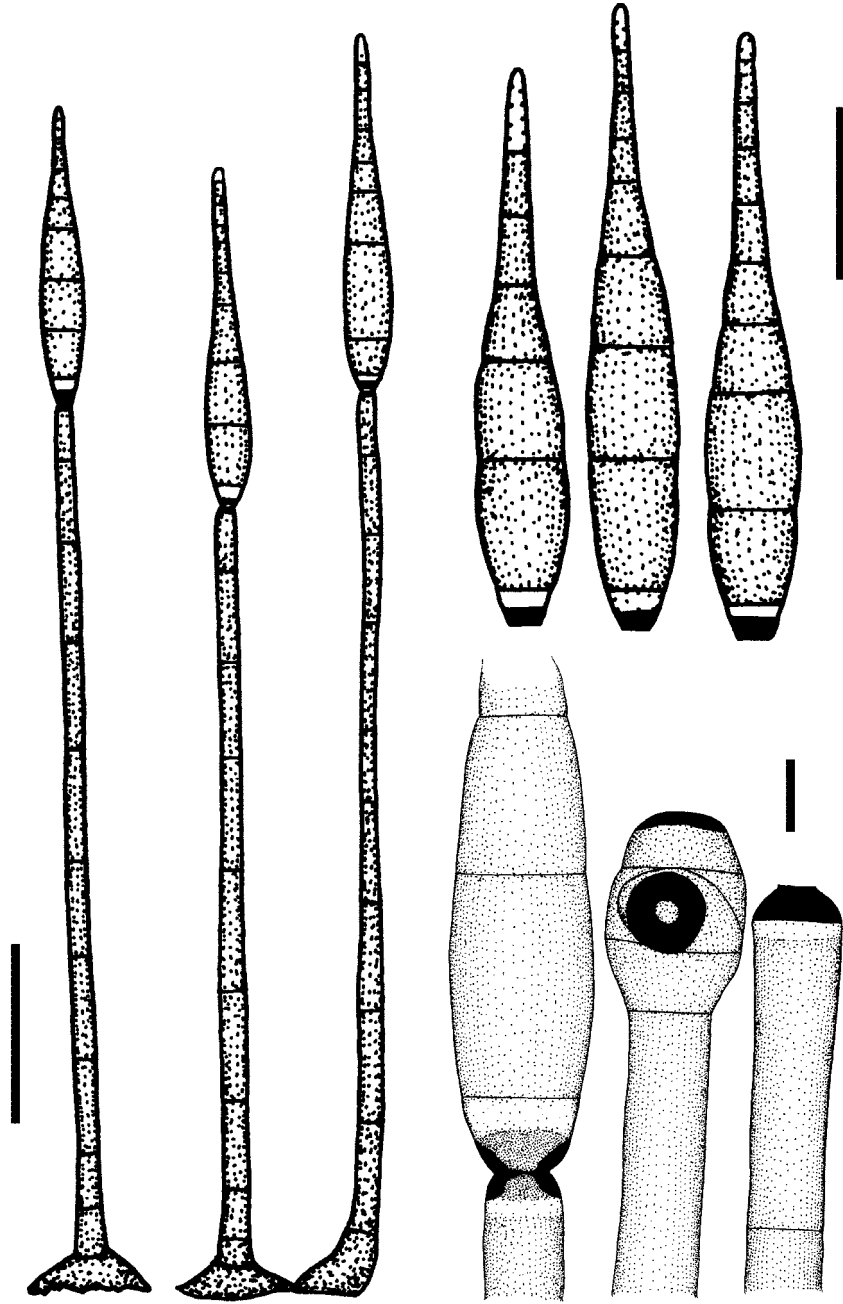


Fig. 4. *Exosporium ampullaceum*. Conidiophores with still attached conidia (bar = 100  $\mu\text{m}$ ), conidiophore apices (bar = 10  $\mu\text{m}$ ), and conidia (bar = 50  $\mu\text{m}$ ).



*Material examined:* PANAMA, Chiriquí, Parque Internacional de la Amistad, Sendero El Retoño, ca. 2.300 m, on dead herbaceous stem, 3 March 2003, R. Kirschner & M. Piepenbring 1788 (PMA).

*Notes:* This species, hitherto not known outside India (Subramanian, 1958; Subramanian and Srivastava, 1994), was first described in *Deightoniella* and later transferred to *Bahusaganda* by Subramanian (1958). It differs from typical species of *Deightoniella* by strongly torulose, echinulate conidiophores and conidia. These characteristics are typical for species of *Dwayabeeja*, *Polyschema*, and *Torula*, but conidia are dimorphic in *Dwayabeeja* species (Subramanian, 1958; Mercado Sierra, 1980) and are catenate and usually narrower in *Torula* species (Ellis, 1971). The conidiophores of *Polyschema* species are generally shorter and lack intermediate non-sporogenous cells as well as the cupulate appearance of the conidiogenous cells after conidium dehiscence (Ellis, 1976; Matsushima, 1980; Crane, 2001). This type of conidiogenous cell is considered significant for the close relationship between species of the genera *Bahusaganda*, *Dwayabeeja*, and *Torula* (Crane, 2001). The presence of non-sporogenous cells between successive conidiogenous cells, considered a diagnostic characteristic for the genus *Bahusaganda* by Subramanian (1958) and (Subramanian and Srivastava, 1994), was rarely found in the specimen from Panama.

***Exosporium ampullaceum*** (Petch) M.B. Ellis (Fig. 4)

*Colonies* appearing pilose by the presence of scattered conidiophores. *Stromata* composed of dark brown hyphae forming a basal plate at the bases of the conidiophores, 15.5-22  $\mu\text{m}$  high and 38.5-66  $\mu\text{m}$  diam. *Hyphae* extending from the stromata into the substratum subhyaline to pale brown, smooth, 2-3.5  $\mu\text{m}$  wide. *Conidiophores* mostly solitary, in some cases 2-3 conidiophores connected at their bases in a common stroma, unbranched, erect, brown, smooth, 390-484  $\times$  11-20  $\mu\text{m}$ , distances between septa 34-68  $\mu\text{m}$ ; conidiogenous cell terminal, apically mostly slightly swollen, with an apical, blackened, 2-4  $\mu\text{m}$  thick scar, in some cases with additional 1-2 lateral scars and often with a subhyaline to light brown, ca. 2  $\mu\text{m}$  thick zone below the blackened apex. *Conidia* solitary, obclavate to rostrate, smooth or irregularly roughened, with 4-9 slightly constricted transverse septa, brown, basal and terminal cells pale brown or subhyaline, proximal half of the basal cell blackened, (105-)142-178(-194)  $\mu\text{m}$  long, (22-)23-28(-31)  $\mu\text{m}$  wide at the broadest part, narrowing to the terminal cell (4-5.5  $\mu\text{m}$  at its broadest part).

*Habitat:* On dead herbaceous stem.

*Known distribution:* subtropical and tropical countries; Cuba, Ghana, Mexico, Micronesia, Peru, Sierra Leone, Sri Lanka, Taiwan (Ellis, 1971;

Matsushima, 1981, 1987, 1993; Heredia *et al.*, 1997; Mercado Sierra *et al.*, 1997). Panama (new record).

*Material examined*: PANAMA, Chiriquí, Parque Internacional La Amistad, Sendero de la Cascada, ca. 2.300-2.500 m, on dead herbaceous stem, 4 March 2003, R. Kirschner & M. Piepenbring 1778 (PMA).

*Notes*: This species can be easily recognised by the conspicuously paler area of the basal cell between the blackened base and the dark brown main body of the conidium (Ellis, 1971).

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