

## HYALOTIASTRUM GEN. NOV., AN ADDITION TO COELOMYCETES FROM PAKISTAN

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### Abstract

*Hyalotiastrum* gen. nov., et *H. salvadorae* sp. nov., belonging to Coelomycetes are illustrated, described and compared with related taxa.

### Introduction

During examination of specimens of *Salvadora oleoides* collected from Karachi, a Coelomycete with three septate, hyaline conidia, and 4-6 apical cellular, unicellular, enucleate, simple appendages was found. This taxon, although closely resembling *Hyalotiella* Papendorf, *Hyalotiopsis* Punithalingam, *Parahyalotiopsis* Nag Raj, *Alpakesa* Subram. & Ramakr., *Alpakesiopsis* Abbas, Sutton, Ghaffar & Abbas. *Pseudorobillarda phragmitis* (Cunnell) Morelet and *Neoalpakesa Punithalingam* is distinctly different. Further, it is also compared with other related genera.

*Hyalotiastrum* Abbas, Sutton, Ghaffar et Abbas gen. nov.

Fig. 1 (A, B, C, D, E, F)

Fig. 2 (A, B, C, D)

**Etym.** *Hyalotia* et *astrum* (L. suff.) incomplete resemblance.

*Conidiomata* pycnidialia vel eustromatica, separata, 4-5 pycnidialia aggregata, nigra, sphaerica vel applanato-globosa vel convoluta vel irregularia, parietes ex strato duobus ex textura angulari compositi, nigri, generatim paries exterior cellula parva consistantes, crassi ad nigri, versus stratum interius hyalini. In conidiomata multilocularia, parietes interloculares, ex cellulis tenuioribus hyalini consistantes. *Conidiophora* absentia. *Cellulae conidiogenae* ex cellulis interioribus parietorum formatae, hyalinae, lageniformes vel cylindratae, discretae, indeterminatae, cylindricae, laeves, proliferationibus. enterogenibus et progressivis, *Paraphyses* hyalinae, laeves, septatae, producentes et inter cellulae conidiogenae formatae. *Conidia* hologenitica, cylindrica vel fusiformia, hyalina, 3 septata, leviter curvata, omnibus cellulis binucleatis, cellula apicali obconica, crassa, basim truncata, 4-6 appendicibus apicalibus tubulosis, hyalinis.

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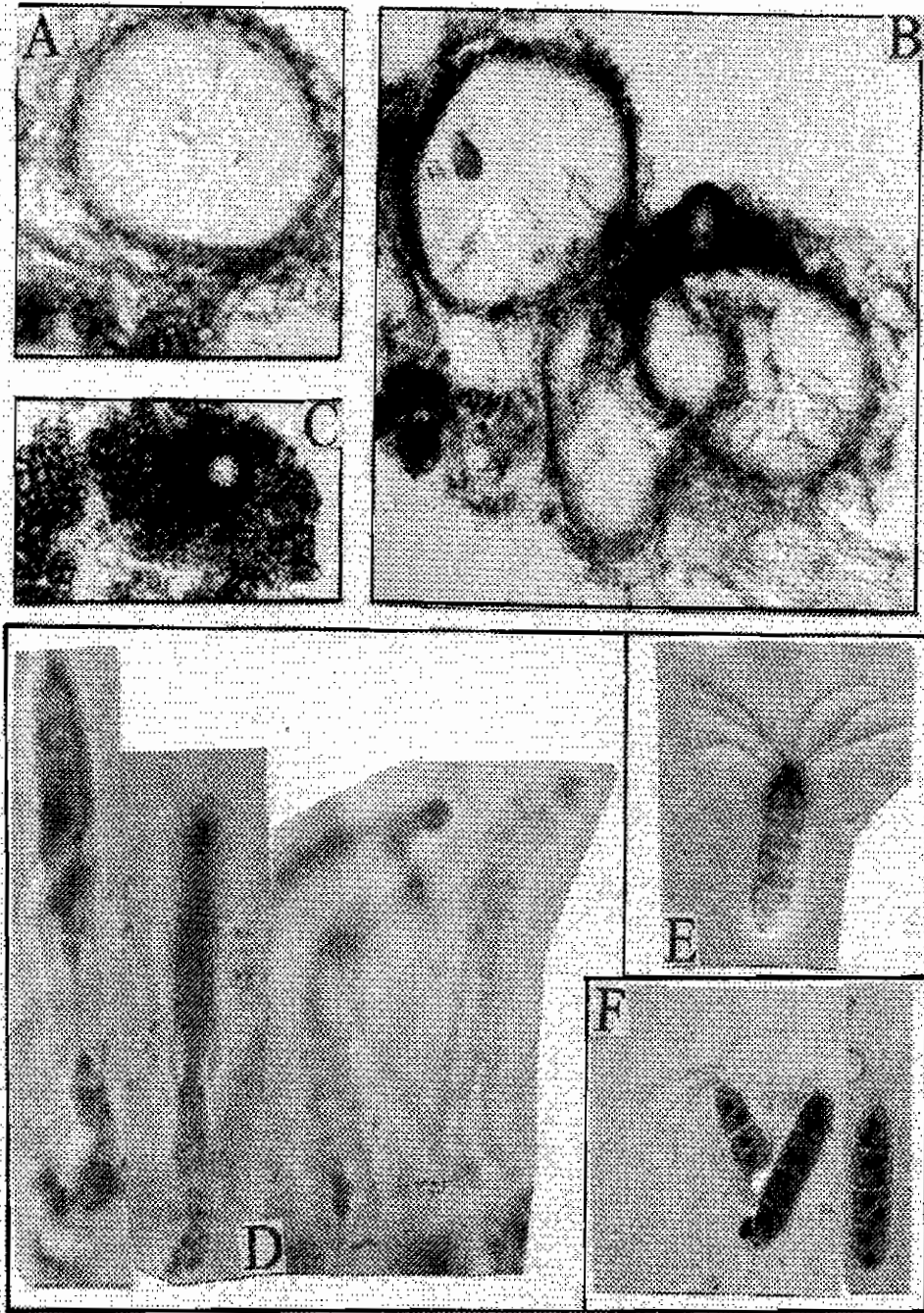


Fig. 1. *Hyalotiastrum salvadorae*. A. V.S. of simple conidioma, 40X; B. V.S. of aggregated conidiomata, 40X; C. Ostiole, 40X; D. Conidiogenous cells, 1800X; E. Conidium, 1800X; F. Conidia stained in Geimsa HCl 1800X.

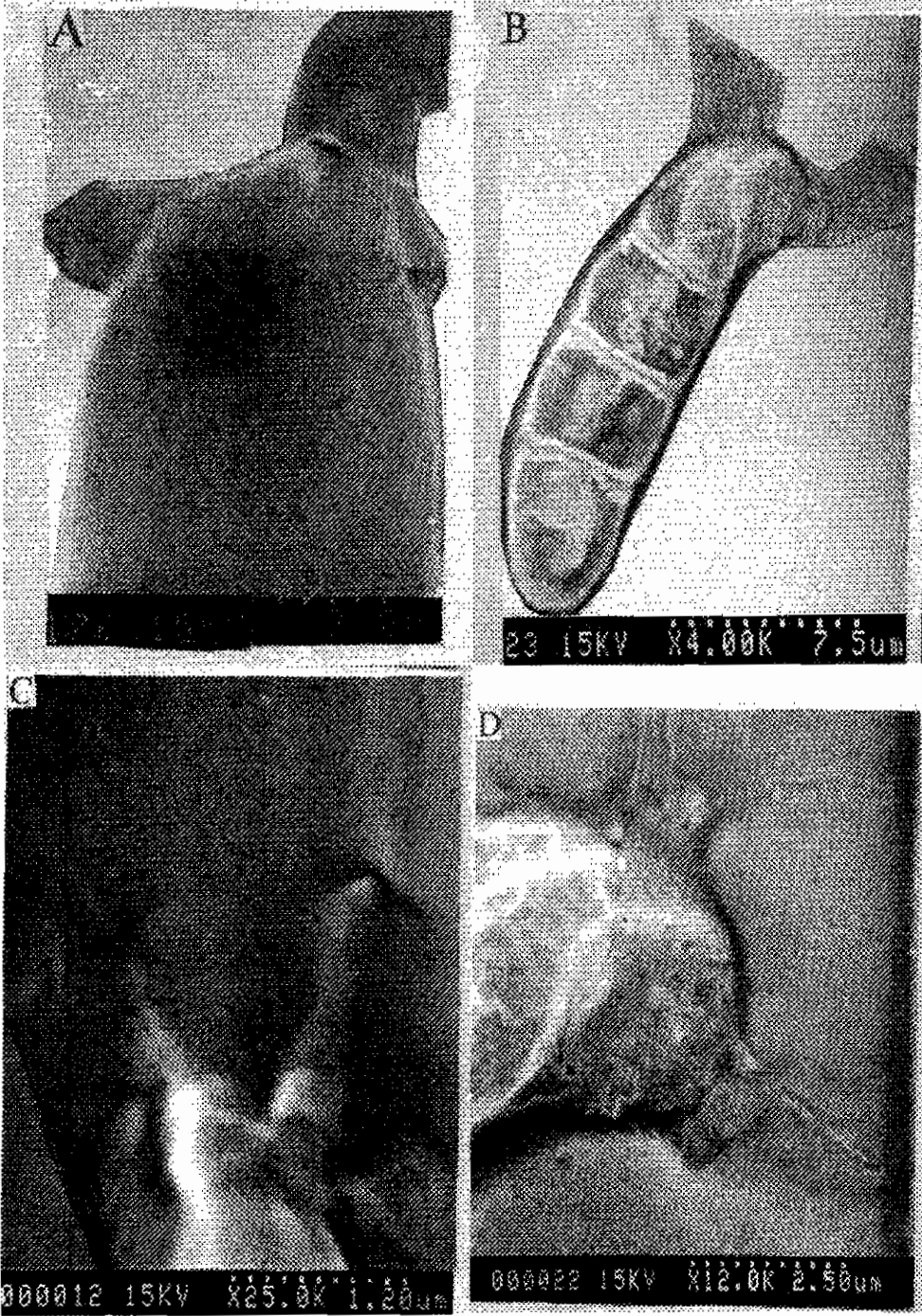


Fig. 2. *Hyalotiastrum salvadorae*. (A, B, C, D) Conidia in SEM.

**Sp. typ.:** *Hyalotiastrum salvadorae* Abbas, Sutton, Ghaffar et Abbas sp. nov.

***Hyalotiastrum* Abbas, Sutton, Ghaffar and Abbas gen nov.**

*Conidiomata* eustromatic, separate or aggregated, black, spherical or appanate-globose, unilocular to multilocular, wall two layers thick of *textura angularis*, consisting of a thick-walled, pale brown outer layer, and a thin-walled hyaline inner layer. *Conidiophores* absent. *Conidiogenous cells* formed from the innermost layer of cells all over the pycnidial cavity, hyaline, lageniform to cylindrical, discrete, smooth, proliferating enterogenous and progressively, intermingled with hyaline, smooth, septate paraphyses. *Conidia* hologenous, 3-septate, cylindrical to fusiform, slightly curved, each cell binucleate, apical cell obconical, pointed with some thickening and by optical microscopy 4-6 apical hyaline appendages are seen originating from a common point. By SEM 2-3 appendages were found to originate separately from two different collar like structures.

*Doliomyces* Steyaert (Sutton, 1980) resembles *Hyalotiastrum* in having conidiogenous cells which proliferate enterogenous and progressively and appendaged conidia. It can easily be separated by the pycnidial conidiomata, simple conidiophores, cylindrical to navicular, unequally 3-5 septate conidia with apical and basal cells hyaline, median cells dark brown and thick-walled, the apical cell with 4-7 lateral appendages and the basal cell with a single excentric, exogenous, cellular appendage. In contrast to this, *Hyalotiastrum* has eustromatic conidiomata, no conidiophores, paraphyses, 3-septate, hyaline, cylindrical, conidia with 4-6 apical appendages and no basal appendage.

*Hyalotiastrum* also resembles *Discosia* Lib., in having eustromatic conidiomata, the absence of conidiophores and 3-septate, hyaline appendaged conidia. However it differs in the enterogenous and progressively proliferating conidiogenous cells and the presence of paraphyses in contrast to non-proliferating conidiogenous cells and absence of paraphyses in *Discosia*. Conidia of both genera are hyaline and 3-septate. In *Discosia* one suprabasal and one subapical appendage is present (Sutton, 1980), whereas conidia in *Hyalotiastrum* have 4-6 apical, simple, hyaline, cellular appendages originating from two collar-like structures, each having 2-3 appendages which arise separately. *Pseudorobillarda phragmitis* (Cunnell) Morelet also resembles *Hyalotiastrum* in the absence of conidiophores, and 3 septate, hyaline, appendaged conidia and the presence of paraphyses (Punithalingam & Woodhams, 1986). However *P. phragmitis* can easily be separated from *Hyalotiastrum* in having pycnidial conidiomata, enterogenous and stationary conidiogenous cells and 3 or sometimes 4 apical, hyaline, parietal appendages. Under optical microscopy, appendages look as if they are formed by the splitting of the cell wall from basal to apical ends (Punithalingam & Woodhams, 1986) but this is not true, because studies under the scanning electron microscopy revealed that 3 or 4 appendages arise from a protuberance at the apex as various branches joining a slightly larger branch from the upper side. *Pestalozzina* (Sacc.) Sacc., is another genus (Nag Raj, 1974) which also resembles *Hyalotiastrum* in having enterogenous and progressively proliferating conidiogenous cells and appendaged conidia, but it can be differentiated by the pycnidial conidiomata, presence of conidiophores and absence of paraphyses. Furthermore conidia of *Pestalozzina* are 4-septate, the individual conidial cells unequal and not uniformly pigmented. Apical and basal cells are hyaline, while the

median cells are pale brown and longer than the apical and basal cells. The apical cell has several simple, hyaline, cellular appendages, arising all over its upper part. In contrast to this, *Hyalotiastrum* has eustromatic conidiomata, conidiogenous cells proliferating enterogenous and progressively. Conidia are 3-septate, hyaline, with 4-6 simple, cellular, enucleate appendages, 2-3 appendages arising separately from two collar-like structures present on the upper part of the apical cell at two different loci towards the margin.

*Bartalina* Tassi (Sutton, 1980) is another genus which has some similarities with *Hyalotiastrum* in having appendaged conidia, no conidiophores and enterogenous and progressively proliferating conidiogenous cells. It differs in the pycnidial conidiomata, absence of paraphyses and four septate, pale brown conidia with one basal and 2-3 apical cellular appendages arising from the upper part of the apical cell. *Alpakesa* Subram. & Ramak., also resembles *Hyalotiastrum* in having 0-3-septate, hyaline, appendaged conidia and no conidiophores. However it differs in the pycnidial conidiomata, non-proliferating conidiogenous cells and absence of paraphyses (Sutton, 1980). Species belonging to *Alpakesa* are 0-3-septate but the main characteristic of the conidia is that the apical cell has a gap-like structure below the appendages. This creates some confusion as to whether the appendages originate from the outer gelatinous sheath or not. Critical studies (Abbas *et al.*, 1998) by light and scanning electron microscopy revealed that the appendages are cellular, having a clear connection to the inner and outer cell wall and cytoplasm. It was also observed that in conidia with an apparently wide gap, there is in fact a wall below the gap and the gap is in reality a small cell, with very little cytoplasm and no nucleus. This cell does not properly stain by conventional methods such as cotton blue and acid fuchsin, however, using a modified Leifson's bacterial staining technique (Punithalingam & Woodhams, 1984) the nature of appendages, cytoplasmic connections and inner and outer walls of appendages become very clear. In *Alpakesa* conidial cells are uninucleate and the upper surface from which appendages originate is very broad. *Hyalotiastrum* can easily be distinguished from *Alpakesa* by having eustromatic conidiomata, enterogenous and progressively proliferating conidiogenous cells, 3-septate, hyaline conidia, an apical conical cell and the absence of small cells with less cytoplasm between appendages. Such cells are visible in *Alpakesa*. Furthermore in *Hyalotiastrum*, the surface from which appendages arise is very narrow, appearing only as a pin-point by optical microscopy. However, by scanning electron microscopy there are two separate collar-like structures of opposite direction from which 2-3 appendages separately arise.

*Hyalotiella* Papendorf (Nag Raj, 1974; Sutton, 1980) resembles *Hyalotiastrum* in having eustromatic conidiomata and 3-septate, appendaged conidia, but it differs in having conidiophores, generally non-proliferating but sometimes hologenous and sympodially proliferating conidiogenous cells, absence of paraphyses and conidia not uniformly pigmented, apical cell hyaline and small, basal cells small and pale brown whereas median cells are pale brown and longer than the apical and basal cells. The apical cell is prolonged into a cellular, hyaline appendage which branches trifurcately. In contrast to this *Hyalotiastrum* is characterized by no conidiophores, enterogenous and progressively proliferating conidiogenous cells and hyaline conidia with 4-6 apical, cellular appendages.

*Hyalotiopsis* Punithalingam (teleomorph *Ellurema* Nag Raj & Kendrick) is another genus which resembles *Hyalotiastrum* in the absence of conidiophores, and 3 septate, appendaged conidia (Abbas *et al.*, 1998; Punithalingam, 1970; Nag Raj, 1975; Nag-Raj & Kendrick, 1985). It differs in having pycnidial conidiomata, generally non-proliferating conidiogenous cells which sometimes do proliferate hologenous and sympodially, and in the absence of paraphyses, the versicoloured pigmented conidia, conidial cells which are unequal but multinucleate and the apical and basal cells which are hyaline and longer than the pale brown median cells. From the apical cells two appendages arise which divide to form 4-6 branched appendages. In contrast to this *Hyalotiastrum* has eustromatic conidiomata, enterogenous and progressively proliferating conidiogenous cells with 3 septate hyaline conidia, the cells of which are binucleate, uniform in size and pigmentation and have 4-6 apical cellular simple appendages. Superficially the appendages of *Hyalotiastrum* resemble *Hyalotiopsis* under optical microscopy, however scanning electron microscopy (Abbas *et al.*, 1998) revealed that the appendages in both genera are very different. In *Hyalotiastrum* 4-6 appendages arise separately from two different collar-like structures, each with 2-3 appendages, while in *Hyalotiopsis* the two apical appendages divide repeatedly and are dichotomously branched.

*Parahyalotiopsis* Nag Raj (Nag Raj, 1976; Abbas *et al.*, 1998) is another genus with some resemblance to *Hyalotiastrum*, in having enterogenous and progressively proliferating conidiogenous cells and 3 septate conidia with 2-4 apical simple appendages. However, it differs in having pycnidial conidiomata, and 3-septate brown conidia, the cells of which are uninucleate. Appendages in both genera are apical, cellular and enucleate, but they differ in ontogeny. In *Parahyalotiopsis*, the apical cell is truncate and 2-4 appendages arise separately from different loci, whereas in *Hyalotiastrum* 2-3 appendages arise separately from two collar-like structures present on the apical part of the conical apical cell. *Chaetospora* Faurel & Schotter (Sutton, 1980) is another genus which resembles *Hyalotiastrum* in having hyaline, septate, appendaged conidia and no conidiophores but it can easily be differentiated by the pycnidial conidiomata, non-proliferating conidiogenous cells, absence of paraphyses, 4-septate conidia with unequal median cells and 3-4 apical, simple, cellular, 1-5 septate appendages. In contrast to this, *Hyalotiastrum* has conidia which are 3-septate, hyaline, the cells, uniform in size, and 4-6 simple, cellular, enucleate appendages arising from two collar-like structures at the apex, from each of which 2-3 appendages arise separately. *Nealpakesa* Punithalingam (1981) is another Coelomycete genus comparable with *Hyalotiastrum*. It can easily be differentiated by having pycnidial conidiomata, paraphyses and aseptate conidia. Its superficial resemblance lies in the appendages which arise in both the genera from two different loci but in *Nealpakesa* the distance between both loci is much greater than it is in *Hyalotiastrum*. By optical microscopy, appendages in *Hyalotiastrum* look as if they are coming from a point but scanning electron microscopy shows that there are actually two collar-like structures from which 2-3 appendages arise separately whereas in *Nealpakesa* two appendages, from two different loci arise and then these appendages divide dichotomously. *Ciliochorella* Syd., (Sutton, 1980) also resembles *Hyalotiastrum* in having eustromatic conidiomata, no conidiophores and septate appendaged conidia, but it differs in bearing subcuticular or

subepidermal, lenticular conidiomata, no paraphyses, conidiogenous cells proliferating enterogenous and stationary and conidia which are 3-septate with median cells very pale brown and longer than the apical and basal cells. The apical cell has two simple cellular appendages, one of which is apical and the other lateral. The basal cell also has one simple cellular appendage. *Japonia* Höhn., (Sutton, 1980) is similar to *Hyalotiastrum* in having hyaline appendaged conidia. However it differs in the pycnidial conidiomata, presence of conidiophores, non-proliferating conidiogenous cells, absence of paraphyses and 2-septate conidia in which the median cell is longer and the apex extended into a single cellular, irregularly branched appendage. *Chrysalidopsis* (Steyaert, 1961; Nag Raj & DiCosmo, 1980) resembles *Hyalotiastrum* in having appendaged conidia, enterogenous and progressively proliferating conidiogenous cells, but it differs by the pycnidial conidiomata, presence of conidiophores, absence of paraphyses and 4-septate conidia. The apical cell is hyaline whereas all the other cells are dark brown, thick-walled and verruculose. The apical cell is extended into a single cellular appendage with 2-4 branches.

*Alpakeiopsis* Abbas, Sutton, Ghaffar and Abbas recently described by Abbas *et al.*, (2003) also resembles *Hyalotiastrum* in having eustromatic unilocular to multilocular conidiomata and 3-euseptate hyaline appendaged conidia, but it differs in the presence of conidiophores, enterogenous stationary conidiogenous cells and absence of paraphyses. Conidial morphology, origin of appendages and number of appendages also differ in both genera. In *Alpakesiopsis* conidia are fusiform, with the apex about twice as wide as the base and the wall thicker at both ends, conidia 3-5-septate, uninucleate with 6-9 apical, simple, cellular appendages which originate from separate loci. In *Hyalotiastrum*, conidia are more or less cylindrical, base obtuse, wider than the acute apex, 3-septate, conidial cells binucleate, with 2-3 simple cellular appendages arising separately from two collar-like structures at the apices and from two different loci.

*Hyalotiastrum* also differ from *Alpakeiopsis*, a genus recently described by Abbas *et al.*, (2003).

***Hyalotiastrum salvadorae* Abbas, Sutton, Ghaffar & Abbas sp. nov.**

**Fig. 1 (A, B, C, D, E, F)**

**Fig. 2 (A, B, C, D)**

*Conidiomata* eustromatica, separata vel aggregata, nigra, sphaerica vel applanato-globosa, unilocularia vel multilocularia, 179-336 x 152-201  $\mu\text{m}$ , parietes 4-8 cellulis crassi ad 14-72 lati ex textura angulari ad stratis duobus compositi, paries exterior 4-8  $\mu\text{m}$  latus, pallide-brunneus, stratum interius 10-64  $\mu\text{m}$  latum, tenuiora versus centrum gradatim hyalinum. *Conidiophora* absentia. *Cellula conidiogenae* ex cellulis interioribus parietonum formatae, hyalinae, lageniformes vel cylindricae, discretae, indeterminatae, laeves 5-6.8x2.4  $\mu\text{m}$ , enterogenitibus progressivis 1-4 proliferationibus. *Paraphyses* hyalinae, laeves 1-septatae 64x1.6  $\mu\text{m}$ , producentes et inter cellulae conidiogenae formatae. *Conidia* hologentica, cylindrica vel fusiformia, 3 septata, hyalina, leviter curvata cum cellulis binucleatis, 21.6-28.8 x 5.2-6.4  $\mu\text{m}$ , apicem obconica, crassa, basim truncata, 4-6 appendicibus apicalibus (28x0.5-0.6  $\mu\text{m}$ ).

In ramis emortuis *Salvadorae oleoidis* S.Q. Abbas UCMH 758 (IMI-320302), holotypus.

***Hyalotiastrum salvadorae* Abbas, Sutton, Ghaffar & Abbas sp. nov.**

*Conidiomata* eustromatic, separate or aggregated, black, spherical or applanate-globose, unilocular to multilocular, 179-336 x 152-201  $\mu\text{m}$ , wall 4-8 cells thick 14-72  $\mu\text{m}$  wide, of two layers of *textura angularis*, the outer layer pale brown and 4-8  $\mu\text{m}$  wide, the inner layer thin-walled, 10-64  $\mu\text{m}$  wide, becoming hyaline towards the centre. The innermost layer forms conidiogenous cells all around the conidiomatal cavity. *Conidiophores* absent. *Conidiogenous cells* hyaline, lageniform to cylindrical, smooth, 1-4, proliferating, enterogenous and progressively 5-6.8 x 2.4  $\mu\text{m}$ , intermingled with hyaline smooth, 1-septate parapyses, which are 64 x 1.6  $\mu\text{m}$ . *Conidia* hologamous, 3-septate, hyaline, cylindrical to fusiform, slightly curved, each cell binucleate, thick-walled, apical cell conical, basal cell truncate, 21.6-28.8 x 5.2-6.4  $\mu\text{m}$ , appendages 4-6 (28 x 0.5-0.6  $\mu\text{m}$ ) apical, simple, hyaline, unicellular, enucleate, 2-3 originating separately from 2 collar-like structures.

**Specimen examined:*****Hyalotiastrum salvadorae* Abbas, Sutton, Ghaffar & Abbas sp. nov.**

On twigs of *Salvadora oleoides*, S.Q. Abbas UCMH 758 (IMI-320302), holotype.

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