

## Notes on Southern African Tuberales

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### ABSTRACT

Three species of Tuberales have been found in Southern Africa. *Terfezia pfeilii* Henn. occurs in the Kalahari Desert and adjacent areas of the Cape Province, Botswana and South-West Africa. The other two, *Terfezia austroafricana* sp. nov. and *Choiromyces echinulatus* sp. nov., are known only from the Cape. *C. echinulatus* is the first representative of that genus to be collected in Africa or the Southern Hemisphere.

### INTRODUCTION

The first record of Tuberales from Southern Africa is Henning's (1897) description of *Terfezia pfeilii* Henn. from Damaraland, South-West Africa. The occurrence of this species in Damaraland was later noted by Pole Evans (1918); the cracks in the Kalahari sand resulting from growth of its hypogeous ascocarps were illustrated by Leistner (1967).

Two other species, *Terfezia boudieri* Chat. and *T. claveryi* Chat., were reported from the Kalahari and Windhoek (Marloth, 1913; Pole Evans, 1918). The "*Terfezia claveryi*" that they examined as well as all others so labelled in the Mycological Herbarium of the National Herbarium, Pretoria (PRE), were subsequently redetermined as *T. pfeilii* by Dr P. H. B. Talbot (unpublished). Marloth did not describe his "*Terfezia boudieri*" and Pole Evans apparently did not see any specimens. We have been unable to locate any collections that were labelled "*Terfezia boudieri*" by either. Accordingly, until now the only member of the Tuberales authenticated for Southern Africa is *T. pfeilii*. The closest record of a different species is that of *Terfezia decaryi* Heim in Madagascar (Heim, 1934).

While noting that three species of *Terfezia* have been reported from South Africa and South-West Africa, Story (1958) stated that "some records are vague and incomplete and cannot be checked." In view of the confusion about these species, we restudied the Tuberales at PRE. Trappe's earlier studies of the types and supplementary collections of the entire order of Tuberales, including the Terfeziaceae (Trappe, 1971) provided interpretive background. Collections of Southern African Tuberales were also kindly provided by the Herbaria of the Botanical Institute of the University of Torino (TO), Oregon State University (OSC), and the U.S. National Fungus Collections (BPI). This paper should be regarded as a preliminary contribution, since further collecting in Southern Africa will almost certainly produce additional hypogeous fungi.

The collections examined were either dried or, in the case of some at PRE, preserved in ethanol-formalin-glycerine-water solutions. Tissues and spores were examined in these mounting media: (1) 5% KOH, (2) cotton blue-lactic acid, (3) Melzer's reagent, and (4) lactophenol, the mount being heated gently over a gas flame. Spores were drawn from the cotton blue mounts. Spores measured the same in all mounting media.

### Key to the Tuberales of Southern Africa

- Asci mostly cylindrical, borne in crowded hymenial layers; spores mostly uniseriate, prominently echinulate, 12-18 $\mu$  broad (including spines)..... 1. *Choiromyces echinulatus*
- Asci subglobose, ellipsoid, reniform, or asymmetric; spores mostly biseriate to irregularly arranged, mostly broader than 18 $\mu$  (including ornamentation):
  - Spores minutely echinulate, mostly 18-23 $\mu$  broad (including spines)..... 2. *Terfezia pfeilii*
  - Spores prominently spinose-reticulate, 25-30 $\mu$  broad (including spines)..... 3. *Terfezia austroafricana*

#### 1. *Choiromyces echinulatus* Trappe & Marasas, sp. nov.

Ascocarpa viva eburnea, in statu sicco nigrescens. Gleba solida, marmorata, hymeniis inclusis. Asci cylindrici, 140  $\times$  12-17 $\mu$ , octospori, in hymeniis mutue compressis. Sporae globosae, 12-18 $\mu$  latae (cum ornamentis), echinulatae bactulis et conis 1-2  $\times$  0,5-1 (-1,5) $\mu$ . Peridium cellulis inflatis multitis.

TYPE: Cape, Gordonias near Upington, in red sand dune, June 1, 1961, Leistner 2612 (PRE 42202, holotype).

Ascocarp pale cream-colored and subglobose when fresh, as dried with a black peridium and a solid gleba marbled by dark brown veins embedding hymenial palisades; opposing hymenial palisades deformed from pressing against each other.

Hymenium of thin-walled, mostly collapsed paraphyses 4 $\mu$  broad growing among hyaline, thin-walled asci. Asci mostly 8-spored; in youth cylindrical to ellipsoid or saccate and with spores irregularly arranged; by maturity mostly cylindrical, 140  $\times$  12-17 $\mu$ ,

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hyaline, thin-walled, with stems long tapered below the basal spores to narrow bases and spores uniseriate or occasionally biseriata.

*Spores* globose, 12–18 $\mu$  broad with ornamentation, 10–14 $\mu$  excluding ornamentation, light yellow to light brownish yellow. Walls 1 $\mu$  thick, light blue in cotton blue. Ornamentation of round-tipped rods and cones 1–2  $\times$  0,5–1(–1,5) $\mu$ , pale yellow in KOH and light blue in cotton blue, ca 20–25 around the spore circumference, unconnected except for some spores on which barely perceptible lines on the spore surface join occasional ornaments.

*Glebal veins* of subparallel-interwoven, hyaline, thin-walled hyphae 3–6 $\mu$  broad at septa, the cells frequently inflated. *Peridium* of hyphae with light yellow, thin walls, 4–8 $\mu$  broad at septa but with many cells greatly inflated (15–50 $\mu$  broad).

CAPE.—Gordonia near Upington, in red sand dune, June 1, 1961, *Leistner* 2612 (PRE 42202, holotype).

This species has the solid gleba marbled with veins and embedded hymenia with elongate asci that typify the genus *Choiromyces*. It differs from previously described *Choiromyces* spp. in having echinulate spores. *C. venosus* (Fr.) Th. Fr., the most common species of Europe, has larger spores prominently ornamented with irregular tubes and rods. The other *Choiromyces* species have either ridged or pitted spores. *C. echinulatus* is the first member of the genus found in Africa or, for that matter, in the Southern Hemisphere.

## 2. *Terfezia pfeilii* Hennings, Engler Bot. Jahrb. 22: 75 (1897).

Ascocarps 2,5–6,5  $\times$  2,5–5,8 cm, subglobose to obpyriform or turbinate, lacking a basal mycelial tuft but with a basal attachment scar. *Peridium* c. 1 mm thick, prominently wrinkled (particularly on the upper surface), blackish brown with the wrinkles yellowish. *Gleba* yellowish white, fleshy, solid, marbled with white veins. Odour rather strongly fungoid.

*Asci* randomly arranged in fertile pockets, (5–) 8-spored at maturity, typically subglobose but sometimes ellipsoid or obovoid, 70–100  $\times$  50–80 $\mu$ , hyaline, thin-walled, sessile to substipitate, readily separable from the glebal hyphae, spores loosely arranged within.

*Spores* globose, (16–)18–23(–26) $\mu$  with ornamentation, at first hyaline, by maturity pale brown. Walls 1,5 $\mu$  thick, 2-layered, blue in cotton blue. Ornamentation appearing as a minutely papillose, mucilaginous-granulose epispore in KOH or lactophenol but clearly seen as densely crowded, minute, deeply staining spines 1–1,5(–2) $\mu$  tall in cotton blue.

*Glebal fertile pockets* separated by sterile but otherwise undifferentiated veins, the hyphae hyaline, thin-walled, 5–12 $\mu$  broad at septa but the cells generally inflated (up to 20 $\mu$ ) to appear pseudoparenchymatous. *Peridium* with an outer layer of large hyphae 4–12 $\mu$  broad at septa but with cells inflated to 15–30 $\mu$  to give a cellular appearance; inner peridial layer of generally circumferentially aligned hyphae 4–10 $\mu$  broad at septa, the cells often slightly inflated.

Fruiting from April to June in sand dunes of the Kalahari Desert and adjacent areas of the Cape, South-West Africa and Botswana.

SOUTH-WEST AFRICA.—Damaraland: *Hennings* (PRE 15021, lectotype; TO, possible isotype). Gibeon: *Burger* (PRE 42082, PRE 42203). Gobabis: *Verbücheln* (PRE 43905), Keetmanshoop: *Kinges* (PRE 36986). Kleinkaras: *Hill* (PRE 17799; BPI, *Lloyd* 38083; OSL, *Trappe* 1309; TO). Windhoek: *Gies* (PRE 42076).

BOTSWANA.—Ghanzi: *Scholtz* (PRE 41869).

KALAHARI DESERT.—*Bottomley* (PRE 44310); *Nash* (PRE 44254); *Weintraub* (PRE 32394). Askham: *Strydom* (PRE 44245). Kalahari Gemsbok National Park: *Storry* 5616 (PRE 41602); *Le Riche* (PRE 41870).

CAPE.—Kakamas: *Oosthuizen* (PRE 26335). Postmasburg: *Hunter* (PRE 11293). Prieska: *MacCleod* (PRE 11619). Upington: *Leistner* 2610 (PRE 42201). Vryburg: *Stephens* 527 (PRE 36103).

*Terfezia pfeilii* has been suggested as a synonym of several other *Terfezia* species by various European authors. Our examinations of the types of all *Terfezia* species have established beyond doubt that *T. pfeilii* is distinct and confirm Mattiolo's (1922) illustrations that clearly show the differences in spore ornamentation between *T. pfeilii* and the other species. The earlier confusion about *T. pfeilii* stems in part from the paucity of good specimens available for study by earlier authors and in part from the nature of the spore ornamentation of *T. pfeilii*. The spines are so crowded and minute that they cannot readily be seen even with an oil immersion objective unless stained.

Only two described species of *Terfezia*, *T. olbiensis* Tul. & Tul. and *T. leptoderma* Tul. & Tul., resemble *T. pfeilii* in having spores ornamented with very small spines. The spores are larger and the spore ornamentation much taller (2–3 $\mu$ ), coarser, and more openly spaced in *T. olbiensis* and *T. leptoderma* than in *T. pfeilii*. Although Mattiolo (1922) correctly illustrated the echinulate nature of spores of *T. pfeilii*, he suspected it to be synonymous with *T. pinoyi* Maire, which he also correctly illustrated as lacking spines. The two are readily separable by this difference in spore ornamentation as well as by the amyloid reaction of asci of *T. pinoyi* with Melzer's reagent and the lack of that reaction by *T. pfeilii*.

Dr Talbot's redetermination of PRE collections labeled "*Terfezia claveryi*" as *T. pfeilii*, noted earlier in this paper, was confirmed by our studies. Accordingly, *T. claveryi* is not known to occur in South Africa, notwithstanding the reports of Marloth (1913), Pole Evans (1918), and Doidge (1950).

## 3. *Terfezia austroafricana* Marasas & Trappe, sp. nov.

Ascocarpace glebis solidis, marmoratis. *Asci* ellipsoidei, obovoidei, subcylindrici, reniformes, vel asymmetrici, 90–140  $\times$  30–80 $\mu$ , plerumque octospori. *Sporae* globosae, 25–30 $\mu$  latae (cum ornamentis), spinosae-reticulatae, spinis (2–)3–5(–6) $\mu$  altis. *Peridia* cellulis inflatis multis.

TYPE: Cape, Griqualand West near Barkly West, E. L. *Stephens* (PRE 35577, holotype).

Dried ascocarps orange brown to brownish black, smooth; dried gleba brown in young specimen, ochraceous in older specimen, solid, marbled with pallid veins.

*Asci* randomly arranged in fertile pockets, (4–)8-spored at maturity, ellipsoid to obovoid, subcylindric, reniform or asymmetric, 90–140  $\times$  30–80 $\mu$ , hyaline,



thin-walled, astipitate or with a short basal protuberance; spore arrangement occasionally uniseriate but mostly incompletely biseriata to irregular.

*Spores* globose, 25–30 $\mu$  broad with ornamentation, 16–22 $\mu$  excluding ornamentation, hyaline in youth and pale yellow at maturity. Walls 1–2 $\mu$  thick, light blue in cotton blue. Ornamentation of truncate to round-tipped spines (2–)3–5(–6)  $\times$  1–3 $\mu$ , connected by walls to form a partial to complete reticulum of 4–6 sided, irregularly sized alveoli; alveoli 3–7 across the spore diameter; reticular walls 0.5 $\mu$  thick, variable in height from very low to as high as the spines. Occasional spores ornamented with rounded warts and no reticulum or with crowded spines only erratically connected by low walls.

*Glebal fertile pockets* separated by sterile but otherwise undifferentiated veins, the hyphae hyaline, thin-walled, 5–12 $\mu$  broad at the septa but the cells generally inflated (up to 22 $\mu$ ). *Peridium* of hyphae 10–30 $\mu$  broad at septa, the cells mostly inflated (up to 60 $\mu$ ) to give a cellular appearance.

CAPE.—Griqualand West: Barkly West, E. L. Stephens (PRE 35577, holotype); Kimberley, April 10, 1918, Wilman (PRE 11542, paratype).

*Terfezia austroafricana* belongs to subgenus *Mattitrolomyces* (Fischer) Trappe by virtue of its large, elongate asci and uncrowded spores (Trappe, 1971). The other species presently assignable to this subgenus—*T. decaryi*, *T. terfezioides* (Matt.) Trappe, and *T. spinosa* Harkn.—also have prominently reticulate spores. Of these, *T. spinosa* of North America most closely resembles *T. austroafricana* in microscopic characters (the fresh ascocarps have not been described for either); *T. austroafricana*, however, has fewer, larger, and more regular alveolae on the spore surface

and larger, more inflated cells in the peridium and gleba. The spores of *T. austroafricana* are larger than those of *T. terfezioides*, and the asci are smaller than those of *T. decaryi*. All of these species are closely related in anatomical characters, but only *T. terfezioides* is known from a large number of collections. As the others are collected again and become better known, some may prove to merit only varietal status.

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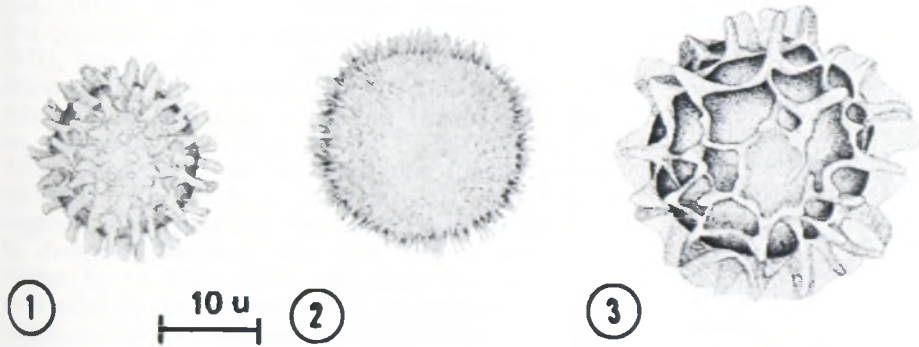


FIG. 1–3.—Ascospores. Fig. 1, *Choiromyces echinulatus*, PRE 42202. Fig. 2, *Terfezia pfeilii*, PRE 17799. Fig. 3, *Terfezia austroafricana*, PRE 15542.

