

An enlarged concept of *Trichobolus* (Thelebolaceae, Pezizales) based on a new eight-spored species¹

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A new eight-spored species, *Trichobolus octosporus*, is described and illustrated. The generic concept has been enlarged to include this taxon. A revised generic description and key are provided. The species is discussed in relation to previously known organisms within the genus.

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Une espèce nouvelle de *Trichobolus*, formant huit spores, est décrite et illustrée, le *T. octosporus*. Les limites du genre ont été élargies de façon à inclure ce nouveau taxon. Une description générique révisée et une clef sont fournies. Les relations de l'espèce décrite avec les représentants déjà connus du genre sont discutées.

Introduction

Until Kimbrough and Cain (1967) raised *Trichobolus* (Sacc.) Kimbr. & Cain to the generic level, this taxon was considered as a subgenus of *Thelebolus* Tode ex Fr. Kimbrough (1966a) was able to show that *Trichobolus* differed from *Thelebolus* in a number of structural and morphological features. In a recent study on the response of growth to temperature in *Thelebolus*, Wicklow and Malloch (1971) have shown that the response to temperature in *Trichobolus zukalii* (Heimerl) Kimbr. is quite different from that in their isolates of *Thelebolus*. This observation would appear to support the removal of *Trichobolus* from *Thelebolus*. At present both genera are placed in the Thelebolaceae of the Pezizales.

Our current concept of *Trichobolus* includes those Discomycetes whose apothecia are fimicolous, small, and setose with the setae each possessing one to several septa. The asci are ovoid, multispored, and few in number, and rupture irregularly. The ascospores are one-celled, and vary from subspherical to subellipsoidal. Kimbrough (1967) added two additional species to the taxon and slightly emended the description but did not alter to any degree the original concept of the genus as based on *T. zukalii*.

As has occurred on a number of previous

occasions, an eight-spored representative has been found which, except for the number of ascospores, would seem to belong to a hitherto multispored genus. This species appears to fit the concept of *Trichobolus* in most other respects. Therefore, it is necessary to enlarge the concept of this taxon as given by Kimbrough and Cain (1967). In addition, a new generic description and key are provided along with the description of this additional species. The previously recognized members of the genus have been described and discussed by Kimbrough (1966a, 1967).

Descriptions

Trichobolus (Sacc.) Kimbr. & Cain in Kimbr. & Korf, *Am. J. Bot.* **54**: 20. 1967, descr. emend.
≡ *Thelebolus* Tode ex Fr. sect. *Setosae* Heimerl, *Jahr. k. k. Ober-Realsch. Bezirke Sechshaus*, Wien, **15**: 30. 1889.

≡ *Thelebolus* Tode ex Fr. subg. *2 Trichobolus* Sacc., *Syll. Fung.* **10**: 35. 1892.

Apothecia setose, solitary to gregarious, superficial, sessile, small, subglobose to discoid, closed when immature, sometimes remaining so until rupturing at spore liberation. *Excipulum* of several layers, outer layers pallid to brownish, of texture angularis, inner layers hyaline, of either texture angularis or texture prismatica. *Setae* stiff, pointed, broader at the base, with 1–10 or more septa, concentrated in the upper portion of the fruit body. *Asci* 1–40 in number, operculate, ovoid to cylindrical, broadly rounded to truncate at the apices, not blueing in Melzer's reagent

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but staining uniformly in Congo Red. *Paraphyses* abundant in young ascocarps but usually difficult to find at maturity, often embedded in a mucilaginous matrix, long, filamentous, frequently branched. *Ascospores* eight or more per ascus, one-celled, subspherical to subellipsoidal, smooth, slightly thick-walled when young but becoming thin-walled at maturity, deBary bubbles present in some but not all species. No conidial state known.

ETYMOLOGY: Greek, *thrix* ($\theta\rho\iota\xi$) = hair, and *bolus* ($\beta\omega\lambda\omicron\varsigma$) = a clod of earth, as in *Ascobolus*, referring to the hairy nature of the ascocarp.

HABITAT: Known from deer, moose, sheep, goat, and rabbit dung.

TYPE SPECIES: *Thelebolus zukalii* Heimerl.

DISTRIBUTION: In Austria from near Vienna (TYPE); in Canada from Alberta, British Columbia, Manitoba, Ontario, and Quebec; in U.S.A. from Colorado, Massachusetts, New York, and Wyoming.

***Trichobolus octosporus* Krug, sp. nov. Figs. 1-6**

Apothecia setosa, gregaria aut raro plusve minusve dispersa, superficialia, sessilia, subglobosa, primum clausa, per maturitatem in crateriformia (sive crasse discoidea) facta, minuta, (200-)300-400 \times (300-)400-600 μ diametris crassa, aurantia aut cinnabarina. Hymenium 75-125 μ crassum. Excipulum iodo rufescens tinctum, textura angulari, 50-70 μ crassum, exterioribus in partibus fuscum, interiori in parte pallidum aut hyalinum; excipuli cellulae angulatae, exterioribus in excipuli partibus cum parietibus crassis, elongatiores, 5-8 \times 5-6 μ magnae, interiori in parte cum parietibus tenuibus, minores. Setae (10-)20-35, solum summa in apothecii parte sitae, rigidae, acuminatae, (0-) 1-2 (raro 3) basilaribus septis praeditae, parietibus 1.0-1.5 μ crassis, 50-125 μ longae, 5-7 μ

diametro basin versus crassae. Asci 25-40 in omni apothecio, octospori, primum clavati, maturitate confirmata plusve minusve cylindracei, 110-130 \times 17.5-20(-22.5) μ magni, summa in parte truncati aut plusve minusve rotundati et annulo praediti, "Congo Red" aequabiliter tincti, iodo non caerulescentes, parietibus 2.0-2.5 μ crassis, superne tenuioribus, stratis tribus sistentibus; stratum internum hyalinum; strata duo externa "Acid Fuchsin" aequabiliter tincta. Paraphyses matrice mucilaginosa circumdatae, primum numerosae, maturitate confirmata raras, filiformes, laxe ramosae, septatae, guttulate, 1-2 μ diametro crassae, ascis longiores, circa ascorum apices inaequaliter patentes. Ascospores unicellulares, subglobosae aut raro subellipsoideae, hyalinae, glabrae, 12-13 \times 10-12 μ magnae, uniseriales, summa in ascorum parte saepe biseriales, parietibus 0.5-1(-2) μ crassis, sine vagina gelatinosa, nec bullulis.

HOLOTYPE: In cervorum fimo, apud Bosler, in Albany comitatu, in Wyomingensibus finibus, in imperio U.S.A., 1 Sept. 1964, Cain, TRTC 43801. In Torontoensis Universitatis Cryptogamarum Herbario.

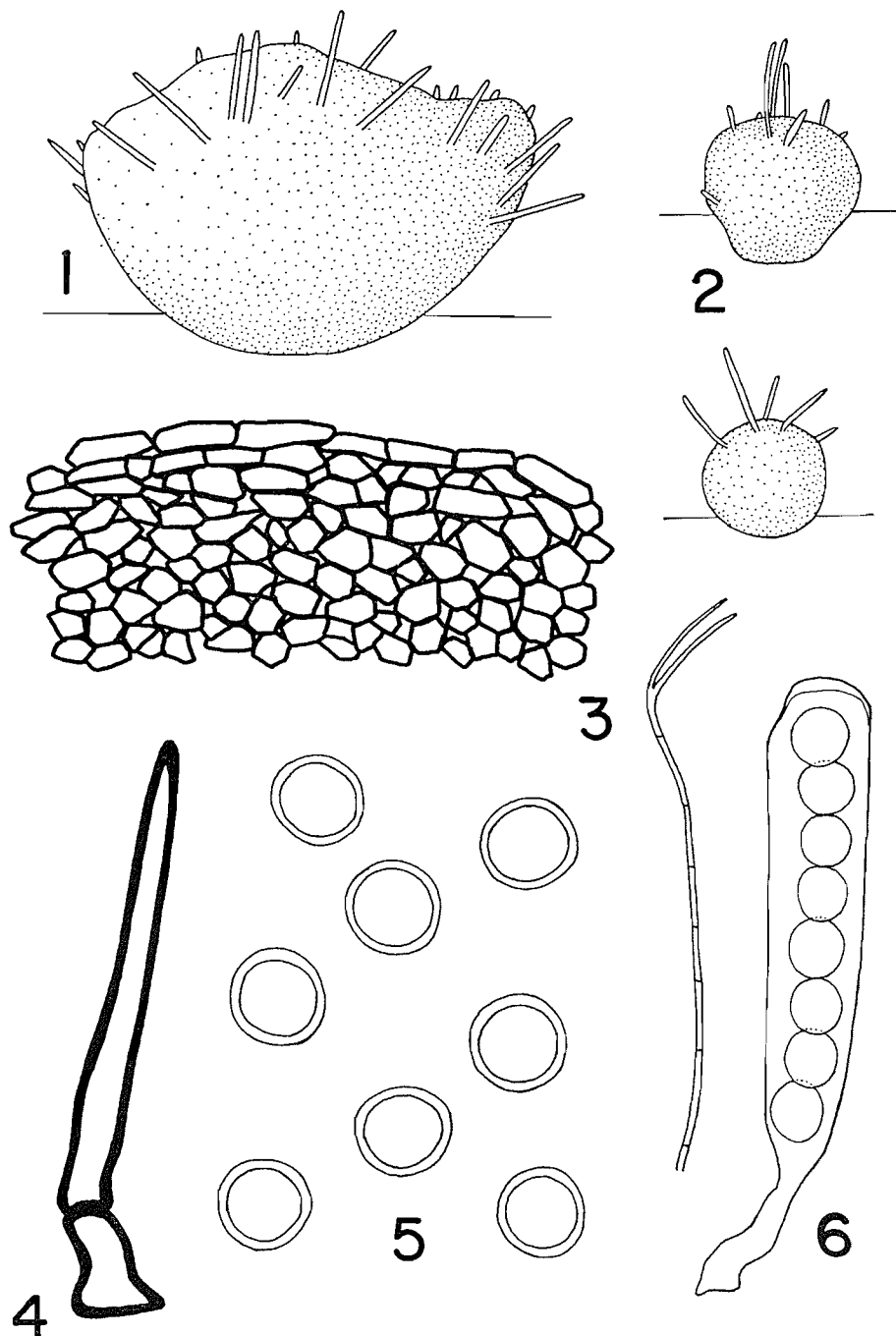
ETYMOLOGY: Greek, *okto* ($\omicron\kappa\tau\omega$) = eight, and *spora* ($\sigma\pi\omicron\rho\alpha$) = seed, referring to the eight-spored asci.

Apothecia setose, gregarious to occasionally somewhat scattered, superficial, sessile, subglobose, closed at first, becoming somewhat bowl-shaped (thickly discoid), minute, (200-)300-400 \times (300-)400-600 μ , orange to orange-red. *Hymenium* 75-125 μ thick. *Excipulum* dextrinoid, of textura angularis, 50-70 μ thick, brownish towards the exterior, pallid to colorless in underlying layers; excipular cells angular, thick-walled, 5-8 \times 5-6 μ , somewhat elongated towards the exterior, becoming smaller and thin-walled towards the interior. *Setae* (10-)20-35 in number,

KEY TO THE SPECIES

For descriptions of the three multispored species, which are not treated in the text, see Kimbrough (1966a, 1967).

1. Apothecia with many asci; setae with one to three septa; asci eight-spored, cylindrical; ascospores subspherical, without deBary bubbles *T. octosporus*
1. Apothecia with relatively few asci; asci multispored, ovoid.....2
2. Apothecia each possessing three asci; setae with a single basal septum; ascospores subspherical to subellipsoidal, with deBary bubbles..... *T. pilosus*
2. Apothecia each possessing a single ascus; setae with three or more septa.....3
3. Ascospores subspherical to subellipsoidal, each with a prominent deBary bubble..... *T. zukalii*
3. Ascospores spherical to subspherical, without deBary bubbles..... *T. sphaerosporus*



FIGS. 1-6. *Trichobolus octosporus* (TRTC 43801). Fig. 1. Mature apothecium shown in lateral view, $\times 100$. Fig. 2. Two immature stages in the development of the apothecium showing the cleistothecial nature of the young ascocarp, $\times 100$. Fig. 3. Section through a portion of the excipulum showing the more elongated exterior cells and the inner angular cells, $\times 1000$. Fig. 4. Single excipular hair, $\times 1000$. Fig. 5. Mature ascospores, $\times 1000$. Fig. 6. Mature ascus with ascospores and an associated paraphysis, $\times 500$.

concentrated in the upper portion of the ascocarp, stiff, pointed, with (zero-)one-two or rarely three basal septa, walls 1.0–1.5 μ thick, 50–125 μ long, 5–7 μ diameter at the base. *Asci* eight-spored, clavate when young, becoming almost cylindrical, 25–40 in each apothecium, truncate to somewhat rounded at the apices, with each possessing a definite apical ring, 110–130 \times 17.5–20(–22.5) μ , staining uniformly in Congo red, not blueing in Melzer's reagent; the wall 2.0–2.5 μ diameter, becoming thinner towards the apex, of three layers, the innermost hyaline and the outer two staining uniformly in acid fuchsin. *Paraphyses* enclosed within a mucilaginous matrix, abundant in young ascocarps, usually difficult to locate at maturity, filamentous, freely branching, septate, guttulate, 1–2 μ diameter, longer than the asci, spreading about the apices of the asci. *Ascospores* one-celled, subspherical or occasionally subellipsoidal, hyaline, smooth, 12–13 \times 10–12 μ , uniseriate, frequently becoming biseriate near the apex of the ascus, with walls 0.5–1(–2) μ diameter, without gelatinous sheath, deBary bubbles absent.

Colonies on V-8 vegetable juice agar growing moderately slowly at room temperature, spreading broadly, thin, cottony, white, forming no aerial hyphae, producing clusters of orange fruiting bodies sporadically and very slowly, reverse uncolored; on phytone – peptone – potato dextrose agar (Miller 1966) growing fairly rapidly, spreading broadly, densely cottony, white, forming a dense growth of aerial hyphae, reverse brownish except around the margins where it is orange-yellow. No conidial state formed.

HABITAT: On deer dung.

SPECIMEN EXAMINED: U.S.A.: Wyoming: Albany Co., Bosler, 1 Sept. 1964, Cain, TRTC 43801 (TYPE).

Isolates derived from the original culture have been dried and will be deposited at BPI, DAOM, K, and PC.

This species differs from those previously described in that there are only eight spores in the ascus, each apothecium contains many asci which are cylindrical as opposed to being ovoid in the other species, there is a definite apical ring in the apex of the ascus, and the excipulum is definitely dextrinoid. Furthermore, there is usually only one basal septum on each seta, although occasionally two or rarely even three may be found.

The presence of a distinct apical structure in the ascus is probably an adaptation for more controlled spore discharge, and the cylindrical nature of the ascus is likely correlated with the reduction in number of ascospores. The dextrinoid reaction in the excipulum is also found in certain representatives of *Lasiobolus* Sacc. Here, observations in this laboratory have shown that this reaction is extremely variable, depending, for example, on age of the ascocarp and mounting conditions. The significant differences between *T. octosporus* and the other species would appear to be the number of spores in the ascus and number of asci in the ascocarp. However, Kimbrough (1966b) has shown that such features, which traditionally have been used to separate genera, are extremely variable. It is not surprising, therefore, to find a further example of a genus with both eight and multi-spored representatives.

Variability in the number of setal septa has already been reported in two of the other species of *Trichobolus*. The occasional absence of any noticeable septum in *T. octosporus* might indicate a relationship with *Lasiobolus*. However, in most respects this species appears to be closer to *Trichobolus* since most of the other distinctions from *Lasiobolus* mentioned by Kimbrough (1966b) appear to be valid. If *T. octosporus* is related to *Lasiobolus*, this species may represent a transitional form between these genera. This might lead one to believe that the generic separation between these two taxa is unnatural. However, Kimbrough (1972), in evaluating various ascus structures in coprophilous *Discomycetes*, found that the two genera are quite distinct in this respect. On the basis of further, yet undescribed, eight-spored species of *Trichobolus* he distinguishes between members of *Trichobolus* and *Lasiobolus*. With the report of a uniascal species of *Lasiobolus*, Kimbrough concludes that *Lasiobolus* and *Trichobolus*, in addition to *Thelebolus*, all contain species ranging from those with uniascal apothecia and multi-spored asci to those with comparatively numerous eight-spored asci.

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