

## *Talbotiomyces*, a new genus for *Entorrhiza calospora* (Basidiomycota)

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**Abstract.** A short overview of the genus *Entorrhiza* is presented. Based on host plant taxonomy and morphological characters, including septal pore structure, a new genus, *Talbotiomyces*, is described for the aberrant *Entorrhiza calospora*. This fungus produces galls on various dicotyledonous host plants, whereas *Entorrhiza* spp. sensu stricto are restricted to monocotyledonous hosts.

**Key words:** *Entorrhiza*, new combination, smut fungi, *Talbotiomyces calosporus*, taxonomy

### Introduction

*Entorrhiza* C.A. Weber is a small genus of plant parasitic microfungi, with 13 known species (comp. Zundel 1953: 232-234; Fineran 1978; Vánky 2002: 54-55). The species produce galls on roots of plants in the *Cyperaceae* and *Juncaceae*. In the hypertrophied host plant cells thick-walled, variously ornamented, pale coloured spores are produced. Spore germination is unique, with a four-celled basidium remaining included in the spores; each basidial cell developing a branch that bears apically and subapically up to four falcate, looped or curved basidiospores (comp. Fineran 1982; Bauer *et al.* 2001). Host-parasite interaction is by local interaction zones, lacking an interaction apparatus, and coiled, septate haustoria are present. The septal pore is a dolipore without membranous plates or caps (Bauer *et al.* 1997). *Entorrhiza* occupies a basal position within the class *Ustilaginomycetes* R. Bauer, Oberw. & Vánky (Begerow *et al.* 1998) and it has been classified in its own subclass of *Entorrhizomycetidae* R. Bauer & Oberw.

Talbot (1956) described and illustrated a curious parasitic fungus, *Entorrhiza calospora* P.H.B. Talbot, producing galls on the roots of several dicotyledonous host plants in South Africa. Repeated attempts by Talbot to obtain spore

germination, under various conditions, failed. The sorus and spore characters correspond well with those of *Entorrhiza*, but, its dicotyledonous host plants in the Centrospermae (e.g. *Molluginaceae*, *Aizoaceae*, *Portulacaceae*) exclude it with certainty from this genus.

The story and characters of this fungus can best be shown by the words of Talbot (1956: 453), in the summary of his paper: "A fungus described here as *Entorrhiza calospora* sp. nov. and assumed to belong to the *Tilletiaceae* in the absence of proof by germination of the chlamydospores, is associated with large galls on the roots of *Limeum glomeratum*, *L. viscosum* and *Trianthema pentandra* . . . . The galls range in diameter from less than 1 mm to 3.5 cm. They are composed of parenchymatous cells in which the fine mycelium, often coiled, bears globose, strongly verrucose, binucleate chlamydospores 17.4-22 µm in diameter, produced singly but forming groups of up to twenty per host cell. The dry chlamydospores are lemon yellow to golden in a mass. A dusty sorus of chlamydospores is not formed; instead the spores are liberated by the gradual disintegration of the galls when the host dies in autumn. Germination of the spores and re-infection of the hosts were not observed. The parasite does not harm the plants as a whole, since its aerial parts show no sign of disease nor of stimulation."

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Fig. 1. Sori of *Talbotiomyces calosporus* producing galls on the roots of *Limeum glomeratum* (isotype). Habit. To the right a part of a healthy plant. Bar = 1 cm

Thirumalachar & Whitehead (1968: 185) examined *Entorrhiza calospora* and considered it to be a species of *Protomycopsis* Magnus (*Protomycetaceae* Gray, *Taphrinales* Gäum. & C.W. Dodge). Below, we discuss some characteristics of this plant parasite, which leads us to the conclusion that it belongs in a new genus.

## Materials and Methods

Sorus and spore characteristics were studied using dried herbarium specimens. To soften the tissues, pieces of mature sori were boiled in a mixture of distilled water and lactophenol with cotton blue, on a microscope slide, and then hand

sectioned with a razor blade under a stereo microscope. Thin sections showing the structure of sori and the spores were mounted in a droplet of lactophenol with cotton blue on a slide, covered with a cover glass, gently heated to eliminate air bubbles, and examined by a light microscope (LM) at 1000× magnification. For scanning electron microscopy (SEM), broken pieces of sori were placed on double-sided adhesive tape, mounted on a specimen stub, sputter-coated with gold-palladium, c. 20 nm, and examined in a SEM at 10 kV. Specimens studied were those which Talbot also studied, preserved in Pretoria, PREM 33 770 (type), PREM 35 291 (topotype), PREM 39 031 (paratype), PREM 41 034 (paratype), as well as H.U.V. 587 (isotype), and PREM 41 932 (lacking sori). For details see below.

The ultrastructure of the spores, hyphae, and septa of the isotype specimen (H.U.V. 587) was studied with a Zeiss EM 109 transmission electron microscope at 80 kV. Samples were fixed overnight with 2 % glutaraldehyde in 0.1 M sodium cacodylate buffer (pH 7.2) at room temperature. Following six transfers in 0.1 M sodium cacodylate buffer, samples were postfixed in 1 % osmium tetroxide in the same buffer for 1 h in the dark, washed in distilled water, and stained in 1 % aqueous uranyl acetate for 1 h in the dark. After five washes in distilled water, samples were dehydrated in acetone, using 10 min changes at 25 %, 50 %, 70 %, 95 %, and 3 times in 100 % acetone. Samples were embedded in Spurr's plastic and sectioned with a diamond knife. Serial sections were mounted on formvar-coated, single-slot copper grids, stained with lead citrate at room temperature for 5 min, and washed with distilled water.

## Results and Discussion

*Talbotiomyces* Vánky, R. Bauer & Begerow, **gen. nov.** (Mycobank 510473)

*Pertinet ad phylum Basidiomycota cum poris simplicibus septalibus et cum haustoriis, in radicibus plantarum nutrientium dicotyledonearum gallas producens. Sporae intracellulares, ornamentatae, subhyalinae, leniter pigmentosae (carens colorem brunneam, nigrum, rubellum vel violaceum).*

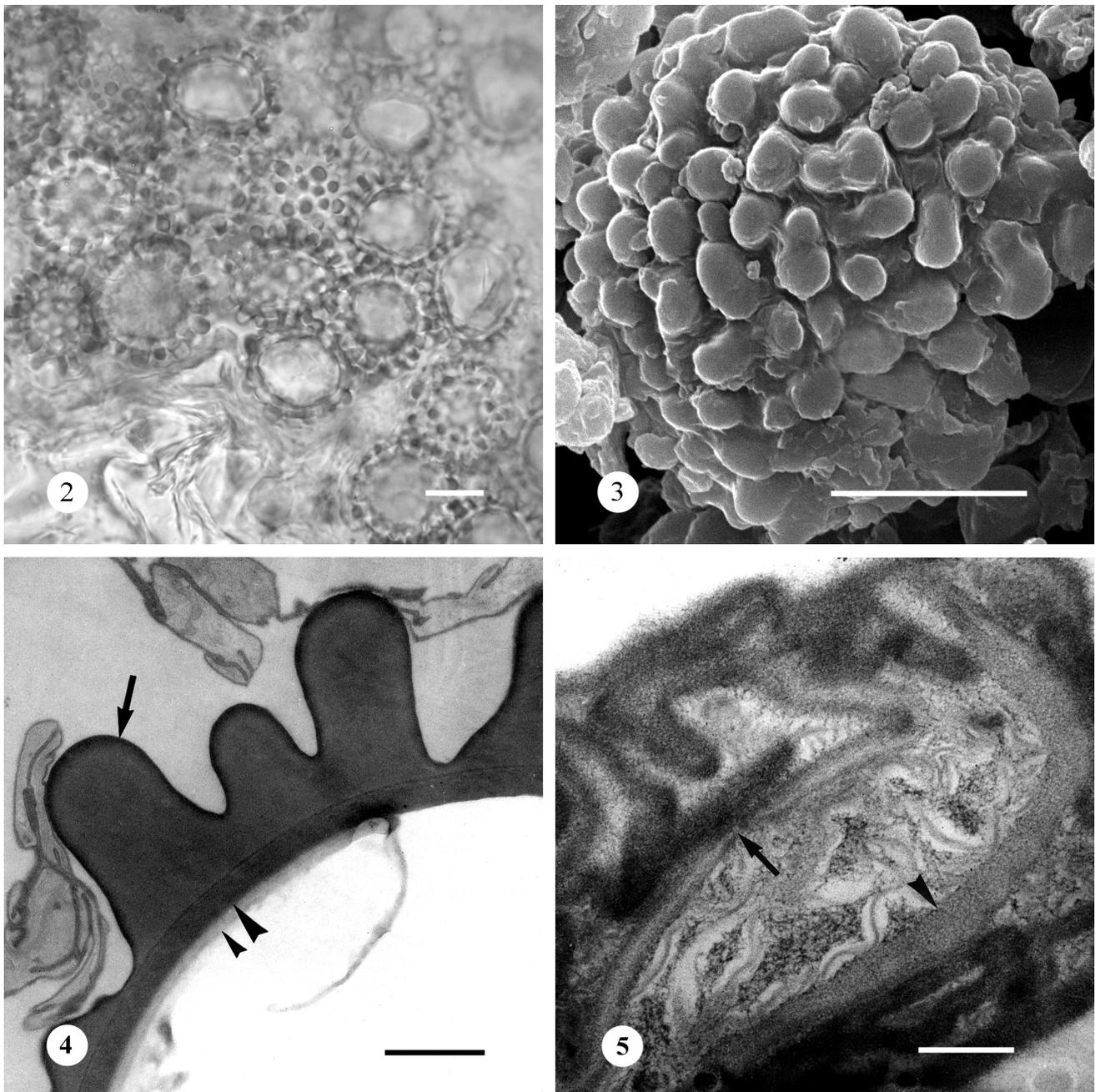
*Typus generis: T. calosporus.*

Member of the phylum *Basidiomycota*, having simple septal pores and haustoria, parasitising dicotyledonous host plants (in *Centrospermae*), producing galls on the roots. Spores intracellular, ornamented, subhyaline, weakly pigmented (lacking brown, black, reddish or violet colour).

Type of the genus:

*Talbotiomyces calosporus* (P.H.B. Talbot) Vánky, R. Bauer & Begerow, **comb. nov.** (Mycobank 510474).

Basionym: *Entorrhiza calospora* P.H.B. Talbot, *Bothalia* 6: 453, 1956. — Type on *Limeum glomeratum* (*Molluginaceae*), South Africa, Transvaal, Pretoria, Brummeria, near Murray Farm, 30 Jan 1943, leg. J.J.O. Pazzi. Holotype PREM 33 770, isotype H.U.V. 587. Topotypes on *L. glomeratum* (= *L. viscosum*



**Figs 2-5.** *Talbotiomyces calosporus* on *Limeum glomeratum* (from isotype). **Fig. 2.** Section of a part of a sorus with spores developed intracellularly in hypertrophied host cells. Cut in lactophenol with cotton blue, in LM. Bar = 10  $\mu$ m. **Fig. 3.** Spore from a ruptured sorus, showing the cylindrical warts of variable size, with a rounded top (from isotype), in SEM. Bar = 10  $\mu$ m. **Fig. 4.** Spore wall with endospore (small arrowhead), exospore (large arrowhead), and warts (arrow), in TEM. Bar = 2  $\mu$ m. **Fig. 5.** Section through a hypha showing the electron-opaque, fibrillate hyphal wall (arrowhead) and a tripartite septum with a central simple pore (arrow), in TEM. Note that the pore is not enclosed by caps or plates. Bar = 0.2  $\mu$ m

var. *glomeratum*), Brummeria, spring 1944, J.J.O. Pazzi, PREM 35 291 & 35 391. Paratypes on *Limeum viscosum*, Orange Free State, Ficksburg, Mar 1955, leg. Dr. Meredith, PREM 41 034; on *Trianthema pentandra* (*Aizoaceae*), Orange Free State, near Malaty, 8 Apr 1945, leg. B.N. Wolff, PREM 39 031.

**Sori** (Fig. 1) on the roots, forming galls from 0.5-35 mm in diameter, first smooth, later rough, tubercular, single, globose, irregular or elongated by fusion, surrounding the root or on

one side, then asymmetrical, pale yellowish brown, compact, composed of vascular bundles and parenchymatous cells with fungal mycelium, 1-2.5  $\mu$ m wide, haustoria and intracellularly developed spores on the top of lateral branches, up to twenty per host cell. Spore mass lemon-yellow to golden-yellow, agglutinated, not powdery. **Spores** (Figs 2-3) globose, subglobose to ellipsoidal, varying in size, 14-21.5  $\times$  16-25  $\mu$ m (including ornamentation), subhyaline to pale yellow; endospore even,

0.5–0.8 µm thick, exospore ornamented with moderately densely situated cylindrical warts, measuring 1.5–2.5 (–3.5) × 1.5–4 µm (Fig. 4). Warts on the same spore variable in length and width, from short, nearly subpyramidal to long cylindrical with rounded tip; number of warts per spore diameter (4–) 5–9, on the equatorial circumference, in optical median view, 12–22 warts can be seen. **Spore germination** unknown. **Hyphae** (Fig. 5) in TEM with electron-opaque, fibrillate walls. **Septa** with the typical tripartite basidiomycetous profile. **Septal pores** simple with more or less rounded pore lips. Caps or plates on the septal pores lacking.

On *Molluginaceae*: *Limeum viscosum* (J. Gay) Fenzl, *L. viscosum* var. *glomeratum* (Eckl. & Zeyh.) Friedrich (*L. glomeratum* Eckl. & Zeyh.); on *Aizoaceae*: *Trianthema penterandra* L.; and on *Portulacaceae*: *Portulaca oleracea* L.

Distribution: South Africa.

**Etymology:** *Talbotiomyces* = Talbot's fungus, named in honour of the excellent South African mycologist and outstanding taxonomist, Patrick Henry Brabazon Talbot (1919–1979) who, as well as publishing a number of new species, took a great interest in fungal microstructure and the principles of fungal taxonomy (Talbot 1968). Talbot, *i.a.*, published the illustrated book *Principles of Fungal Taxonomy* (1971), and also described *Entorrhiza calospora* in a paper which can serve as an example for every mycologist publishing new species. He named his fungus with beautiful spores '*calospora*' (calo- = beautiful, in Greek).

*Talbotiomyces calosporus* is a peculiar parasitic microfungus. Whereas all known *Entorrhiza* species form galls on roots of *Cyperaceae* and *Juncaceae*, *T. calosporus* parasitises dicotyledonous host plants, members of at least three genera of the *Centrospermae*. It is known only from South Africa, where it seems not to be rare. It is certainly much overlooked due to the hidden sori and no sign of disease on the host plants. Its geographic distribution is probably much larger. The members of *Entorrhiza*, as well as *T. calosporus*, form coiled, septate haustoria. In addition, the morphology of the sori and spores of *T. calosporus* is very similar to that of *Entorrhiza* species. However, the ultrastructure of their septal pores is different. (For types of septal pores within the smut fungi see Vánky 2002: 12). Thus, whereas the septal pore in species of *Entorrhiza* is a dolipore without membranous plates and caps (Bauer *et al.* 1997, 2001), that in *Talbotiomyces* is a simple pore without membranous plates and caps (Fig. 5). The ultrastructure of the hyphal wall and septa clearly demonstrate that *T. calosporus* is a member of the *Basidiomycota* R.T. Moore. Within the *Basidiomycota*, however, without additional data concerning the cellular interaction, basidial morphology and/or DNA sequences it is not possible to ascribe *Talbotiomyces* to any family, order or class. Members of the subphyl. *Pucciniomycotina* R. Bauer, Begerow, J.P. Samp., M. Weiss & Oberw., the order *Entorrhizales* R. Bauer & Oberw. (subphyl. *Ustilaginomycotina* R. Bauer, Begerow, J.P. Samp., M. Weiss & Oberw.) and the order *Cystofilobasidiales* Boekhout & Fell (subphyl. *Agaricomycotina* R. Bauer, Begerow, J.P. Samp., M. Weiss & Oberw.) also have pores without membranous

caps (Bauer *et al.* 1997, 2006; Weiss *et al.* 2004). Therefore, *T. calosporus* could be a member of the *Pucciniomycotina*, *Entorrhizales* or the *Cystofilobasidiales*. Within the *Pucciniomycotina* and *Cystofilobasidiales*, *Talbotiomyces* would be unique by its soral morphology, and within the *Entorrhizales* and *Cystofilobasidiales*, *Talbotiomyces* would be unique by its simple pores. Therefore, in each case erection of a new genus for it is justified. Because of the morphological and ecological similarities between *Entorrhiza* and *Talbotiomyces* we favour a position within the *Entorrhizales*, but this suggestion must be confirmed by additional data.

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