 Contributions to the smut fungi of Africa. 6. New records of *Tilletia oplismeni-cristati*, *T. vittata*, and *T. perotidis*

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Abstract. Additional records to three *Tilletia* species are reported from Africa: *T. oplismeni-cristati* on *Acroceras calcicola* from Madagascar, *T. vittata* on *Oplismenus burmannii* from Senegal, and *T. perotidis* on *Perotis patens* from Madagascar. *Acroceras calcicola* is a new host association for *T. oplismeni-cristati*, currently known only on species of *Oplismenus*. Descriptions, illustrations, and taxonomic notes are provided for these taxa.

Key words: *Acroceras calcicola*, Africa, grasses, Madagascar, *Oplismenus burmannii*, *Perotis patens*, Poaceae, Senegal, smut fungi, taxonomy, *Tilletia oplismeni-cristati*, *Tilletia perotidis*, *Tilletia vittata*

Introduction

*Tilletia* is a large genus of smut fungi (*Ustilaginomycotina*) comprising 189 species on grasses (*Poaceae*) (Bao et al. 2010; Vánky 2011, 2013; Denchev & Denchev 2013; Li et al. 2014; Denchev, T. & Denchev 2018a, b; Denchev et al. 2018). Most commonly, their sori are produced in the ovaries, which fill with a semi-agglutinated or powdery spore mass intermixed with sterile cells. In some species, the sori are formed in leaves and culms, as streaks. Exceptionally, the sori appear as swellings on the culms or cover the surface of the leaves, or form witches’ brooms (Vánky 2013; Denchev, T. & Denchev 2018a).

For many of the currently recognized species in *Tilletia*, only one to few specimens exist. This is mostly due to the cryptic nature of most members of this genus and to the fact that its highest diversity occurs in the tropical and subtropical regions of the world.
where very few collection trips for smut fungi have been carried out and where many species of this genus are yet to be discovered. The inadequate amount of Tilletia collections hinders the assessment of the species diversity, distribution, and host spectrum. Therefore, accumulation of data for poorly known species from understudied regions of the world, like Africa, is of high interest.

**Material and methods**

Dried specimens from the herbaria of the Meise Botanic Garden, Belgium (BR; herbarium codes according to Thiers 2020+) and National Museum of Natural History, Paris (P) were examined under a light microscope (LM) and scanning electron microscope (SEM). For LM observations and measurements, spores and sterile cells were mounted in lactoglycerol solution (w : la : gl = 1 : 1 : 2) on glass slides, gently heated to boiling point to rehydrate the spores and sterile cells, and then cooled. The measurements of spores are given as min–max (extreme values) (mean ± 1 standard deviation). For SEM, spores and sterile cells were attached to specimen holders by double-sided adhesive tape and coated with gold in an ion sputter. The surface structure of spores and sterile cells was observed and photographed at 10 kV accelerating voltage using JEOL JSM-7100F LV (for Tilletia vittata) and Hitachi SU3500 (for T. oplismeni-cristati and T. perotidis) scanning electron microscopes. The descriptions below are based entirely on the specimens examined. The shapes of spores and sterile cells are arranged in descending order of frequency.

**Taxonomy**


*Pat. & Har., in sched. (nom. inval.)*]


Sori in ovaries of some spikelets of infected plant, exceeding glumes, elongated, curved on top, ca 2.5 × 0.8 mm, covered by a thick, grayish brown, hispid pericarp that later ruptures exposing a powdery, blackish brown mass of spores and sterile cells. **Sterile cells** irregular, broadly ellipsoidal, subglobose or reniform, (6–)7–20(–23) × (5.5–)6.5–17(–20.5) μm, often attached to spores, usually light yellowish brown, sometimes subhyaline; cell wall usually with two, thin layers (sometimes hard for observation), 1.0–1.5(–1.7) μm thick, occasionally with papilla. In SEM on the free surface rugulose to minutely verruculose, sometimes wrinkled, on the contact surface foveolate, with patterns corresponding to the ornaments of the spore to which the sterile cell was attached. **Spores** globose, subglobose, sometimes broadly ellipsoidal, (17.5–)18–21.5(–22) × (17–)17.5–21(–21.5) (20.1 ± 0.8 ×
Figs 1–5. *Tilletia oplismeni-cristati* on *Acroceras calcicola* (P-P02329448). 1. Habit. 2, 3. Spores and sterile cells in LM (in median and surface view, respectively). 4, 5. Spores and sterile cells in SEM. Scale bars: 1 = 0.5 cm, 2, 3 = 10 μm, 4, 5 = 5 μm
Figs 6, 7. *Tilletia oplismeni-cristati* on *Acroceras calcicola* (P-P02329448) — spores and sterile cells in SEM. Figs 8, 9. *Tilletia vittata* on *Oplismenus burmannii* (BR 0000015694461). 8. Habit. 9. Spores and sterile cells in LM (in median view). Scale bars: 6, 7 = 5 μm, 8 = 0.5 cm, 9 = 10 μm
19.7 ± 0.8) μm (n = 100), dark reddish brown; spore wall (3.2–)3.4–4.3(–4.6) μm thick (including ornaments), two-layered (inner layer 0.6–1.1 μm thick), with warts aggregated in clusters; clusters (12–)13–16(–17) on equatorial circumference, in optical median view with a flattened tip, (0.6–)0.8–1.6(–2.0) μm high, in surface view appearing as irregular, darker areas; hyaline sheath not seen. In SEM, spore wall with warts aggregated in clusters, sometimes solitary; clusters consist of up to 15(–20) warts, clusters sometimes connected by low, fine ribs.


Distribution – On Poaceae: Acroceras calcicola, Oplismenus burmannii, and Oplismenus sp.; North America (Mexico), Central America (Guatemala, Costa Rica, and Guadeloupe), and Africa (Malawi and Madagascar).

Tilletia oplismeni-cristati is known to infect Oplismenus burmannii in North and Central America (Durán & Fischer 1961; Durán 1987; Piepenbring 2003; Vánky 2004, 2011) and Oplismenus sp. in Malawi (Vánky 2004; Vánky et al. 2011). This smut fungus is reported here for the first time from Madagascar, on a new host plant, Acroceras calcicola.

Oplismenus and Acroceras (Poaceae: Panicoideae) are small genera (7 and 13 species, respectively) in the tribe Paniceae, subtribe Boivinellinae (Soreng et al. 2017). The morphological characters of the smut fungus on Acroceras calcicola, especially the spore wall ornamentation, match well that of T. oplismeni-cristati (comp. Durán 1987; Piepenbring 2003; Vánky 2004). No recent collections are available to confirm by molecular data the conspecificity of the fungus on Oplismenus burmannii and Acroceras calcicola.

The immature spores of Tilletia oplismeni-cristati have light yellowish brown colour, similar to that of the sterile cells. The two-layered spore wall can be observed much easily in the light-coloured, immature spores. Occasionally, the wall of sterile cells appears laminated with up to four thin, faint, and hard for observation layers. Rarely, some spores can have a short papilla.

The spore wall ornamentation of T. oplismeni-cristati, composed of warts aggregated in clusters, is unique in the genus Tilletia and is the best morphological feature by which this species can be identified or differentiated from T. imbecillis Vánky and T. vittata (Berk.) Mundk., which also infect hosts in Oplismenus (Vánky 2004, 2011). Another species of Tilletia, T. acroceratis Vánky, is also known on Acroceras from Africa (Vánky 2011; Vánky et al. 2011), but T. acroceratis differs by its larger spores (up to 25 μm long) and the type of spore wall ornamentation (dense, polyangular projections with flattened top) (Vánky 2011).

Sori in ovaries of some spikelets of infected plant, exceeding glumes, lemon-shaped to cylindrical, often curved, 2.5–10 × 0.8–1.2 mm, with a short, acute tip, bearing rudimentary style and stigmas, covered by a thin, brown, and hispid pericarp with parallel veins. The powdery, blackish brown mass of spores and sterile cells become evident upon rupturing of the pericarp. Sterile cells variable in size, irregular, broadly ellipsoidal, subglobose or reniform, (9.5–)10.5–18.5(–21) × (9–)10–16.5(–19.5) (14.9 ± 2.6 × 13.4 ± 2.1) μm (n = 50), light yellowish brown; cell wall usually two-layered (sometimes hard for observation), 0.7–1.7(–2.0) μm thick. In SEM rugulose to wrinkled. Spores globose, subglobose, sometimes broadly ellipsoidal, (15.5–)16.5–20.5(–21.5) × (14.5–)15.5–19.5(–20.5) (18.7 ± 1.2 × 18.2 ± 1.2) μm (n = 100), dark reddish brown; spore wall indistinctly two-layered, moderately verruculose to verrucose, (1.6–)1.8–2.5(–2.8) μm thick (including the 0.4–1.0 μm high ornamentation and hardly visible 0.5–0.9 μm thick inner layer); usually covered by thin hyaline sheath. In SEM, warts densely spaced, often confluent in small groups or short rows.

Specimen examined – On Oplismenus burmannii (Retz.) P. Beauv.: SENEGAL, ZIGUINCHOR REGION, Basse Casamance (Lower Casamance), Djobonker, 5 m, 22 Oct 1988, leg. C. Vanden Berghen, no. 8599 (BR 00001569461).


Tilletia vittata is reported here for the first time from Senegal.

Although Tilletia vittata and T. oplismeni-cristati are similar in the way they convert the infected ovaries into more or less elongated, often curved bodies, covered by a hispid pericarp, they can easily be differentiated by their spore wall ornamentation.


Sori in all ovaries of infected plant, inconspicuous, concealed by glumes, fusiform, 0.6–0.9 × 0.15–0.3 mm, with a short, acute tip, bearing rudimentary style and stigmas, covered by a thin, greenish to yellowish brown pericarp that later ruptures exposing a powdery, dark reddish brown mass of spores and sterile cells. Sterile cells irregular, subglobose or broadly ellipsoidal, smaller than spores, (8–)10–15(–16) × (7–)9–14(–15) (13.2 ± 1.6 × 12.1 ± 1.5) μm (n = 50), hyaline; cell wall 1.2–3.0(–3.3) μm thick. In SEM, wall rugulose to wrinkled. Spores subglobose, broadly ellipsoidal or globose, (23.5–)24.5–29(–30) × (22–)23–26(–27.5) (26.4 ± 1.2 × 24.4 ± 1.1) μm (n = 100), medium reddish brown; spore wall (3.7–)4–5(–5.3) μm thick (including ornamentation, composed of 1.7–3.5(–3.8) μm high subconical ornaments); ornaments 17–24 on equatorial circumference, 5–7 per spore diameter, (0.6–)0.8–1.6(–2.0) μm high; hyaline sheath not seen. In SEM, ornaments usually not confluent, interconnected by thin ribs.
Figs 14–17. *Tilletia perotidis* on *Perotis patens* (P-P01929966). 14. An infected inflorescence (left) and a healthy inflorescence (right). 15, 16. Spores and sterile cells in LM (in median and surface view, respectively). In Fig. 15, arrows indicate sterile cells, arrowheads show immature spores. 17. Spores and sterile cells in SEM. Scale bars: 14 = 0.5 cm, 15, 16 = 10 μm, 17 = 5 μm
Figs 18, 19. *Tilletia perotidis* on *Perotis patens* (P-P01929966) – spores and sterile cells in SEM. Scale bars = 5 μm

**Specimen examined** – On *Perotis patens* Gand.: MADAGASCAR, MENABE REGION, Mahabo, Ferme de Mahabo, ca 20°23’S, 44°40’E, 1952–1955, leg. J. Dequaire, no. 27162 (P-P01929966, as ‘*Perotis latifolia* Aiton’, q.e. *P. indica* (L.) Kuntze).

**Distribution** – On Poaceae: *Perotis indica, P. patens*; South Asia (India), Africa (Malawi, Madagascar) (Thirumalachar & Pavgi 1952; Vánky 2011; Vánky et al. 2011).

It is worth noting that in the examined specimen of *T. perotidis* all ovaries are infected while in the protologue (Thirumalachar & Pavgi 1952: 392, and Fig. 9) and descriptions in Durán & Fischer (1961) and Vánky (2011), the infection is given as restricted only to some ovaries.

*Perotis* (Poaceae: Chloridoideae) is a small genus (16 species) in the tribe Cynodonteae, subtribe Perotidinae (Soreng et al. 2017). *Perotis patens* is endemic to Africa, distributed in Tropical and South Africa and Madagascar (Klaassen & Craven 2003). On this host, *Tilletia perotidis* has been previously recorded only from Malawi (Vánky et al. 2011). This smut fungus is reported here for the first time from Madagascar.

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