

# The genus *Pericladium* (*Ustilaginales*). *Pericladiaceae* fam. nov.

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**Abstract.** An historical account is given of the smut fungus genus *Pericladium* and its species, parasitic on members of *Grewia* (*Malvaceae*). The genus is characterised and descriptions of its three recognised species, *P. grewiae*, *P. piperis* and *P. tiliacearum* are given, along with their synonyms, host plant range and geographic distribution. A key to the species of *Pericladium* is presented. Arguments are enumerated for the necessity of description of a new family, *Pericladiaceae*, to accommodate *Pericladium* within the *Ustilaginales*.

**Key words:** *Grewia*, *Malvaceae*, *Pericladiaceae*, *Pericladium*, *P. grewiae*, plant parasite, smut fungi, *Ustilaginales*

## Introduction, history

Passerini (1875) described a fungus on the twigs of *Grewia* cf. *mollis* (*Malvaceae*, earlier *Tiliaceae*), collected by O. Beccari in Abyssinia [Ethiopia; Eritrea]. The fungus was placed in a new genus as *Pericladium grewiae* Pass. Passerini thought that this genus belonged to the *Uredinales* (under *Aecidinei*). Hennings (1900) studied this smut and considered it to be a peculiar *Ustilago*, and reduced the genus *Pericladium* to a synonym of *Ustilago*. Mundkur (1944: 293) recognised and restored the genus *Pericladium* as a smut fungus. In contrary, Ciferri (1954) considered *Pericladium* “a stilbaceous fungus”, allied with *Munkia*, *Graphiola*, *Stylina* and *Endocalyx*.

*Pericladium grewiae* is a remarkable smut, partly because of the nearly 1700 known species of smut fungi (comp. Vánky 2012) there are very few which parasitize woody plants. The smuts on woody plants comprise *Geminago nonveilleri* (Zambett. & Foko) Vánky & R. Bauer on *Triplochiton scleroxylon* (*Sterculiaceae*) from Central Africa, two species of *Uleiella*, *U. chilensis* Dietel & Neger and *U. paradoxa* J. Schröt., both on *Araucaria* species (*Araucariaceae*) in South America, four species of *Georgefischeria* on members of *Convolvulaceae* in India, and five species of *Mundkurella* on *Araliaceae*, from various continents.

Thirumalachar (1950: 176) described a second species of *Pericladium*, *P. tiliacearum* Mundk. & Thirum. on *Grewia rotundifolia* Juss. from India, possessing smaller spores. The next species in *Pericladium* was *P. piperis* (Zundel) Mundkur (1944: 293, as “*piperii*”), based on *Xylosorium piperis* Zundel

(1939: 576, as “*piperii*”) on supposed *Piper* sp., South Africa. However, I am rather convinced that the host plant of this species is also a *Grewia*. A further species, *Pericladium flavesci* Prasad & Tyagi (1961: 498) was described from India on *Grewia flavescens* Juss. It differs very little from the polyphagous *P. grewiae*, and it was considered to be a synonym (Vánky 2006: 41).

Joshi (1957) studied the spore germination of *Pericladium grewiae* on various substrates, describing the germination very briefly and giving no illustration of it. Mordue (1988) studied in detail *Pericladium* and the published literature, concluding that “the smuts on *Grewia* are best regarded as a single species.”

## Materials and methods

Sorus structure and spore characteristics were studied using dried herbarium specimens. For microscopic studies of the soral characters, thin twigs with sori were rehydrated in gently heated water, and fixed in 2% glutaraldehyde in 0.1 M Na-cacodylate buffer at pH 7.2 for several days. After six transfers in 0.1 M Na-cacodylate buffer, the material was postfixed in 1% osmiumtetroxide 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, the material was dehydrated in acetone series, embedded in Spurr's plastic and sectioned with a diamond knife. Semi-thin sections were stained with new fuchsin and crystal violet, mounted in “Entellan” and studied in a light microscope.

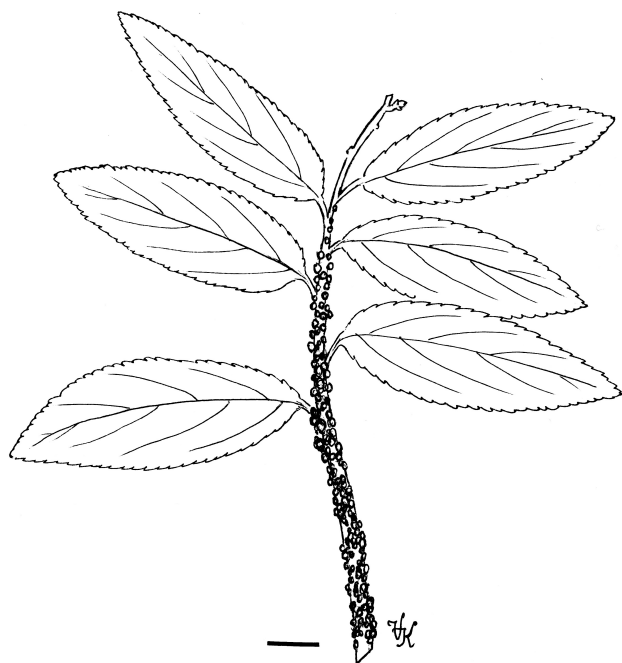


Fig. 1. Sori of *Pericladium grewiae* on a twig of *Grewia retusifolia*, forming aggregates of pustules (H.U.V. 18365). Habit. Bar = 1 cm

For light microscopy (LM) spores were suspended in a small droplet of lactophenol, covered with a cover glass, gently heated to boiling point to rehydrate the spores and eliminate air bubbles from the preparation, and studied at 1000 $\times$  magnification. For scanning electron microscopy (SEM), spores were placed on double-sided adhesive tape, mounted on a specimen stub, sputter-coated with gold-palladium, ca 20 nm, and examined in a SEM at 10 kV. Germination of freshly collected spores was obtained on MYP (malt-yeast-peptone-agar), at room temp., in 2–7 days.

## Results and discussion

The genus *Pericladium* and its species are characterised as follows:

*Pericladium* Pass., Nuovo Giorn. Bot. Ital. 7: 185, 1875.

*Xylosorium* Zundel, Mycologia 31: 576, 1939. — Type: *X. piperis* Zundel (as “*piperii*”, = *Pericladium piperis* (Zundel) Mundkur) on “*Piper* sp.” (= *Grewia* sp.; Vánky), South Africa (syn. by Mundkur 1944a: 293).

**Sori** as pustules or galls on twigs of plants in *Malvaceae* (earlier treated as *Tiliaceae*), formed by a hard, coriaceous peridium of hypertrophied host tissue and fungal elements, enclosing dark spore masses. **Spores** single, pigmented (brown), semi-agglutinated to powdery at maturity. **Sterile cells** absent. **Spore germination** results in either ovoid, multinucleate holobasidia giving rise to septate, ramified hyphae on which secondary sporidia develop, or by ramified, septate

hyphae that arise directly from the spores. In nutrient media, germinated spores produce brick-red yeast cultures. **Host-parasite interaction** by intracellular hyphae, coated by an electron-opaque matrix. Mature **septa** poreless.

Type of the genus: *P. grewiae*.

*Pericladium grewiae* Pass., Nuovo Giorn. Bot. Ital. 7: 185, 1875.

*Ustilago grewiae* (Pass.) Henn., Beibl. Hedwigia 39: (75), 1900. — Type on *Grewia* cf. *mollis* Juss., Abyssinia [= Ethiopia; Eritrea], near Sciotel, Zedamba, VI.1870, leg. O. Beccari.

*Pericladium flavesce* Prasad & Tyagi, J. Indian Bot. Soc. 40: 498, 1961. — Type on *Grewia flavesce*ns, HCIO 27004; isotypes IMI 80640, H.U.V. 15507! (syn. by Vánky 2006: 41).

**Sori** (Figs 1–3) on the twigs as globose, pustular bodies, c. 1–2 mm in diameter, gregarious to crowded, sometimes entirely surrounding the stems. Single sori formed by a brown, coriaceous peridium of an outer layer of host epidermis and hypertrophied host tissue mixed with fungal elements, enclosing a central mass of dark brown, semi-agglutinated spores. At maturity, the peridium ruptures irregularly revealing the semi-agglutinated to powdery spores. **Spores** (Figs 4–5) when mature solitary, roll-shaped, more or less flattened on one half, convex on the opposite side, in side view (5–) 5.5–7  $\mu$ m wide, in plane view circular to elliptic, 7–8.5  $\times$  7–10.5  $\mu$ m, chestnut-brown; wall c. 1  $\mu$ m thick, on the flattened side pale yellowish brown, thin, c. 0.2  $\mu$ m, smooth. **Spore germination** (Fig. 10) results in one-celled, ovoid holobasidia on a short pedicel, basidium 6–9  $\times$  10–15  $\mu$ m, with 8–12 nuclei. From the basidium septate, ramified hyphae arise on which secondary, ovoid sporidia develop measuring 3–5.5  $\times$  5.5–8  $\mu$ m. From the spores directly may arise septate, ramified, 2.5–3.5  $\mu$ m thick hyphae, similar to those resulting from germinated basidia; most probably the basidia remaining enclosed in the spores. In nutrient media, germinated spores produce brick red yeast cultures.

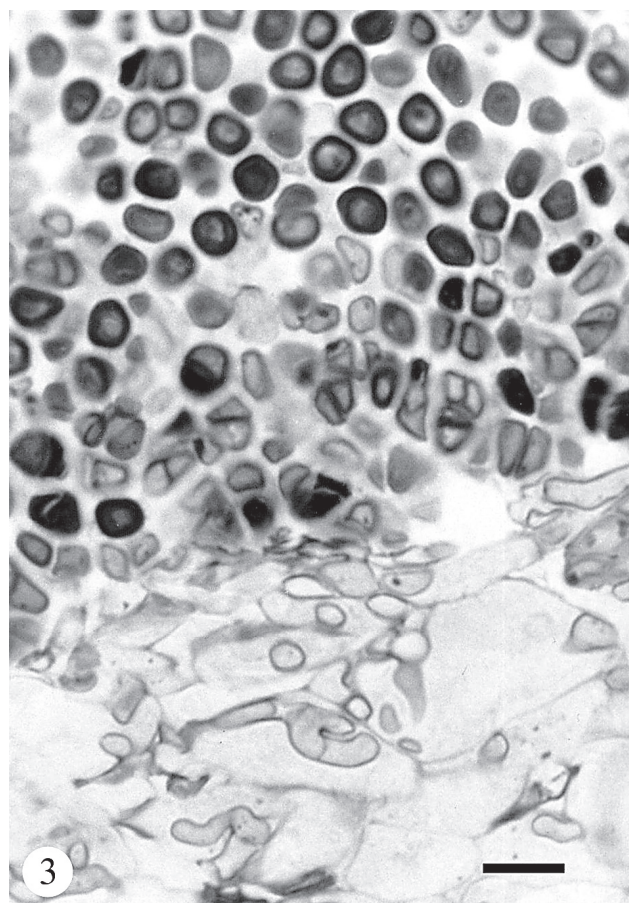
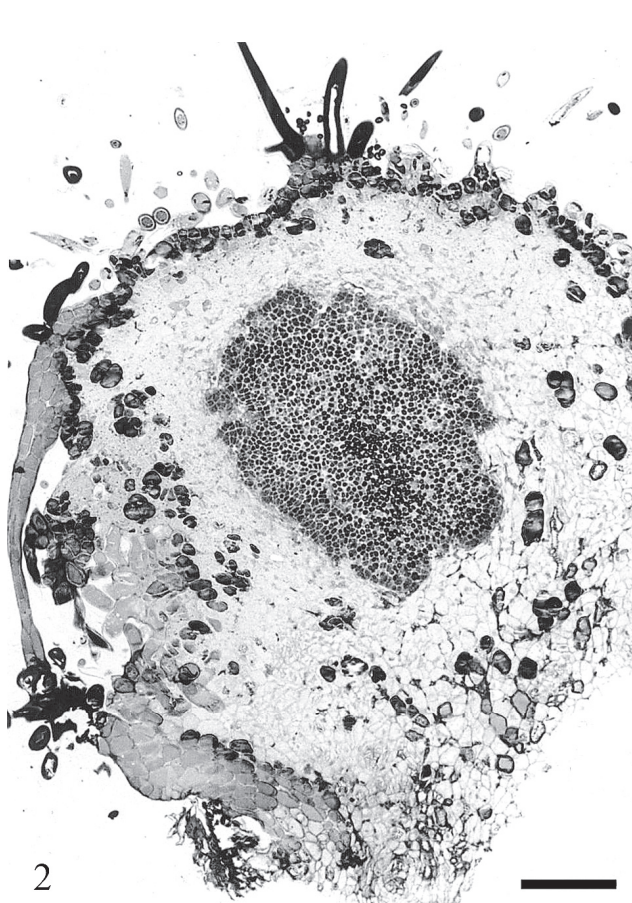
On *Malvaceae*: *Grewia asiatica* L., *G. breviflora* Benth., *G. carpinifolia* Juss., *G. columnaris* Sm., *G. flavesce*ns Juss., *G. microcarpa* K. Schum., *G. mollis* Juss., *G. orbiculata* Rottler, *G. retusifolia* Kurz, *G. venusta* Fresen, *G. villosa* Willd., *Grewia* sp.; Africa, S Asia, Australia.

*Pericladium piperis* (Zundel) Mundk., Mycologia 36: 293, 1944 (as “*piperii*”).

*Xylosorium piperis* Zundel, Mycologia 31: 576, 1939 (as “*piperii*”). — Type on “*Piper* sp.” (= *Grewia* sp., K. Vánky), South Africa, Transvaal, XI.1915, A. Rogers, PREM 11806; isotypes BPI 17940, H.U.V. 14716!

**Sori** on the twigs forming hard, coriaceous, globoid, agglomerated bodies of hypertrophied host tissues permeated by sporogenous hyphae, 1.5–2.5 mm in diam., or larger by coalescence of 2–3 bodies, filled with a dark brown, agglutinated to semi-powdery mass of spores. At maturity, the sori split irregularly and the spores are exposed. **Spores** (Figs





Figs 2–3. Semi-thin, stained transversal section of a young sorus of *Pericladium grewiae* on *Grewia villosa* (H.U.V. 8885). 2. TS of a sorus showing the central mass of dark spores and the thick, covering peridium of hypertrophied host tissue and fungal elements. Bar = 100  $\mu$ m. 3. Enlarged the area of spore formation. Bar = 10  $\mu$ m

6–7) variable in shape and size, subglobose, ovoid, ellipsoidal, elongate, usually more or less irregular, subpolyhedral, 6.5–11  $\times$  7.5–13.5 (–14)  $\mu$ m, yellowish brown, darker on one side; wall uneven, 1–1.5  $\mu$ m thick on the dark side, c. 0.3–0.4  $\mu$ m on the pale side (thicker after years in lactophenol), in LM smooth, in SEM very finely low verruculose.

On *Malvaceae*: *Grewia* sp.; S Africa. Known only from the type collection.

No leaves or flowers, only twigs of 3–5 mm diameter, surrounded by sori are present in the type collection of *Pericladium piperis*. Based especially on host specificity of the numerous collections of *Pericladium*, belonging to at least 13 *Grewia* species, and on spore morphology, I am rather convinced that the host plant of this species is not a *Piper* (*Piperaceae*) but a misidentified *Grewia* (*Malvaceae*). Parasitic microfungi are usually good “taxonomists” in identifying their host plants.

*Pericladium tiliacearum* Mundk. & Thirum., in Thirumalachar, Lloydia 13: 176, 1950.

Type on *Grewia rotundifolia*, India, Mysore, Channapatna, Hanumantarayanagudi, 23.XI.1943, K.B.A. Kahn, HCIO

10428; isotypes IMI 4946, H.U.V. 15508! Paratype on *Grewia tiliacefolia*, India, Mysore, Santaveri, 24.X.1943, M.J. Thirumalachar.

Sori as in *Pericladium grewiae* but the spores (Figs 8–9) are smaller, measuring 5–7  $\times$  5.5–8  $\mu$ m.

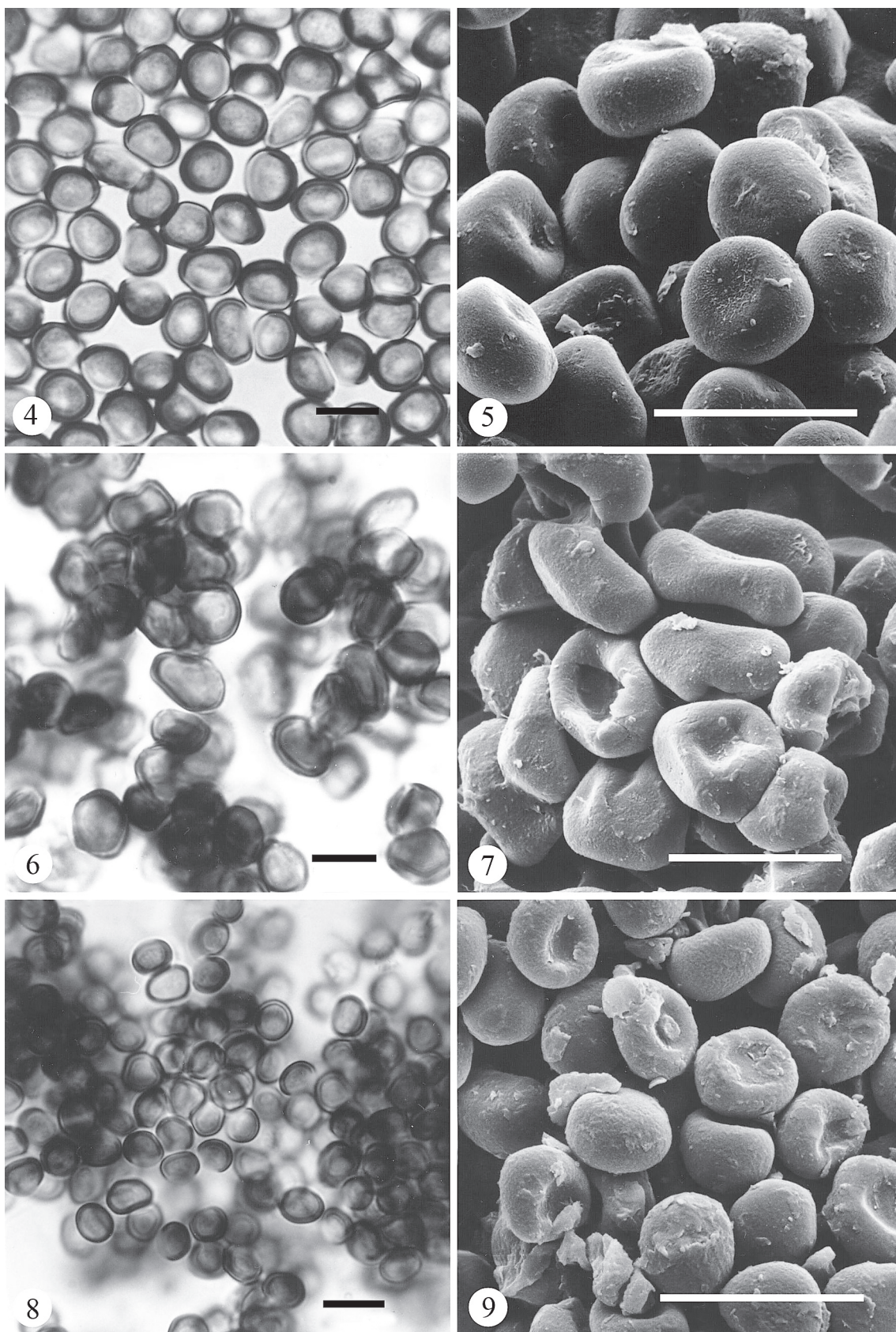
On *Malvaceae*: *Grewia rotundifolia* Juss., *G. tiliacefolia* Vahl, *G. villosa* Willd.; S Africa, S Asia.

#### Key to the species of *Pericladium*

- 1 Spores 5–7  $\times$  5.5–8  $\mu$ m ..... *P. tiliacearum*
- 1\* Spores larger ..... 2
- 2 Spores 7–8.5  $\times$  7–10.5  $\mu$ m ..... *P. grewiae*
- 2\* Spores 6.5–11  $\times$  7.5–13.5 (–14)  $\mu$ m ..... *P. piperis*

Ultrastructurally, the order *Ustilaginales* is characterised by poreless septa, the *Ustilaginaceae* family by poreless septa, intracellular hyphae and phragmobasidia. Based on these characters, the *Ustilaginaceae* represents a heterogeneous





**Figs 4–5.** Spores of *Pericladium grewiae* on *Grewia retusifolia* in LM and in SEM (H.U.V. 18365). Bars = 10  $\mu$ m. **Figs 6–7.** Spores of *Pericladium piperis* on “*Piper* sp.” (= *Grewia* sp.) in LM and in SEM (isotype). Bars = 10  $\mu$ m. **Figs 8–9.** Spores of *Pericladium tiliacearum* on *Grewia rotundifolia* in LM and in SEM (isotype). Bars = 10  $\mu$ m

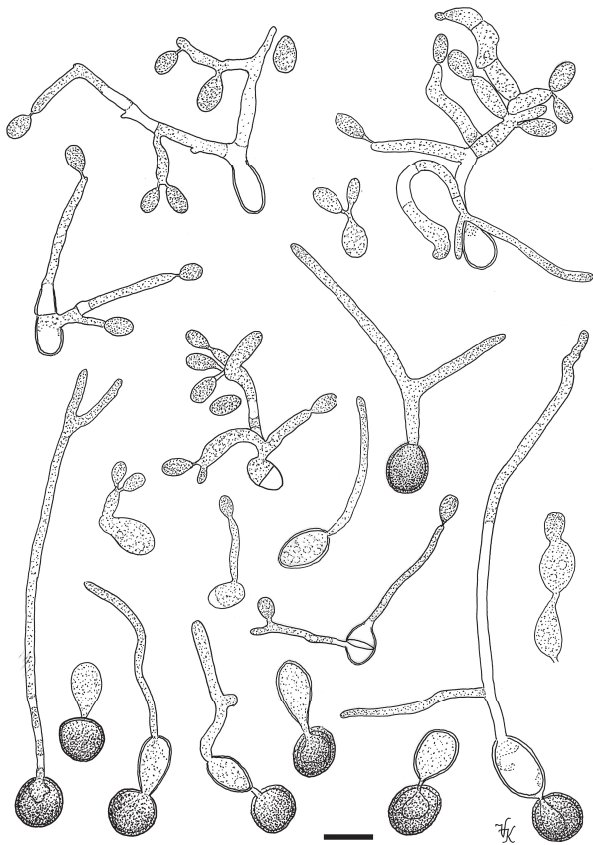


Fig. 10. Germinating spores of *Pericladium grewiae* on *Grewia retusifolia* (on MYP, at room temp., in 2–7 days, R. Bauer; in lactophenol with cotton blue) (H.U.V. 18334). Bar = 10  $\mu$ m

assemblage of 31 genera (comp. Weiss *et al.* 2004: 7–48), with most various morphology and parasitizing both mono- and dicotyledonous host plants of unrelated systematic position (gymnosperms, angiosperms). Vánky (2002: 18), in a more restricted sense, enumerated 14 genera under this family, and in a recent paper (Vánky 2012) 8 genera: seven on monocotyledonous host plants, e. g. *Franzpetrakia* (3 species), *Macalpinomyces* (45), *Moesziomyces* (1), *Parvulgo* (1), *Sporisorium* (330), *Tranzscheliella* (17), *Ustilago* (200), and one genus, *Melanopsichium* (2 spp.), on dicotyledonous host plants (*Polygonaceae*). This last one is explained as a result of a remote “host jump” from a *Poaceae* to *Polygonaceae* (comp. Weiss *et al.* 2004: 38).

Based on its ultrastructure and spore morphology only, *Pericladium* could be, and was considered a member of the *Ustilaginaceae* family. However, taking into consideration other criteria such as host plant taxonomy, sorus-, (teliospore-), basidial-, basidiospore-, cultural- and molecular phylogenetic characters, *Pericladium* should be excluded not only from the *Ustilaginaceae*, but also from all other described families of smut fungi.

Regarding host plant taxonomy, the dicotyledonous, woody *Grewia* (*Malvaceae*) is not related to the monocotyledonous, non-woody species of *Poaceae*, host plants of seven smut fungus genera in the *Ustilaginaceae*, enumerated above. A remote “host jump” of a smut from a member of one of these genera on *Poaceae* to a *Malvaceae* can with certainty be excluded (confirmed also by DNA analyses).

The hard, coriaceous, globose sorus structure of *Pericladium*, composed of a thick, outer layer of hypertrophied host tissue permeated by sporogenous hyphae, and a central mass of dark, agglutinated to semi-powdery mass of spores is unique for this genus.

Smooth, brown, roll-shaped spores with a more or less flattened, thin-walled half and a convex, thick-walled opposite side may occur also in other smut fungi, although not often.

Spore germination, resulting in a one-celled, short-pedicelled, ovoid, multinucleate holobasidium, from which septate, ramified hyphae arise producing secondary, ovoid sporidia is unique for *Pericladium*; such a germination could be called “*Pericladium*-type”. Rarely, from the spores directly, septate, ramified hyphae may arise, most probably the basidia remaining enclosed in the spores. The holobasidium excludes *Pericladium* from the *Ustilaginaceae*, which is characterised by phragmobasidia.

In nutrient media, the germinating spores produce brick red yeast cultures. Brick red yeast culture in true smut fungi is also unique.

Finally, molecular phylogenetic analysis (Begerow *et al.* 2007: 910, Fig. 1) shows an isolated position for *Pericladium* within the numerous genera of *Ustilaginomycetes*.

In conclusion, the following new family is proposed to accommodate *Pericladium*:

***Pericladaceae* Vánky, fam. nov.**

MYCOBANK # MB 563812

*Membrum ordinis Ustilaginales G.P. Clinton, emend. R. Bauer & Oberw., habens hyphae intracellulares, cum septis sine poris, et characteres proprios generis Pericladium: Sori sicut gallae plantarum dicotyledonearum (Malvacearum), massis sporarum atris, semi-agglutinatiss peridio crasso involutantes. Sporae singulae, pigmentatae (brunneae), mature pulverae. Cellulae steriles nullae. Germinatio sporarum holobasidiis ovoideis, multinucleatis, erigerontibus hyphis septatis, ramificatis sporidia secundaria evolutis, vel immediate hyphis sporarum septatis, ramificatis. In mediis nutritivis basidiosporae germinantes fermentum colore laterum producentes.*

***Typus familiae:*** *Pericladium* Pass.

Member of the order *Ustilaginales* having intracellular hyphae, poreless septa and characters of the genus *Pericladium*: **Sori** as galls on dicotyledonous plants (*Malvaceae*), composed of a peridium of hypertrophied host tissue and fungal elements, enclosing dark, semi-agglutinated spore masses. **Spores** single, pigmented (brown), powdery at maturity. **Sterile cells** absent. **Spore germination** results in ovoid, short-pedicelled, multinucleate holobasidia, giving rise to septate, ramified hyphae on which secondary sporidia develop, or



from the spores directly septate, ramified hyphae arise. In nutrient media, germinated spores produce brick red coloured yeast cultures.

**Type of the family:** *Pericladium* Pass.

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