The mycobiota of the cactus weed *Pereskia aculeata* in Brazil, with comments on the life-cycle of *Uromyces pereskiae*

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A survey of the fungi associated with the cactus weed *Pereskia aculeata*, conducted in Southeastern Brazil, revealed three phytopathogenic fungi. *Pseudocercospora pereskiae* sp. nov., a new cercosporoid fungus, found associated with leaf spots on *P. aculeta* is described and illustrated. This is the second species in *Pseudocercospora opuntiae* a fungus that attacks *Opuntia* sp. in Mexico. The other being *Pseudocercospora opuntiae* a fungus that attacks *Opuntia* sp. in Mexico. The other fungi collected on *P. aculeata* were the leaf spot fungus *Cercospora apii* and the rust *Uromyces pereskiae*. The polyphagous species *Cercospora apii* is reported for the first time on a member of the *Cactaceae*. Our observations indicated that *Uromyces pereskiae* and *Aecidium pereskiae* are two distinct heteroecious rust fungi, infecting two distinct *Pereskia* spp., *viz. P. aculeata* and *P. grandiflora*, respectively, and do not pertain to a single autoecious rust species as previously assumed. The potential use of these species as classical biological control agents of *P. aculeata* is discussed.

Key words: classical biological control, *Cactaceae*, fungal survey, new species, plant disease, taxonomy, *Cercospora*, *Pseudocercospora*, *Uromyces*.

Introduction

Pereskia aculeata Miller, known as Barbados gooseberry (local Brazilian names: lobrobrô, ora-pro-nobis) is a primitive woody member of the *Cactaceae*, characterized by having true leaves, short, curved thorns occurring in pairs close to the base of the leaves on young stems, white flowers and yellowish fruits resembling gooseberries. Leaves of *P. aculeata* are eaten cooked and marginally cultivated in many places in Brazil as vegetables. It is widely distributed and known to occur naturally in the Brazilian Atlantic Rain

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Forest (Lombardi and Gonçalves, 2000). This plant species has been introduced in South Africa since at least 1858. Its fruits are used for making jams and the plant is also utilized as hedge plant (Moran and Zimmermann, 1991). In 1979, P. aculeata was declared an invasive weed (Campbell, 1988). It is very thorny, climbs over indigenous and commercially planted forests, and can eventually kill the supporting plants (Moran and Zimmermann, 1991). Once it becomes established, it is virtually impossible to eradicate without damaging the surrounding vegetation (Byford-Jones, 1990). A South African biological control program has involved the release of the flea beetle Phenrica guérini Bechyné (Chrysomelidae, Alticinae), but the beetles have become abundant at only one locality in the Eastern Cape and their impact is unknown (Klein, 1999). Host-specificity tests on additional agents resulted in the rejection of two moth species (Klein, 1999). Since 2002, a systematic field survey of the mycobiota associated with *P. aculeata* was carried out in a part of its center of origin (Southern and Southeastern Brazil). The purpose of this survey was finding potential biocontrol agents against this weed. An account of the fungal species found attacking P. aculeata and observations about the diseases they cause is given.

Materials and methods

Prior to the field survey, a complete list of collecting localities from herbarium records of P. aculeata was compiled from the following Brazilian herbaria: Herb. IAC, Herbário Fanerogâmico e Criptogâmico do Instituto Agronômico; Herb. SP, Herbário Maria Eneyda P. K. Fidalgo; Herb. RB, Instituto de Pesquisas Jardim Botânico do Rio de Janeiro; Herb. R, Herbário do Museu Nacional do Rio de Janeiro; Herb. VIC, Universidade Federal de Viçosa; Herb. MBM, Museu Botânico Municipal; Herb. OUPR, Herbário José Badini. The southern and southeastern Brazilian states Minas Gerais, Rio de Janeiro, Espírito Santo, São Paulo, Paraná, Santa Catarina and Rio Grande do Sul were visited between January 2003 and December 2005. Selected sites were explored in each state. For details of the survey methodology and the laboratory studies see Barreto and Evans (1994). Freshly collected samples were examined under a stereomicroscope. Hand free sections containing the fungal structures were mounted in lactophenol. Observations, measurements and line drawings were prepared using an Olympus BX 50 light microscope fitted with a camera lucida. The collections examined were deposited in the herbarium of the Universidade Federal de Viçosa (VIC). Additional specimens previously deposited at VIC were also studied.

Results

Three fungal species were found associated with diseased *P. aculeata*: two cercosporoid leaf-spot fungi and a rust fungus. The fungi involved are described below.

Pseudocercospora pereskiae O.L. Pereira, R.W. Barreto & U. Braun, **sp. nov.** Mycobank 500719 (Figs 1-7)

Etymology: named in reference to the host genus.

Differt a *Pseudocercospora opuntiae* conidiis 2-2.5 µm latis, hyphis superficialibus cum conidiophoris solitariis.

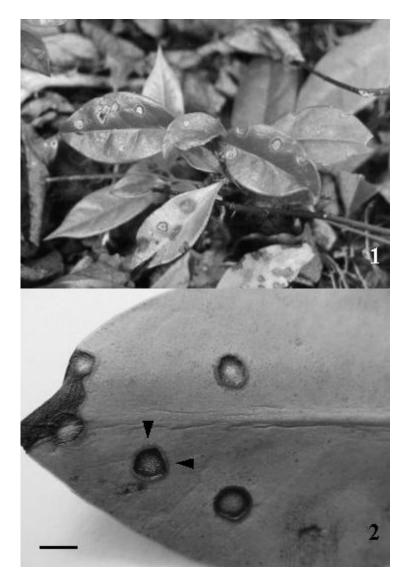
Leaf spots amphigenous, circular, depressed, 0.2-0.7 cm diam., whitish to gravish at center, with a purplish-black well defined border, surrounded by a chlorotic area. Colonies punctiform, dark brown to blackish, with a whitish mass of subhyaline conidia on abaxial leaf surfaces. Mycelium internal and external. Internal hyphae 2-5 µm diam., septate, branched, olivaceous-brown, thin-walled, sometimes in monilioid sequences, forming groups of anastomosing swollen hyphal cells, smooth. External hyphae present, 2-3.5 septate, pale-olivaceous, thin-walled, smooth, bearing μm, solitary conidiophores. Stromata well-developed, immersed to erumpent, composed of dark brown textura angularis, initially on young spots higher than wider, 22.5- $57.5 \times 25-90 \ \mu\text{m}$, with few conidiophores, becoming depressed in the middle during development with an increasing number of conidiophores and finally forming a cupulate stromata with many conidiophores in dense layers, $35-40 \times$ 165-175 µm. Conidiophores hypophyllous, erect, straight, subcylindrical, unbranched, commonly reduced to conidiogenous cells only, $6-25 \times 2-4 \mu m$, 0-2-septate, olivaceous, thin-walled, smooth. Conidiogenous cells integrated, terminal, pale olivaceous, proliferating sympodially but sometimes with percurrent proliferations, smooth. Conidiogenous loci inconspicuous, not darkened, unthickened. Conidia solitary, obclavate-cylindrical, straight to curved (sometimes nearly falcate), $16-82 \times 2-2.5 \mu m$, apex obtuse or subacute, base obconically truncate (sometimes with a truncate cell projection at the basis), 1-7-septate, subhyaline, thin-walled, smooth, hila unthickened and not darkened, commonly remaining attached to the conidiogeous cells.

Teleomorph: not seen.

Habitat: on living leaves of Pereskia aculeata.

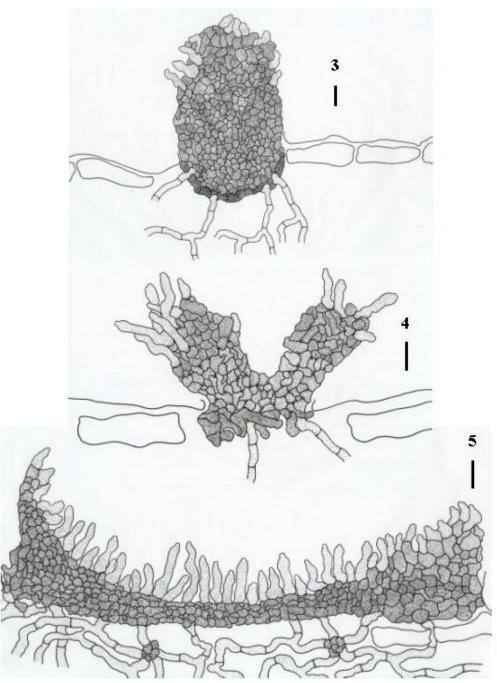
Known distribution: Minas Gerais (Brazil), Rio de Janeiro (Brazil), Bahia (Brazil). Paraná (Brazil).

Material examined: BRAZIL, Minas Gerais, Tiradentes, on living leaves of *Pereskia aculeata*, 15 May 2004, O.L. Pereira (VIC 29425; **holotype**). BRAZIL, Minas Gerais, on living leaves of *Pereskia aculeata*, 15 May 2004, O.L. Pereira (HAL 1899 F; **isotype**).

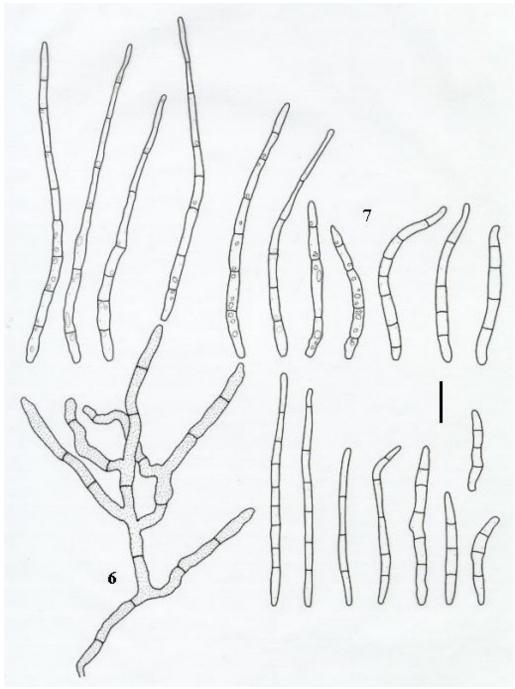


Figs 1-2. *Pseudocercospora pereskiae.* **1.** Symptoms on severely infected plants in the field. **2.** Spot detail showing many conidiophore layers with whitish mass of subhyaline conidia (arrowheads). Bar = 0.5 cm(2).

Other collections examined (all on Pereskia aculeata, Brazil): Bahia, 27 July 2000, R.W. Barreto (VIC 22154); Rio de Janeiro, 27 February 2001, R.W. Barreto (VIC 22156); Minas Gerais, 15 May 2004, O.L. Pereira (VIC 29426); Minas Gerais, 19 May 2004, O.L. Pereira (VIC 29424); Minas Gerais, UFV, 17 July 2004, O.L. Pereira (VIC 29423, HAL 1900F) and 10 August 2004, O.L. Pereira (VIC 29429).



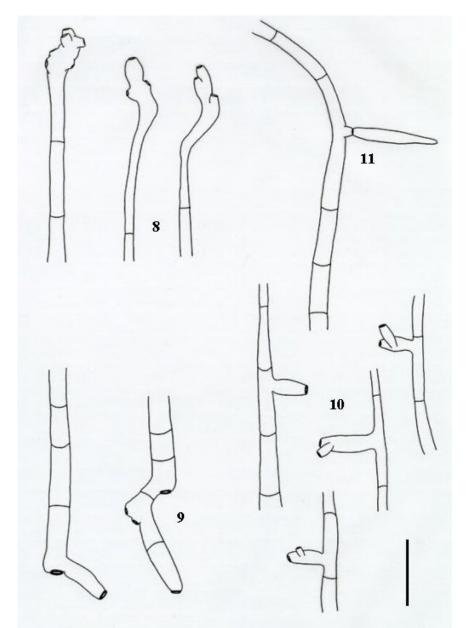
Figs 3-5. Stroma development of *Pseudocercospora pereskiae* (VIC 29425, HAL 1899F). **3.** Immature stroma on young spots with few conidiophores. **4.** Developing stroma with inner depression and more conidiophores. **5.** Mature cupulate stromata, on well developed spots, with many conidiophores in dense layer. Bars = $10 \mu m$.



Figs 6-7. *Pseudocercospora pereskiae* (VIC 29425, HAL 1899F). **6.** Superficial hyphae with conidiophores. **7.** Subhyaline conidia. Bars = $10 \mu m$.

Cercospora apii Fresenius (Figs 8-11)

Material examined: on *P. aculeata*, Brazil, Minas Gerais, 15 July 2004, O.L. Pereira (VIC 29420) and 17 July 2004 (VIC 29421).



Figs 8-11. *Cercospora apii* on *Pereskia aculeata* (VIC 29420, VIC 29421). **8.** Refractive dark scars on acicular conidial apex. **9.** Refractive dark scars on acicular conidial basis. **10.** Cicatrized conidiogenous cells along conidia. **11.** Secondary conidia formation on conidiogenous cells formed on well developed acicular conidia. Bars = $10 \mu m$.

Uromyces pereskiae Dietel

(Figs 12-15)

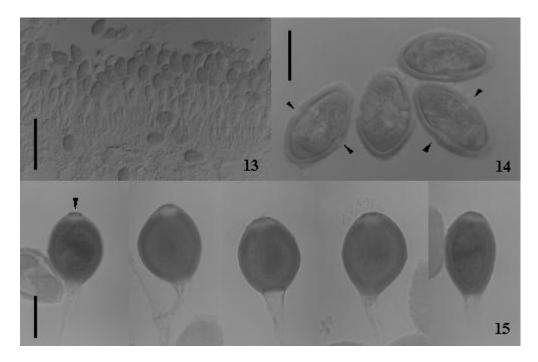
Lesions on living leaves, amphigenous, discolored, initially 1-3 mm diam, becoming confluent covering the whole leaf surface, leading to defoliation. *Spermogonia* and *aecia* unknown. *Uredinia* rarely abaxial, commonly adaxial, 250-600 μ m diam., 297-346.5 μ m high, erumpent, pulverulent, gregarious, reddish-brown. *Urediniospores* ellipsoid to obovate, commonly with a truncate rarely rounded base, pale yellowish, 34.5-37 × 21-25 μ m, wall 1.5-2.5 μ m thick, with 3-4 equatorial pores. *Telia* abaxial, erumpent, mixed with uredinia, dark brown. *Teliospores* 1-celled, smooth, reddish brown to chestnut-brown, 27-34.5 × 23-27 μ m, wall 2.5-4 μ m thick, with a prominent apical pore, pedicellate. *Pedicel* colorless, usually deciduous, 3.5-6 μ m wide.



Fig. 12. Rust symptoms caused by *Uromyces pereskiae* on a heavily infected leaf of *Pereskia aculeata* showing uredinia on adaxial leaf surface.

Material examined: on *P. aculeata*, Brazil, Minas Gerais, 16 July 1999, R.W. Barreto (VIC 22153) and 31 July 2001 (VIC 22152); 17 July 2004, O.L. Pereira (VIC 29422).

Other colections examined: Aecidium pereskiae Henn. on P. grandifolia, Brazil, São Paulo, 04 January 2001, R.W. Barreto (VIC 22155); 28 November 2005, O.L. Pereira, D.J. Soares & R.W. Barreto (VIC 29427).



Figs 13-15. Uromyces pereskiae on Pereskia aculeata (VIC 22152). **13.** Uredia with urediniospores. **14.** Finely echinulate uredospores with equatorial pores (arrowheads). **15.** Pedicellate 1-celled teliospore, with distinct apical pore (arrowhead). Bar = 100 μ m (13); 20 μ m (14, 15).

Discussion

Cercospora-like fungi (= cercosporoids) have been the subject of renewed interest to mycologists during the last decades, resulting in significant taxonomic changes and descriptions of numerous new taxa (e.g.: Crous and Braun, 2003; Kirschner *et al.*, 2004; Schubert and Braun, 2005; Braun *et al.*, 2006; Crous *et al.*, 2006a, b; Pereira and Barreto, 2006). Only one cercosporoid fungus was known to occur on members of the family *Cactaceae*, *viz.*, *Pseudocercospora opuntiae* Ayala-Escobar, U. Braun & Crous described on *Opuntia* sp. from Mexico (Ayala-Escobar *et al.*, 2006). *Pseudocercospora pereskiae* is similar to *P. opuntiae* in several morphological aspects but differs from it by the presence of a superficial hyphae bearing solitary conidiophores and its narrower conidia.

Crous and Braun (2003) listed numerous hosts belonging to many distinct plant families for *C. apii*, however, this new host addition represents the first

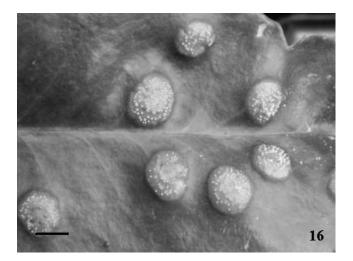
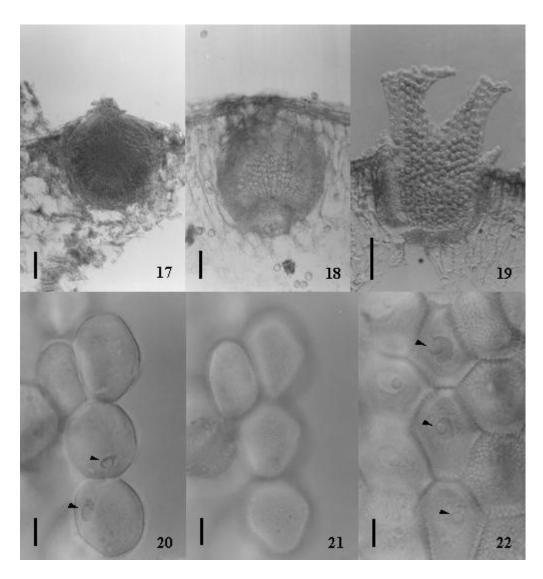


Fig. 16. *Aecidium pereskiae* attacking a leaf of *Pereskia grandifolia* (VIC 29427). Note raised areas with many aecia. Bar = 0.5 cm.

report of *C. apii* on a member of the *Cactaceae*. The material of *C. apii* on *P. aculeata* has an uncommon feature for a member of the genus *Cercospora* which is the presence of branched conidial chains. Nevertheless, although *Cercospora* has solitary conidia, short conidial chains can occasionally be produced in *C. apii* under moist conditions (Crous and Braun, 2003). Although *C. apii* can form some well-defined spots on leaves of senescing branches of *P. aculeata*, this fungal species is known to have an extremely wide host range (Crous and Braun, 2003). Therefore it cannot be considered as sufficiently damaging nor specific for use as a classical biological control agent.

Uromyces pereskiae and Aecidium pereskiae (reported as the anamorph of U. pereskiae, Figs 16-22) are the only known rust fungi reported on the genus Pereskia (Cactaceae). In publications about U. pereskiae, the host genus has been erroneously spelled as "Peireskea", "Peirescia" as well as "Peireskia" (Hennen et al., 2005). The type species of U. pereskiae was reported on Pereskia sp. from "St. Eduardo" (São Eduardo), RJ, Brazil, by Dietel (Dietel, 1899). Hennings (1898) described A. pereskiae from the same sample. The type collection of A. pereskiae is the same as that of U. pereskiae, but on a different leaf (Hennen et al., 2005). Later, the two synonyms Uromyces pereskiae H.S. Jackson & Holway and Aecidium pereskiae H.S. Jackson & Holway were proposed for specimens collected on Pereskia grandifolia Haw. from 'Nictheroy' (Niterói), RJ, Brazil (Jackson, 1931), but the family of the host was mistakenly reported as belonging to the Hippocrateaceae. During our survey, we collected U. pereskiae only on P. aculeata and A. pereskiae only on P. grandifolia.



Figs. 17-22. Aecidium pereskiae on Pereskia grandifolia (VIC 29427). **17.** Subepidermal spermogonia group V (type 4). **18.** Subepidermal aecial initial development. **19.** Cup-shaped subepidermal aecia with well-developed peridium. **20.** Catenate thin-walled aeciospores. Note the germ-pore (arrowheads). **21.** Finelly vertuculose surface of the catenate aeciospores. **22.** Internal surface of the peridial cells showing central oil drops (arrowheads). Bar = 50 µm (17); 100 µm (18, 19); 10 µm (20, 21, 22).

Both cactaceous species are commonly planted in Brazil for different purposes. Pereskia aculeata is very much appreciated as a green vegetable and P. grandifolia is planted for ornamental purposes, particularly in hedges. These species are somewhat similar being distinguished by the seasonal white flowers on P. aculeata and the purplish flowers on P. grandifolia. When not fertile, both species can be easily confused, but the presence of short curved thorns on the young stems occurring only on P. aculeata is a useful character for separating the two species. In Minas Gerais, for over four years, we have observed that P. aculeata showed rust symptoms seasonally, whereas P. grandifolia plants were always healthy and A. pereskiae was never observed in either species. In addition, fresh aeciospores of A. pereskiae collected on P. grandifolia from São Paulo were inoculated on P. aculeata but no disease resulted from such inoculations. Uromyces pereskiae and A. pereskiae seem to be two different heteroecious rust species and not two states of a single autoecious species as stated in Hennen et al. (2005). Jackson (1931) was of the opinion that: 'It is quite possible that this Uromyces is connected with the Aecidium described as Aecidium pereskiae. There is no evidence of such a connection except that they are both collected in the same region on the same host. It seems best, until more positive evidence is available of their identity, to describe the two forms as independent species'. Interestingly the type collection of A. pereskiae is the same as that of U. pereskiae, but material of each species came from a separate leaf (Hennen et al., 2005). We may conjecture that the original collections of U. pereskiae and A. pereschiae were in fact from different species of *Pereskia* which were mistakenly mixed by the collector.

Pseudocercospora pereskiae appears to cause a sufficiently severe disease on *P. aculeata* to justify its study as a potential biocontrol agent. The rust disease caused by *Uromyces pereskiae* is highly damaging to *P. aculeata* and rust fungi are often highly specific and, therefore, regarded as safe for classical introduction (Evans, 1987; Evans *et al.*, 2001). Nevertheless, a clarification of the life cycle of this species would be necessary before it is further considered for use as a biocontrol agent since another host plant may be involved.

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