RECENTLY DISCOVERED COLLECTIONS EXTEND THE GEOGRAPHICAL RANGE OF THE SMUT FUNGUS SPHACELOTHECA POLYGONI-SERRULATI TO CAMEROON AND ZAMBIA

MARCIN PIĄTEK, JOLANTA PIĄTEK & DOMINIQUE C. MOSSEBO

Abstract. The smut fungus *Sphacelotheca polygoni-serrulati* Maire is newly reported from Cameroon on *Persicaria decipiens* (R. Br.) K. L. Wilson and from Zambia on *P. pulchra* (Blume) Soják. *Persicaria pulchra* is a new host plant species for this smut fungus. The species is described and illustrated with line drawings of infected plants as well as LM and SEM micrographs. Information on the habitat of this species (based on the observation in Cameroon) is given for the first time, its host range is reviewed and the global distribution is discussed and mapped. *Sphacelotheca polygoni-serrulati* is currently known from two widely spaced areas of occurrence, African and Australasian, with no intermediate localities in the Asian tropics.

Key words: Africa, Australasia, Basidiomycota, Microbotryales, Persicaria, Pucciniomycotina, smut fungi, Sphacelotheca

Marcin Piqtek, Department of Mycology, W. Szafer Institute of Botany, Polish Academy of Sciences, Lubicz 46, PL-31-512 Kraków, Poland; e-mail: m.piatek@botany.pl

Jolanta Piqtek, Department of Phycology, W. Szafer Institute of Botany, Polish Academy of Sciences, Lubicz 46, PL-31-512 Kraków, Poland; e-mail: j.piatek@botany.pl

Dominique C. Mossebo, University of Yaoundé 1, Mycological Laboratory, B.P. 1456 Yaoundé, Cameroon; e-mail: dmossebo@ yahoo.fr

INTRODUCTION

The members of the fungal genus Sphacelotheca de Bary are parasitic on the host plants of the family Polygonaceae (genera Fagopyrum Mill. and Persicaria Mill.). The sori are produced in the ovaries; an individual sorus is composed of a peridium and a central columella, and filled with dark, violet-tinted spores that are connected by disjunctors in the young stage. When mature, the spores are separated but retain the disjunctors on both sides of the spore wall in the form of small pale violaceous appendages. Sterile cells intermixed with spores are absent (Vánky 2002), although some descriptions of Sphacelotheca hydropiperis (Schumach.) de Bary (Mordue 1991) or Sphacelotheca polygoni-serrulati Maire (Vánky & Oberwinkler 1994) contradict this. However, sterile cells in these descriptions are probably misidentified and in fact represent peridial cells. Although traditionally treated as smut fungi, the Sphacelotheca species are more closely related to

rust fungi than to true smuts, and together with genera such as Aurantiosporium M. Piepenbr., Vánky & Oberw., Bauerago Vánky, Fulvisporium Vánky, Liroa Cif., Microbotryozyma Suh, Maslov, Molestina & Zhou, Microbotryum Lév., Ustilentyloma Savile, and Zundeliomyces Vánky belong to the order Microbotryales within the subphylum Pucciniomycotina (Bauer et al. 2001, 2006; Weiss et al. 2004; Vánky 2008; Suh et al. 2012). Five species are currently recognized within Sphacelotheca (Vánky & Oberwinkler 1994), but identification of many specimens is difficult and it seems that the most easily identifiable species is S. polygoni-serrulati because of the characteristic reticulate ornamentation of the spore wall.

A population of *Sphacelotheca polygoni-serrulati* on its type host *Persicaria decipiens* (R. Br.) K. L. Wilson was found during a field survey of phytopathogenic fungi in western Cameroon

conducted in March 2007. Subsequently, an examination of unidentified materials of smuts from Kew Herbarium revealed a collection from Zambia on an unnamed Polygonum sp. which was determined as Persicaria pulchra (Blume) Soják during the present study. Both findings represent the first records of Sphacelotheca polygoni-serrulati in these respective countries (the Cameroonian record was included on a recent checklist of African smuts by Vánky et al. 2011) and Persicaria pulchra is a new host plant for this smut. In the present work, the newly discovered collections of Sphacelotheca polygoni-serrulati are described and illustrated in detail, the information on the habitat of this smut species (based on the observation in Cameroon) is given for the first time and its host range and global distribution are reviewed.

MATERIALS AND METHODS

Sori and spore characteristics were studied using dried herbarium material. The herbarium specimens are deposited in K and KRAM F. The specimens were examined by light microscopy (LM) and scanning electron microscopy (SEM).

For light microscopy (LM), small pieces of sori were mounted in lactic acid, heated to the boiling point and cooled, and then examined under a Nikon Eclipse 80i light microscope. LM micrographs were taken with a Nikon DS-Fi1 camera. Fifty spores and at least 10 peridial cells were measured from each collection, using NIS-Elements BR 3.0 imaging software, and the variation is presented as a range, with extreme values given in parentheses. Except for the spore walls and peridial cell walls, the measurements are adjusted to the nearest $0.5 \,\mu$ m. Mean and standard deviation values calculated from *n* spores are given after the spore size ranges. Only extreme values are given for peridial cells.

For scanning electron microscopy (SEM), spores and peridial cells were mounted on the carbon tabs and fixed to an aluminium stub with double-sided transparent tape. The stubs were sputter-coated with carbon using a Cressington sputter-coater and viewed under a Hitachi S-4700 scanning electron microscope, with a working distance of *ca* 12–13 mm. SEM micrographs were taken in the Laboratory of Field Emission Scanning Electron Microscopy and Microanalysis at the Institute of Geological Sciences of Jagiellonian University, Kraków (Poland).

RESULTS

Sphacelotheca polygoni-serrulati Maire Figs 1–3

Bull. Soc. Hist. Nat. Afrique N. 8: 74 (1917) – for full typification and synonymy see Vánky and Oberwinkler (1994), Vánky and McKenzie (2002) or Vánky and Shivas (2008).

Sori usually in all swollen ovaries of the inflorescences, rarely a few ovaries of the inflorescence may escape infection, forming ovate to cylindrical bodies, 2-6 mm long, 1.0-2.5 mm wide, covered by a dirty white or brownish peridium [on Persicaria decipiens: $2-5 \times 1.0-1.5$ mm, dirty white peridium; on P. pulchra: 2-6 × 2.0-2.5 mm, brownish peridium], composed of peridial cells; peridium during maturity ruptures irregularly from the apex, revealing dark purplish-black, semi-agglutinated to powdery mass of spores surrounding one central, stout, narrowing or not narrowing columella of the sorus length. Spores pale violaceous or violaceous, globose, subglobose, broadly ellipsoid or rarely ovoid, $9.0-14.0(-14.5) \times 9.0-12.5(-14.0) \ \mu m$, av. \pm SD, $11.9\pm1.1 \times 11.0\pm0.9 \ \mu m \ (n = 100)$ [on Persicaria decipiens: (10.5–)11.0–14.0(–14.5) × $(9.5-)10.0-12.5(-13.0) \ \mu m, av. \pm SD, 12.3\pm1.0 \ \times$ 11.2 \pm 0.9 µm (n = 50); on P. pulchra: 9.0–14.0 \times 9.0–12.5(–14.0) µm, av. \pm SD, 11.6 \pm 1.2 \times $10.8\pm1.0 \ \mu m \ (n = 50)$], with one spherical body (lipid body) in the cytoplasm; spores at first connected together by disjunctors, forming short chains, later separated and becoming single, but usually with two small, pale violaceous appendages on the opposite sides as the remnants of disjunctors; wall even or uneven, 0.8-2.5 µm thick [on Persicaria decipiens: 0.8-2.5 µm; on P. pulchra: 1.2-1.8(-2.5) µm thick], somewhat lighter at the appendages, surface finely, regularly to irregularly reticulate or sometimes somewhat labyrinthiform in LM and SEM, interspaces rugulose or tuberculate as seen by SEM. Peridial cells hyaline or subhyaline, globose, subglobose or broadly ellipsoidal, very variable in size, $5.5-22.0 \times 5-19 \ \mu m$ [on Persicaria. decipiens: $5.5-20.0 \times 5-17 \mu m$; on *P. pulchra*: $5.5-22.0 \times 5.5-19.0 \,\mu\text{m}$, with one spherical body (lipid body) in the cytoplasm; wall



Fig. 1. The macroscopic appearance of infection and sori of *Sphacelotheca polygoni-serrulati* Maire, respectively, on A & B – *Persicaria decipiens* (R. Br.) K. L. Wilson (KRAM F-48775) and on C & D – *Persicaria pulchra* (Blume) Soják [K(M) 154771]. Scale bars: A & C = 1 cm, B & D = 5 mm.

even, or rarely uneven, 0.8–3.3 μm [on *Persicaria decipiens*: 0.8–3.3 μm; on *P. pulchra*: 0.8–3.3 μm], surface smooth in LM and SEM.

SPECIMENS EXAMINED. CAMEROON. NORTH-WEST REGION: *ca* 10 km S of Bamenda, 05°52'13.3"N, 10°09'03.8"E, elev. 1805 m, marshy place, on *Persicaria decipiens* (R. Br.) K. L. Wilson, 8 March 2007, *leg. A. L. Njouonkou, J. Piątek, M. Piątek, C. Vánky & K. Vánky*, KRAM F-48775; ZAMBIA. No further details, on *Persicaria pulchra* (Blume) Soják, Oct. 1937–Feb. 1938, *leg. E. Milne-Redhead 3403*, K(M) 154771.

LOCATIONS, HABITAT AND POPULATION SIZE. The Cameroonian locality was found in the Bamenda Highlands, at an elevation of 1805 m, which belong to the Afromontane archipelago-like regional centre of endemism according to the classification of White (1983). A small population of the host plant inhabited a swampy place that was a very small remnant of probably natural vegetation surrounded by agricultural fields. Sphacelotheca polygoni-serrulati was rare in this place and we did not observe more than ten infected plants at the end of the dry season. Two other interesting, previously undescribed fungi were found at this place: the rust fungus Phakopsora dissotidis R. Berndt & M. Piątek on Dissotis thollonii Cogn. ex Büttner var. elliotii (Gilg.) Jacq.-Fél. (Berndt et al. 2008) and a still unnamed Pseudocercospora Speg. on the same species of Dissotis Benth. Details of the Zambian locality are unknown.

DISCUSSION

Sphacelotheca polygoni-serrulati was first described by Maire (1917) from two collections on Polygonum serrulatum Lag. (now Persicaria decipiens) from Algeria and Greece. Later, Vánky and Oberwinkler (1994) demonstrated that both collections represented two different species. By designating the lectotype, they attributed the name Sphacelotheca polygoni-serrulati to the material from Algeria, while they assigned the Greek specimen to the new species Sphacelotheca serrulati-magna Vánky & Oberw. Smuts with similar phenotypic characters were later described by other authors under the names Sphacelotheca doliaris Liro (Liro 1924), *S. tropico-africana* Zundel (Zundel 1944), and *S. polygoni-persicariae* G. Deml & Oberw. (Deml *et al.* 1985). Revising the smut fungi on Polygonaceae, Vánky and Oberwinkler (1994) found that they represented the same morpho-species and synonymized them with the oldest available name, *Sphacelotheca polygoniserrulati*.

The specimens on Persicaria decipiens from Cameroon and P. pulchra from Zambia were similar to each other in most morphological characters and also agreed quite well with a revised, short description of Sphacelotheca polygoni-serrulati offered by Vánky and Oberwinkler (1994). However, some minor morphological differences were observed between collections, including macro- and microscopic features. Macroscopically, the sori were somewhat smaller on Persicaria decipiens, measuring $2-5 \times 1.0-1.5$ mm, and had a dirty white peridium, than those on *P. pulchra*, which measured $2-6 \times 2.0-2.5$ mm, and had a brownish peridium. The colour of the peridium, however, may depend on the varied age of the specimens. Microscopically, the sizes of spores and the spore wall in the specimen on Persicaria decipiens differed slightly from those on P. pulchra (see species description). These minor morphological differences are considered as variability of the same morpho-species. Nevertheless, the cryptic speciation, similar to that observed in the closely related genus Microbotryum (Lutz et al. 2005, 2008; Kemler et al. 2006, 2009; Le Gac et al. 2007; Refrégier et al. 2008; Denchev et al. 2009; Piątek et al. 2012), cannot be entirely excluded within this morpho-species, but may be proved only by analyses of appropriate sampling of fresh specimens using methods of molecular phylogeny.

The host plant names of *Sphacelotheca polygoni-serrulati* included in various literature sources are *Polygonum barbatum* L., *P. persicaria* L., *P. salicifolium* Brouss. *ex* Willd., *P. serrulatum*, *P. setosulum* A. Rich., *Polygonum* sp. as well as *Persicaria decipiens* (Vánky & Oberwinkler 1994; Vánky & McKenzie 2002; Vánky & Shivas 2008; Vánky *et al.* 2011). These names in fact refer only to four species: *Persicaria barbata* (L.) H. Hara,



Fig. 2. Sphacelotheca polygoni-serrulati Maire on Persicaria decipiens (R. Br.) K. L. Wilson (KRAM F-48775). A–D – Spores seen by LM, median and superficial views; note the disjunctors indicated by arrows. G – Peridial cells seen by LM. – Sphace-lotheca polygoni-serrulati Maire on Persicaria pulchra (Blume) Soják [K(M) 154771]. E & F – Spores (intermixed with peridial cells) seen by LM, median and superficial views; note the disjunctors indicated by arrows. H – Peridial cells (intermixed with some spores) seen by LM. Scale bars = 10 μ m.

P. decipiens (= Polygonum salicifolium, P. serrulatum), Persicaria maculosa Gray (= Polygonum persicaria) and Persicaria setosula (A. Rich.) K. L. Wilson (= *Polygonum setosulum*). *Persicaria pulchra* is added here as a fifth host plant for *Sphacelotheca polygoni-serrulati*. It is worth

noting that McKenzie and Vánky (2001), in the checklist of New Zealand's smut fungi, reported two additional hosts for this species: *Polygonum hydropiper* L. [now *Persicaria hydropiper* (L.) Spach] and *Polygonum punctatum* Elliott [now *Persicaria punctata* (Elliott) Small], but they did not include them in the monograph of smut fungi of New Zealand (Vánky & McKenzie 2002).



Fig. 3. Sphacelotheca polygoni-serrulati Maire on Persicaria decipiens (R. Br.) K. L. Wilson (KRAM F-48775). A – Spores seen by SEM. B – Short chain of three spores connected together by disjunctors seen by SEM. C – Ornamentation of spore seen by SEM; note rugulose interspaces and well visible disjunctor. – Sphacelotheca polygoni-serrulati Maire on Persicaria pulchra (Blume) Soják [K(M) 154771]. D – Short chain of four spores connected together by disjunctors seen by SEM; note tuberculate interspaces and well visible disjunctor. F – Spores and peridial cell seen by SEM; note tuberculate interspaces and well visible disjunctor. F – Spores and peridial cell seen by SEM; note tuberculate interspaces and well visible disjunctor. Scale bars: A & B = 10 μ m, C–F = 5 μ m.



Fig. 4. Global distribution of *Sphacelotheca polygoni-serrulati* Maire: \bullet – on *Persicaria barbata* (L.) H. Hara, \bullet – on *Persicaria decipiens* (R. Br.) K. L. Wilson, \bullet – on *Persicaria maculosa* Gray, \bullet – on *Persicaria pulchra* (Blume) Soják, \bullet – on unknown *Polygonum* species.

These two hosts were probably misidentified in the former publication.

Sphacelotheca polygoni-serrulati shows a very interesting pattern of geographical distribution (Fig. 4), having two main areas of occurrence: African (with one extra-limital locality in southern Europe) and Australasian, both separated by a considerable gap where no records of this smut have been known so far. The African records of *S. polygoni-serrulati* reported to date are sparse and include single localities in Algeria (Maire 1917, type locality), the Democratic Republic of the Congo (Zundel 1944, type locality for *S. tropico-africana*), Portugal – Madeira Island (Deml *et al.* 1985, type locality for *S. polygoni-persicariae*), Rwanda (Liro 1924, type locality for *S. doliaris*), and Uganda (Vánky *et al.* 2011).

The Rwandan location needs further comment. In the protologue of *Sphacelotheca doliaris*, the type locality is given as 'Afrika, Hohasi? 30.7.1907: J.M. (632)' (Liro 1924). This entry was later repeated by all other smut researchers, but this is certainly based on misreading the original label of the specimen. Some details of this collection (date, abbreviation J.M., which probably denotes

Johannes Mildbraed) indicate that the material was collected during 'Deutsche Zentral-Afrika-Expedition 1907–1908' under the command of Adolf Friedrichs, and with the participation of Johannes Mildbraed, who was responsible for the botanical part of the expedition. In July 1907 the team collected materials around Lac Mohasi (also spelt Lake Muhazi), a lake located in eastern Rwanda, which is evident from the localities recorded in the book describing botanical and mycological discoveries, including smut fungi, made during the expedition (Mildbraed 1914). Smuts from these materials were elaborated by Hans and Paul Sydow, who reported three species from Lac Mohasi (Mildbraed 1914), although they did not mention any specimen referable to the collection numbered 632, which later served as a holotype of Sphacelotheca doliaris. It was probably caused by difficulties in identifying this specimen. The letter M (in Mohasi) was apparently mistaken for the letter H (in Hohasi) on the hand-written label. Thus, it is evident that the type locality of Sphacelotheca doliaris is Lac Mohasi in eastern Rwanda.

The present discoveries of *Sphacelotheca* polygoni-serrulati in Cameroon and Zambia fill

the gap in its distribution in Africa and suggest that it may be more common on this continent. The African area of occurrence of this smut is extended to southern Europe, where only one locality is known in eastern Spain (Almaraz 2002) and it is still not certain whether this is an isolated locality or part of a larger population.

The Australasian localities are more abundant than African localities although they were discovered only at the beginning of the 21st century as a result of a taxonomic re-evaluation of several old collections. *Sphacelotheca polygoni-serrulati* is known there from several places in the North Island of New Zealand, mostly in its northern, subtropical part (Vánky & McKenzie 2002) and from several places in southern Australia in the states of New South Wales, South Australia, Tasmania, and Western Australia (Vánky & Shivas 2008).

It is striking that besides African and Australasian areas of occurrence no intermediate localities are known in Asian tropics despite the occurrence of host plants. For example, *Persicaria decipiens*, a host plant of *S. polygoni-serrulati* in Africa and Australasia, is infected by *Sphacelotheca hydropiperis* in Pakistan (Kakishima & Ono 1993). Future studies should reveal whether *Sphacelotheca polygoni-serrulati* is indeed absent or has not been found in Asia.

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