Erysiphe quercicola causing powdery mildew on *Cassia fistula* and *Tamarindus indica* in Brazil

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Abstract

Powdery mildew on flowers of *Cassia fistula* and folioles of *Tamarindus indica*, caused by *Erysiphe quercicola*, is reported for the first time in Brazil. The identity of the pathogen was confirmed by morphological characteristics and molecular sequence analyses of ITS and 28S rDNA regions. This is the first report of *E. quercicola* causing powdery mildew on *C. fistula* and *T. indica* in Brazil.

Keywords Forest pathology · Obligate pathogen · Phylogeny · Powdery mildew

Cassia fistula is an ornamental tree known in Brazil as “Cassia imperial”, and internationally as Indian laburnum. It is native to Southeast Asia and was grown throughout the world due its several uses, for example in medicine and as an ornamental (CABI 2019). *Tamarindus indica* is a fruit tree, with Brazilian name Tamarindo, native to Africa, grown in the tropical regions of the world. Its fruits are used in many food applications (Bhadoriya et al. 2011).

During the 2018–2019 summer, plants of *C. fistula* and *T. indica* with typical symptoms of powdery mildew were observed in Minas Gerais state, in the cities of Viçosa and Monte Carmelo, respectively. The fungus had a whitish powdery sporulation on petal surfaces of *C. fistula* and on folioles of *T. indica* (Fig. 1a and b), being more abundant on the adaxial surface.

For morphological examination, reproductive structures were detached, using a sterile needle, from powdery mildew colonies on fresh flowers and folioles from the plants, and were mounted in lactoglycerol on a glass slide. Conidia were detached from the colonies, placed on a glass slide and left in a moist chamber for 24–48 h to induce germination of conidia and appressoria formation. Observations, measurements

and micrographs of morphological characteristics were made using an Olympus BX53 light microscope equipped with a digital camera (Q-Color 5 Olympus). At least 30 measurements were taken for each fungal structure. Voucher specimens from the collected materials were deposited at the herbarium of the Universidade Federal de Viçosa (Herbarium VIC)—Acc. N° VIC 47256, VIC 47257 and VIC 47258.

The pathogen was morphologically similar on the two hosts, having superficial mycelium and hyphae with 5.5–8.5 µm in diameter. The conidiophores were upright, cylindrical and hyaline, measuring 16.5–55 × 5.5–8.5 µm (Fig. 1c and d). However, some conidiophores of the specimen on *T. indica* (VIC 47257) were up to 105 µm long (Fig. 1g and h). Foot-cells of conidiophores are upright and measured 16.5–26 × 5.5–8.5 µm, followed by a cell prior to the conidia. These were hyaline, ellipsoid-shaped, single and measured 20–38.5 × 11.5–21.5 µm (Fig. 1e, i and j). Conidial germ tubes were produced only by the specimens from *C. fistula* (VIC 47256 and VIC 47258), in the perihilar position, and the lobate appressoria were produced at the terminal part of the tubes (Fig. 1f). The surface of the conidia showed angular/rectangular wrinkling of the outer walls (Fig. 1f and j). No chasmothecia were observed. These morphological characters are typical of the asexual structures of *Erysiphe quercicola* as described by Braun and Cook (2012) and similar to the description of this pathogen on *C. fistula* and *T. indica* by Meeboon and Takamatsu (2020).

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Fig. 1 Powdery mildew caused by *Erysiphe quercicola* on *Cassia fistula* (**a**, **c–f**) and *Tamarindus indica* (**b**, **g–j**). Symptoms on flowers of *C. fistula* (**a**). Symptoms on folioles of *T. indica* (**b**). Conidiophores (**c**, **d**). Conidia (**e**). Germinating conidia and showing the the angu-

lar/rectangular pattern of the outer wall (**f**). Greater conidiophore (**g**). Detail of conidium formation on conidiophore (**h**). Conidia (**i**). Conidia showing the angular/rectangular pattern of the outer wall (**j**). Scale bar = 10 µm

To confirm morphological identification, gDNA was extracted from colonies (mycelium, conidia and conidiophores), using the Chelex 100 method (Walsh et al. 1991). The internal transcriber spacer (ITS) regions including 5.8 rDNA and 28S rDNA including D1/D2 regions were amplified, using primer pairs ITS1F (CTT GGT CAT TTA GAG GAA GTA A) (Gardes and Bruns 1993) and LR7 (TAC TAC CAC CAA GAT CT) (Vilgalys and Hester 1990). The PCR products were sequenced by Macrogen (Seoul, Korea) and the sequences obtained, from 900 to 1200 nucleotides, were deposited in the GenBank (MN165985, MN165986 and MN165987). Phylogenetic trees were obtained by the Bayesian inference (BI) and Maximum-likelihood (ML) methods, using 30 sequences representing the genus *Erysiphe* available in GenBank and according with the Group II phylogeny from Takamatsu et al. (2015). The BI trees were constructed using MrBayes 3.2.2 on CIPRES SCIENCE GATEWAY, with 10,000,000 generations, and the posterior probabilities (PP) were

determined from a majority-rule consensus tree generated from 7,500 remaining trees. The ML trees were constructed using the MEGA X software system and the tree robustness was evaluated with 1,000 bootstrap (BS) values.

An NCBI megaBLAST search of these sequences showed over 99% similarity with ITS and 28S sequences of *E. quercicola*. Phylogenetic trees based on concatenated sequences of ITS and 28S regions placed the specimens of this study in the same clade with sequences of *E. quercicola*, with 100% PP and 100% BS (Fig. 2).

Takamatsu et al. (2015) showed that *E. quercicola* comprises a phylogenetic clade that includes powdery mildews from a wide range of host plants. This species is well-known on *Quercus* spp. (Fagaceae) from Asia, Australia and Europe, where the sexual phase is present and well defined. In tropical and subtropical Africa and South America, only the asexual phase is widespread on several hosts (Braun and Cook 2012; Meeboon and Takamatsu 2020). Although other species are associated with powdery

Fig. 2 Phylogenetic trees obtained by Maximum-Likelihood method using sequences of ITS and 28S from a combined dataset. Bootstrap (over 70%) and posterior probability (over 90%) values are indicated on the nodes. Bold specimens were obtained in this study. The tree was rooted with *Erysiphe glycines* (MUMH 396). T= Type material. Branches that are crossed by diagonal lines are shortened by 50%

mildews on *C. fistula* and *T. indica* (Farr and Rossman 2020), *E. quercicola* has been recently found associated with this disease in these hosts in Thailand (Meeboon and Takamatsu 2020). In Brazil, this pathogen is associated with *Anacardium occidentale*, *Bixa orellana*, *Cinnamomum camphora*, *Clitoria fairchildiana*, *Delonix regia*, *Hevea brasiliensis*, *Rumex crispus* and *Quercus robur* (Dallagnol et al. 2012; Cardoso et al. 2017; Dorneles et al. 2018, 2019; Piveta et al. 2018; Takamatsu et al. 2018; Fonseca et al. 2019a, b; Farr and Rossman 2020). Therefore, our study provides important information about *E. quercicola* on *C. fistula* and *T. indica*, and is the first to report this pathogen to these forest species in Brazil.

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