New records of *Pseudocercospora oenotherae* and *Synchytrium fulgens* on the invasive coastal plant *Oenothera laciniata* in Taiwan

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Abstract

Oenothera laciniata is a naturalized plant occurring on sand coasts and rural places in Taiwan. *Pseudocercospora oenotherae* (*Dothideomycetes*) is first recorded for this host species and for Taiwan. *Synchytrium fulgens* (*Chytridiomycota*) is a new record for Taiwan. Descriptions and illustrations are provided for both fungal species. The time spans for spread and detection of introduction of both the host as well as the parasites are discussed.

Key words – cercosporoid hyphomycetes–introduced species –Onagraceae – sand dune ecology

Introduction

Oenothera laciniata Hill (*Onagraceae*, Fig. 1) being native to North America has been introduced in many other countries. In Taiwan, the species was first recorded in 1986 mainly from sand coasts in Northern Taiwan (Peng & Huang 1986) and spread to sandy rural places also in the inland (Chen 2008) and finally became classified as invasive and weed (Wu et al. 2004a, b). Casual collections of plant parasitic fungi in Taiwan yielded two species on this plant and are recorded here in detail.

Materials & Methods

Specimens of diseased *Oenothera laciniata* were collected on a foredune of the sand coast and at a rural place along a rice field closely behind the foredune of Zhunan Township, Miaoli County, Taiwan in the years 2013 and 2014. Specimens were morphologically investigated as described in Kirschner (2013). DNA methods were applied as in Yeh & Kirschner (2014).

Results

Two species of fungi were associated with galls or leaf spots particularly on the rosette leaves of the infected plant (Figs 2, 6) and identified to belong to *Chytridiomycota* and asexual Ascomycota, respectively.

Pseudocercospora oenotherae (Ellis & Everh.) Y.L. Guo & X.J. Liu

Figs 2–5

Leaf spots amphigenous, irregular, pale brown with diffuse yellow margin, 1 mm diameter to extending to the whole lobe of the leaf. Hyphae internal, pale brown, smooth, 2–5 µm wide. Stromata amphigenous, in the substomatal chambers, pale to medium brown, (22–)23–39(–45) µm wide and (30–)33–46(–50) µm deep (n=10), giving rise to amphigenous fascicles of 10–45 conidiophores penetrating through stomata. Conidiophores unbranched (or rarely branched at the base), erect or somewhat prostrate, cylindrical, straight or slightly undulate or sometimes geniculate at the apex, pale brown, smooth, 0–2-septate, (12–)13–22(–30) × 3–4(–5) µm (n=30), conidiogenous cells terminal, (7–)10–15(–17) × 3–4(–4.5) µm (n=30), with 1–2 apical, unthickened, undarkened, 1.5–2 µm wide conidiogenous loci. Conidia solitary, narrowly obclavate-cylindrical, straight or slightly curved, pale brown, smooth, 1–9-septate, (20–)39–82(–128) × 3–3.5 µm (n=30), basal hilum unthickened, undarkened, 1–1.5 µm wide.

Known hosts and distribution – on *Oenotheraalbicaulis*, *Oe. biennis*, *Oe. laciniata* (new host species), *Oe. lamarckiana*, *Oe. odorata*, *Oe. serrulata*, and *Oe. tanacetifolia* (Onagraceae), PR China (Guo et al. 1998, Zhai et al. 2011), Japan (Katsuki 1965), Korea (Shin & Kim 2001), Taiwan (new record), USA (Crous & Braun 2003).

Material examined –Taiwan, Miaoli County, Zhunan Township, north of Dragon-Phoenix Fishing Port, 3 September 2014, R. Kirschner 4084 (TNM) – Living culture BCRC FU30354. ITS sequence (555 bp) GenBank KP053621.

Notes –*Pseudocercospora oenotherae* is distinguished from *Ps. didymospora* (Ellis & Barthol.) U. Braun & Crous by the low number of conidial septa (0–3) in the latter species (Crous & Braun 2003), and from *Ps. oenotherae-speciosae* U. Braun & Crous by the conidiogenous loci being not typical of *Pseudocercospora*, but appearing as minute circles in the latter (Braun et al. 2003). Guo et al. (1998) and Zhai et al. (2011) reported external hyphae giving rise to solitary conidiophores for *Ps. oenotherae* in China. Katsuki (1965) and Shin & Kim (2001), however, did not find external hyphae in the collections from Japan and Korea. External hyphae were also not detected in the specimen from Taiwan. The ITS sequence of the specimen from Taiwan (GenBank KP053621) was 100% identical to those from Korea (GenBank GU269856: 484 bp; GU269755: 490 bp). A BLAST search with the whole length (555) of the sequence from the Taiwanese strain, however, retrieved as the closest matches sequences of two *Pseudocercospora* specimens from members of *Oleaceae*: *Syringa reticulata* (GenBank DQ184477: 554/554, 0 different position) and *Fraxinus rhynchophylla* (GenBank GQ852766: 553/554, 1 different position), whereas the sequences of *Ps. oenotherae* did not appear among the 100 closest matches routinely presented after a BLAST search. This example demonstrates the strong impact of sequence length on molecular species identification and challenges the credibility of species lists based on BLAST searches and the commonly applied 97% similarity threshold.

Synchytrium fulgens J. Schröt.

Figs 6–10

Leaf discoloration absent, spots only caused by crowded resting spores. Inconspicuous galls formed on both sides of the leaf by hypertrophy of infected host cells. Resting spores amber to dark reddish brown, 1–4 per plant cell, slightly irregularly subglobose to ellipsoid, subglobose ones 33–85 µm diameter, ellipsoid ones 35–90 × 25–80 µm, cell wall 2–7 µm thick, outer residues brown, up to 10 µm thick. Prosori inconspicuous, seen only in the form of depressed empty spaces below mature or emptied sori. Sori 75–175 µm wide and 100–150 µm deep, containing numerous polyhedral zoosporangia appearing yellow when fresh, 17–37 × 15–28 µm, becoming released by apical rupture of the sorus and host cell. Liberated zoospores not seen.

Known hosts and distribution – on Onagraceae: *Boisduvalia densiflora*, *B. glabella*, *Clarkia elegans*, *Gaura* sp., *Oenotheraabramsii* (= *Sphaerostigma pallidum*), *Oe. biennis*, *Oe. bistorta*, *Oe. laciniata*, *Oe. lamarckiana*, *Oe. muricata*, *Oe. odorata*, *Oe. rhombipetala*, *Oe. sinuata*, *Oe. tarquensis*, in Ecuador, Germany, Japan, Switzerland, USA (Karling 1964), and Taiwan (new record).

Material examined – Taiwan, Miaoli County, Zhunan Township, north of Dragon-Phoenix Fishing Port, 30 Mar 2013, R. Kirschner 3853 (TNM).

Notes –The taxonomy of *Synchytrium* species on members of *Oenothera* was complicated because Karling (1954) separated *S. brownii* from *S. fulgens* by the formation of a prosorus which had not been recorded for *S. fulgens*. Later (1956), Karling assumed that the prosorus of *S. fulgens* was overlooked by the previous authors so that his *S. brownii* was a synonym of *S. fulgens*. After study of authoritative specimens of Schröter and numerous other specimens, Karling (1958) concluded that the prosorus was in fact overlooked by previous authors and confirmed the synonymy. In our own study, the presence or absence of prosori was also difficult to verify, because only mature and over mature stages with collapsed prosori were found. The single other *Synchytrium* species also known from *Oe. laciniata* is *S. macrosporum* Karling which belongs to a group of species in which only resting spores are known (Karling 1964).

Fig.1– Flowering *Oenothera laciniata* at the sand coast of Taiwan.

Discussion

The time span between introduction and subsequent scientific record of naturalized organisms is difficult to estimate. Because plants are more conspicuous than plant parasitic microfungi, more data about introduced plants are available than about introduction of the associated fungi. Data are, however, gradually accumulating also for fungi so that ratios for the time spans of discovery of the introduced plant and the subsequent record of the associated fungus could be estimated, but such analyses hitherto remained mainly restricted to Europe and North America (Kreisel & Scholler

1994, Desprez-Loustau et al. 2007, Eschen et al. 2014). Because of their geographically isolated positions, areas such as Taiwan and New Zealand are particularly suitable for studying the occurrence of introduced species. For example, *Phomasela ginellicola* has recently been shown to be parasitic on *Selaginella kraussiana* in New Zealand, where the record of the associated fungus followed almost one hundred years later to that of the plant (McClymont et al. 2013). The majority of approximately 100 smut fungus species in New Zealand are introduced and restricted to introduced host plants, but have been recorded considerably later than the host plants (McKenzie & Vánky 2001). Since recording naturalized plants in Taiwan is well-established (Wu et al. 2004a, b), some hypotheses about the spread of plant-associated fungi are possible. In Taiwan, *Oe. laciniata* was first recorded in 1986, but since villagers remembered the occurrence of the plant at sand coasts for the previous 20 years, it might have escaped the attention of field botanists (Peng & Huang 1986). Again, after more than 20 years, we present the first records of two pathogenic fungi on this plant in Taiwan. Both fungi are considered specific on members of Onagraceae as host plants. All five species of *Oenothera* known for Taiwan were introduced and recorded in the 1980s, except for *Oenothera tetraptera* Cav. recorded in the 1960s (Wu et al. 2004a). This species occurs at middle elevations in contrast to the low coastal elevation of *Oe. Laciniata* (Peng & Huang 1986), so that overlap between the distribution areas of the two *Oenothera* species and transmission of pathogens from one species to the other are not likely. Given the time required from first introductions to naturalization and scientific record of 20 years as in the casual note for *Oe. laciniata*, the two fungal species could not have been introduced before the 1960s.

Similarly, *Plasmopara obducens*, a pathogenic fungus on *Impatiens walleriana* which had been introduced to Taiwan as an ornamental in the 1960s, was also recorded in Taiwan only recently (Kirschner 2013). In both cases, we could estimate the record of the fungus approx. 50 years later than the introduction of the host plant. The host plant and the parasitic fungus had been taxonomically known many decades before these records and, therefore, may provide more reliable data for such estimates of non-epidemic spread than in cases, where the pathogen is taxonomically poorly known or only recently scientifically described. The latter case is not rare in the notoriously underinvestigated fungi, e.g. in the record of the powdery mildew of *Cassia fistula* in Taiwan approx. 250 years after introduction of its host plant to Taiwan and almost simultaneous scientific description of the fungus from India where the host plant is native (Kirschner & Chen 2008).

A pilot study accompanying the increased imports of agricultural plant products to Taiwan after joining the World Trade Organization in 2002 lead to the discovery of 19 fungal species previously unknown for Taiwan (Tzean & Huang 2009). Introduction of fungal inocula to Taiwan occurs, therefore, probably by transportation of humans and goods rather than by wind or other natural dispersal. With respect to invasive plants, such as *Oe. laciniata*, the subsequent introduction of their highly specific pathogens might reduce the impact of the naturalized plants without negative impact on native plants (McKenzie & Vánky 2001, Ellison & Barreto 2004, Kirschner 2013, McClymont et al. 2013).

Figs 2–5—*Pseudocercospora oenotherae* on *Oenothera laciniata* (R. Kirschner 4084, TNM). 2, Leaf spots on host. 3, Intercellular hyphae and fascicle of conidiophores penetrating from stroma through stoma in adaxial part of transversal leaf section. 4, Part of fascicle with conidiophores arising from stroma. 5, Conidia. —Bars 3, 4 = 10 μ m, 5 = 20 μ m.

Figs 6–10 – *Synchytrium fulgens* (R. Kirschner 3853, TNM). 6, Resting spores on fresh leaf of *Oenothera laciniata*. 7, Resting spore in epidermis cell. 8, Sorus with zoosporangia. 9, Two resting spores in an epidermis cell and an opened sorus with zoosporangia stained with cotton blue. 10, Fresh zoosporangia not stained by cotton blue, but showing their natural yellow color. Bars = 50 μm .

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