



## First record of *Erysiphe magnifica* on the new host *Magnolia × alba* in Taiwan indicates high morphological plasticity of the anamorph under tropical conditions

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### Abstract

The identification of the first record of *Erysiphe magnifica* for Taiwan is based on anamorphic material on diseased leaves of *Magnolia × alba*. Internal Transcribed Spacer (ITS) sequence comparison shows 100% identity with other sequences from *E. magnifica* with a clear barcode gap to other species. The morphology, however, deviates considerably from that reported in the literature by up to over 100 µm long foot cells of the conidiophore, which in some cases is correlated with a shift of the basal septum at a distance above the supporting hypha. These deviations may be correlated with adaptability to tropical environment. The potential evolutionary and phytopathological implications of such morphological anamorph plasticity are discussed.

**Key words** – Fungal biogeography – Erysiphales – Fungi – Magnoliaceae – tropical ecology

### Introduction

*Magnolia × alba* Figlar was introduced from mainland China to Taiwan in the 17th century. This evergreen tree requires high temperature, light and moisture conditions for optimal growth (Hsueh & Yang 2008). Since it does not produce mature fruits and is known only from cultivation, it is believed to be a hybrid. Analyses including the phytochrome A gene, LEAFY, and the Gibberellic-Acid Insensitive gene (Nie et al. 2008) support the assumption that *M. champaca* (L.) Baill. ex Pierre is one of the parent species, whereas *M. montana* (Blume) Figlar as potential other parent species (Nooteboom 1985) needs confirmation. In tropical Asia, including Taiwan, *M. × alba* is an important ornamental whose flowers are sold on the market and an important source of income for local people.

*Erysiphe magnifica* (U. Braun) U. Braun & S. Takam. is the most widespread of the ca. five powdery mildews recorded from *Magnolia* hosts, with respect to geography and host species (Ondrušková et al. 2014). The fungus is known from several *Magnolia* species and lotus (*Nelumbo nucifera* Gaertn.) in Europe, North and South America, and within Asia from Iran, Korea, and Japan (Kirschner 2010, Cho et al. 2014, Ondrušková et al. 2014, Khodaparast et al. 2019). In spite of the cultivation of *M. × alba* for centuries, this species has not been recorded as host of a powdery

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mildew. Young leaves of *M. × alba* in Taipei City, Taiwan, showed powdery mildew symptoms. The aim of the study was to identify the associated powdery mildew fungus to species.

## Materials & Methods

A specimen was collected in the city of Taipei and investigated immediately in the fresh state. For light microscopical characterization, samples were removed with a transparent tape and placed in 10% KOH; sizes of hyphae, conidiophores, conidiophore foot cells, and conidia were measured at 1000× magnification and presented as extreme values or as mean value ± standard deviation of 30 measurements with extreme values in brackets. The specimen was dried on an electrical dryer and deposited at the National Museum of Natural Science, Taichung (TNM) and the Department of Plant Pathology and Microbiology, National Taiwan University Herbarium, Taipei, Taiwan (PPMH). For measuring the foot cells of a German specimen, a duplicate of a published specimen (Kirschner 2010) in the private collection of R. Kirschner was used: On leaves of *Nelumbo nucifera* Gaertn., Frankfurt am Main, Botanical Garden, Sep/Oct 2009, R. Kirschner 3360, deposited at the Herbarium Senckenbergianum (FR), Frankfurt am Main, Germany (FR).

DNA was isolated from freshly collected anamorph material as in Wei & Kirschner (2017). The internal transcribed spacer (ITS) region of the rDNA was amplified, sequenced, and edited as in Wei & Kirschner (2017). The ITS sequence was used for BLAST searches and deposited in GenBank (accession number MT510149). Alignments were based on sequences selected from BLAST search results and Takamatsu et al. (2015). Alignments and Maximum Likelihood analyses based on the Kimura-2 parameter model with gamma distribution and 1000 bootstrap replicates were done as in Yeh et al. (2019). Following Takamatsu et al. (2015), *E. heraclei* DC. was chosen as outgroup.

## Results

The ITS sequence from the Taiwanese specimen was 100% identical with all six sequences of *E. magnifica* in GenBank exceeding 540 bp, whereas other species had identities of 98% or lower and 14 or more different bp. In the Maximum Likelihood analysis, the clade comprising the sequences of *E. magnifica* showed 100% support (Fig. 1). Morphologically, the specimen differed considerably with respect to the length of the foot cells of the conidiophores (Table 1). The full description is given below. Because of the results of the molecular data analysis, the species was identified as *E. magnifica*.

*Erysiphe magnifica* (U. Braun) U. Braun & S. Takam., Schlechtendalia 4: 10 (2000)

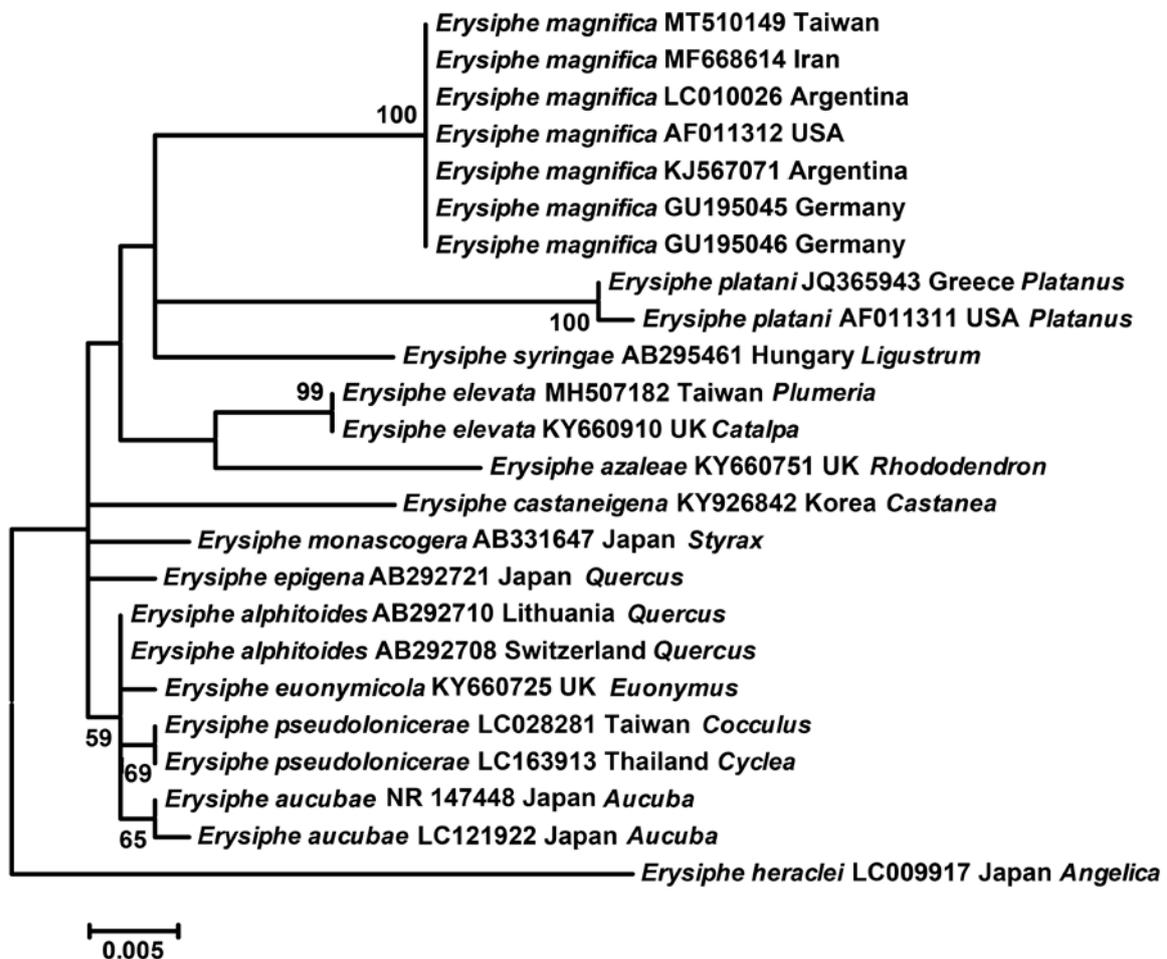
Figs 2, 3

Infection on *Magnolia × alba* limited to young leaves which become distorted and form large irregular purplish brown spots. Colonies amphigenous, effuse. Hyphae verruculose, 3–6 µm wide. Hyphal appressoria nipple-shaped to lobed, simple or in scattered opposite pairs. Conidiophores arising from the middle or margin of the hyphal mother cell, verruculose, composed of the foot cell and 0–2 apical cells, (55–)69–118(–225) × (7–)8–10(–11) µm (n = 30). Foot cell straight or in some cases curved at the base, basal septum at the same level as the upper surface of the hyphal mother cell, occasionally displaced apically up to 15 µm apart, (21–)37–109(–160) × (5–)5.5–7(–8) µm (n = 30). Conidia solitary, covered with fine longitudinal bacilliform scales, primary conidia ovoidal with truncate base and above middle narrowing to the rounded apex, ca. 21–26 × 13–14 µm (n = 2), secondary conidia doliiform with truncate ends, (24–)28–35(–40) × (13–)15–17.5(–20) µm (n = 30), germinating with short hypha and terminal strongly lobed appressorium.

Teleomorph – not found.

Known distribution – Europe, North and South America, Asia: Iran, Korea, and Japan (Pijpers 2009, Kirschner 2010, Braun & Cook 2012, Cho et al. 2014, Ondrušková et al. 2014, Khodaparast et al. 2019), Taiwan (**new record**).

Material examined – on young leaves of *Magnolia × alba* Figlar, Taiwan, Taipei City, Daan District, Chang Hsing Street, dormitory of National Taiwan University, 25.016408, 121.547846, 30 Apr 2020, R. Kirschner 4925 (PPMH, TNM) – ITS GenBank MT510149.



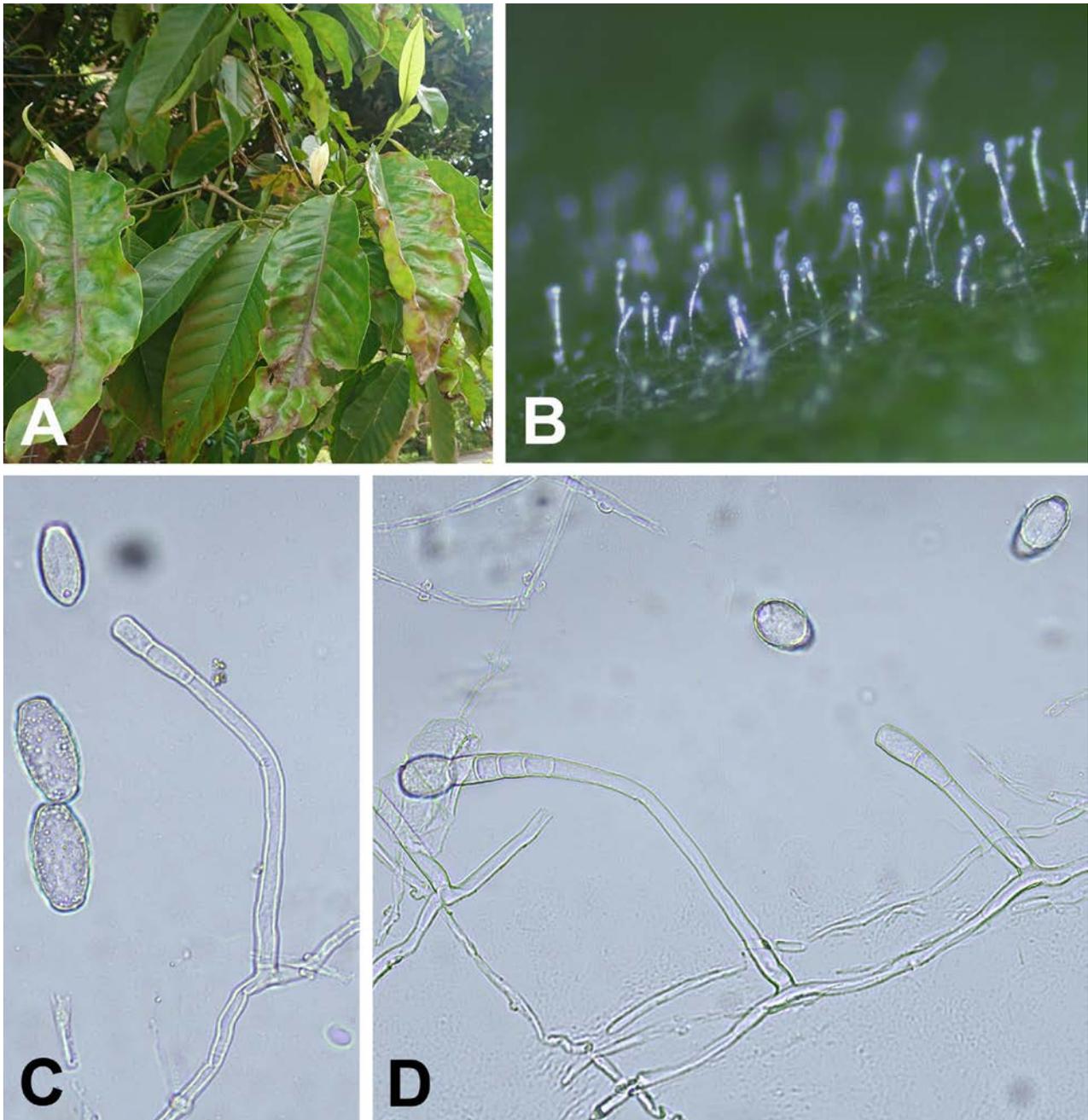
**Fig. 1** – Maximum Likelihood analysis of ITS sequences of *Erysiphe magnifica* on *Magnolia × alba* from Taiwan and other origins (on the top of the phylogenetic tree) as well as of related powdery mildew species with their GenBank numbers, location, and genus name of the respective host plant.

## Discussion

Except for a record from *M. ovata* (Shin 2000), all other *Magnolia* hosts of *E. magnifica* are deciduous species. *Erysiphe magnifica* is a new record for Taiwan and the evergreen *Magnolia × alba* is a new host. In spite of the presence of native *Magnolia* species in Taiwan, the fungus might have been introduced. The ITS sequence from the Taiwanese specimen was 100% identical with all six sequences of *E. magnifica* in GenBank exceeding 540 bp, whereas other species had identities of 98% or lower and 14 or more different bp. Among the powdery mildews on *Magnolia* hosts, no data exist for the anamorph and DNA of *E. bulbosa*. The specimens from Iran and Taiwan have both been identified by the host and ITS sequence identity, but differ by their anamorph morphology from the previous records of *E. magnifica*. Both non-typical specimens may, therefore, belong to *E. bulbosa* or another species whose ITS is not yet available but identical to those of *E. magnifica*. Although this probability cannot be totally excluded, we follow the other authors (Cho et al. 2014, Khodaparast et al. 2019), that this probability can be neglected under the condition of

100% ITS identity among specimens from different continents and clear barcode gap to other species.

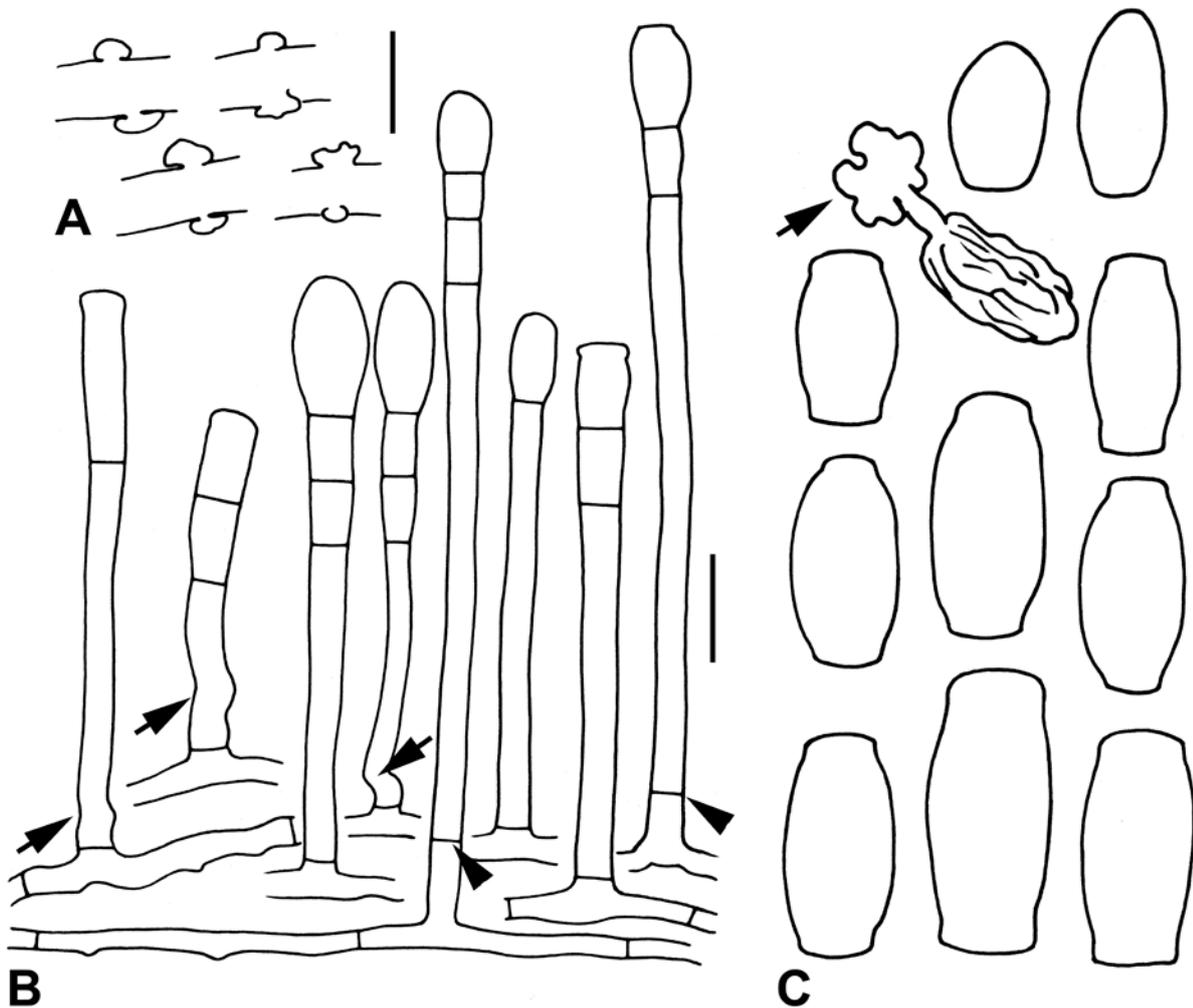
As shown by Takamatsu et al. (2015), other species being closely related, although with poor support, may be *E. syringae*, *E. platani*, *E. elevate*, and *E. azaleae* all also preferring woody host plants. A resolution of the interspecific relationships, however, was not the aim of this study.



**Fig. 2** – Photographs of *Erysiphe magnifica* from *Magnolia × alba* in Taipei, Taiwan. A Symptoms of young host leaves. B Conidiophores on leaf surface. C, D Conidiophores and conidia. For scales of microscopic structures see Fig. 3.

The length of conidiophore foot cells is often considered a diagnostically useful morphological feature in the taxonomy of powdery mildews (Braun & Cook 2012, Cho et al. 2014). Long foot cells were morphological key characteristics for segregating the newly proposed species *E. magnoliicola* S.E. Cho, Takam. & H.D. Shin from *E. magnifica* (Cho et al. 2014). For *E. magnifica*, however, Khodaparast et al. (2019) challenged this concept since they found

considerably longer foot cells of the conidiophores in their specimen from Iran equaling those of *E. magnoliicola*. In the Taiwanese specimen of *E. magnifica*, foot cells again could become as twice as those from the Iranian specimen (Table 1).



**Fig. 3** – Drawings showing microscopic characteristics of *Erysiphe magnifica* from *Magnolia × alba* in Taiwan. A Hyphal appressoria. B Conidiophores; bent foot cells indicated with arrow, elevated basal septa with arrowhead. C Conidia, one of them germinating with a lobed appressorium (arrow). Scale bars: A = 10  $\mu$ m, B, C = 20  $\mu$ m.

**Table 1** Sizes of conidiophore foot cells given for *E. magnifica* from different areas.

Origin	Foot cell	Reference
Iran	25–75 $\times$ 5–8.5 $\mu$ m	Khodaparast et al. (2019)
Germany	(15–)21–36(–40) $\times$ 5–8(–9) $\mu$ m (n = 15)	This study
Korea	15–45 $\times$ ca. 5–6 $\mu$ m	Cho et al. (2014), Shin (2000)
Taiwan	(21–)37–109(–160) $\times$ (5–)5.5–7(–8) $\mu$ m	This study

The two reports of particular long foot cell sizes of *E. magnifica* from subtropical Iran and Taiwan compared to those from rather temperate collection sites indicate morphological plasticity and correlation of anamorph morphology with climatic conditions. Other cases of morphological plasticity were shown for *E. necator* Schwein. and *Golovinomyces neosalviae* M. Scholler et al. which under greenhouse conditions form non-typical excessive long and/or branched conidiophores (Riaz et al. 2013, Götze et al. 2019). In some cases, the conidiophore terminated into a conidium

directly situated on the top of an extended foot cell (Riaz et al. 2013). Future studies with typically developed specimens may clarify which environmental factors, such as high temperature, high humidity, low air circulation, or a specific combination, may be responsible for inducing deviations from the morphology under temperate outdoor conditions and whether such deviation is reversible or not. Since the Taiwanese specimen was found outdoor, such environment-dependent deviations cannot be simply considered artifacts of greenhouse cultivation, but also occur spontaneously in nature. Perhaps such plasticity leading to more exposed conidia enables certain powdery mildews to spread under unfavorable conditions. In powdery mildews in Taiwan we did not find such morphological deviation under the same climatic conditions (Wang et al. 2019, Yeh et al. 2019). This phenomenon may be exceptional and limited to a few species, but such morphological plasticity may be genetically fixed and should be taken into consideration in the taxonomy in certain species of powdery mildews.

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