



Insight from morphology and phylogeny in species delimitation of *Diaporthe*

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Received: 13 January, 2018; Accepted: 15 February, 2018; Published online: 21 February, 2018

Introduction

Members of *Diaporthe* are known as plant pathogens, endophytes or saprobes on a wide range of host plants. *Diaporthe* species are well-known as the causal agents of many important plant diseases; including root and fruit rots, dieback, stem cankers, leaf spots, leaf and pod blights, and seed decay (Udayanga *et al.*, 2011). *Diaporthe helianthi* is the causal agent of one of the most important diseases of sunflower (*Helianthus annuus*) worldwide, and is also listed in the Chinese quarantine directory (Thompson *et al.*, 2011). For plant pathologists, studying of phytopathogenic *Diaporthe* spp. is therefore particularly important to work on wide range of plant diseases (e.g. grapes, sunflowers, soybean, and various diseases associated with ornamentals and forest trees). The taxonomy of *Diaporthe* spp. has recently been reviewed in several impactful studies (Udayanga *et al.*, 2011; Gomes *et al.*, 2013).

Before the molecular era, morphological characteristics such as size and shape of conidia

or ascospores, presence or absence of guttulate, color and growth rate of the colonies, and host associations, formed the basis for studying and comparing the taxonomy of *Diaporthe* spp. (Udayanga *et al.*, 2011). However, the utility of morphology has generally been shown to be of a little use for species identification as there is considerable plasticity of the characters within a species (Santos and Phillips, 2009). Host association has also been shown to bear little significance when referring to phylogenetic relationships (Santos and Phillips, 2009; Udayanga *et al.*, 2011). More than one species of *Diaporthe* can often be recovered from a single host, however, only one species can be associated with many different hosts (Santos and Phillips, 2009; Gomes *et al.*, 2013).

A multi-locus phylogenetic analysis has become a conventional procedure to identify novel fungal species, especially in those genera that lack distinctive morphological characters. The nuclear ribosomal internal transcribed spacer (ITS) has been chosen as the universal

barcode for the Kingdom Fungi, but confusions occur when it is used alone in phylogenetic studies (Schoch *et al.*, 2012). Gome *et al.*, (2013) suggested that ITS and HIS or TUB are recommended for the description of novel taxa in *Diaporthe*.

Udayanga *et al.*, (2015) reported that *Apn2*, *TEF1- α* and *HIS* genes showed the highest net phylogenetic informativeness, with *TEF1- α* showing the highest informativeness per site. Now, we selected *CAL*, *HIS*, *ITS*, *TEF1- α* and

TUB loci for the phylogenetic reconstruction as suggested from previous studies. However, some strains still could not be confidently identified to species level and several clades such as *D. amygdali* complex and *D. eres* complex remain unclear. Thus, it is necessary to be supplemented with additional suitable single-copy markers for satisfactory phylogenetic resolution in the future.

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