Diaporthalean fungi associated with canker and dieback of trees from Mount Dongling in Beijing, China

Haiyan Zhu¹, Meng Pan¹, Guido Bonthond², Chengming Tian¹, Xinlei Fan¹

¹ The Key Laboratory for Silviculture and Conservation of Ministry of Education, Beijing Forestry University, Beijing 100083, China ² GEOMAR Helmholtz Centre for Ocean Research Kiel, Düsternbrooker Weg 20, 24105, Kiel, Germany

Corresponding author: Xinlei Fan (xinleifan@bjfu.edu.cn)

Academic editor: Kevin Hyde | Received 8 July 2019 | Accepted 23 September 2019 | Published 16 October 2019

Citation: Zhu H, Pan M, Bonthond G, Tian C, Fan X (2019) Diaporthalean fungi associated with canker and dieback of trees from Mount Dongling in Beijing, China. MycoKeys 59: 67–94. https://doi.org/10.3897/mycokeys.59.38055

Abstract
Diaporthales is a fungal order comprising important plant pathogens, saprobes and endophytes on a wide range of woody hosts. It is often difficult to differentiate the pathogens in this order, since both the morphology and disease symptoms are similar among the various species. In the current study, we obtained 15 representative diaporthalean isolates from six tree hosts belonging to plant families Betulaceae, Fagaceae, Juglandaceae, Rosaceae, and Ulmaceae from Mount Dongling in China. Six species were identified residing in four families of Diaporthales (Diaporthaceae, Erythrogloeaceae, Juglanconidaceae and Melanconidaceae). Based on morphological comparison and the phylogenetic analyses of partial ITS, LSU, cal, has3, rpb2, tef1-a and tub2 gene sequences, we identified five known species (Diaporthe betulina, D. eres, D. rostrata, Juglamconis oblonga and Melanconis stilbostoma) and one novel species (Dendrostoma donglinensis). These results represent the first study of diaporthalean fungi associated with canker and dieback symptoms from Mount Dongling in Beijing, China.

Keywords
Ascomycota, Diaporthales, new species, phylogeny, taxonomy

Introduction
Diaporthales is an important order in class Sordariomycetes containing taxa that have broad host ranges and widely distributed as plant pathogens, endophytes or saprobes (Fan et al. 2018a, Crous et al. 2019). Most families of the Diaporthales are responsible for diseases on a wide range of host plants, some of which are economically important worldwide, causing anthracnose, blights, cankers, dieback, leaf spots and rots of root and
fruit (Alvarez et al. 2016, Guarnaccia and Crous 2017, Voglmayr et al. 2017, Jiang et al. 2019a, Xavier et al. 2019, Fan et al. 2020). The order is characterized by perithecia often with elongate beaks, immersed in stromatic tissues, producing deliquescent paraphyses and unitunicate asci that generally deliquesce, become detached from the perithecial wall when mature, and have a characteristic refractive apical annulus in sexual morph; and acervuli, pycnidia or rarely synnemata, producing phialidic or annellidic conidiogenous cells with 0–1-septate conidia in asexual morph (Barr 1978, Rossman et al. 2007, Fan et al. 2020). The classification of Diaporthales has been confused over the past decades because of the wide variation in morphological characters. Several recent studies have helped to resolve taxonomic problems of Diaporthales by multigene phylogenetic analyses and accepted 30 families in the order (Senanayake et al. 2017, 2018, Braun et al. 2018, Fan et al. 2018a, Crous et al. 2019, Guterres et al. 2019, Xavier et al. 2019).

Mount Dongling has a high diversity of plant species in western Beijing, which is considered as a biodiversity hotspot with more than 1000 plant species (Ma et al. 1995). As more plant species were recorded in this region, the exploration of fungal diversity gradually increased as most fungi are often linked to particular host plants as parasites or endophytes. Alternaria, Diaporthe, Leptostroma, Pestalotiopsis and Phoma were the most commonly isolated endophytic fungi from Pinus tabuliformis, and later additional 38 endophytic taxa were identified from Acer truncatum from the Mount Dongling (Guo et al. 2008, Sun et al. 2011). Further, pathogens of Botryosphaeriales have been identified from Mount Dongling, including species from the genera Aplosporella, Botryosphaeria and Phaeobotryon (Zhu et al. 2018).

During the trips to collect forest pathogens causing canker or dieback symptoms in Mount Dongling in Beijing, several specimens associated with typical diaporthalean symptoms were collected from various tree hosts, i.e., Betula dahurica (Betulaceae), Juglans regia, J. mandshurica (Juglandaceae), Prunus davidiana (Rosaceae) and Quercus mongolica (Fagaceae). As the higher-level phylogeny of many genera within the diaporthalean taxa remains largely unresolved in this region, the current study aims to clarify the systematics and taxonomy of these diaporthalean fungi with detailed descriptions.

### Materials and methods

#### Sampling and isolation

Fresh specimens of diaporthalean fungi were collected from infected branches of six hosts from Mount Dongling in Beijing, China (Table 1), during the course of cognitive practice at the Beijing Forestry University (BJFU). Diaporthalean canker symptoms include elongated, slightly sunken and discolored areas in the bark, which often splits along the canker margin, forming several prominent dark sporocarps immersed and erumpent through the surface of the bark (Fig. 1). A total of 15 isolates were obtained by removing the mucoid spore mass from conidiomata or ascomata of fresh material, which was cut horizontally with a sterile blade and mixed in a drop of sterile water on a
glass slide. The contents were broken up further with the blade until a spore suspension was obtained. The suspension was spread over the surface of 1.8 % potato dextrose agar (PDA). Single germinating spores were transferred on to fresh PDA plates. Specimens and isolates were deposited in the Key Laboratory for Silviculture and Conservation of the Ministry of Education in BJFU, and the working Collection of X.L. Fan (CF) housed at the BJFU. Axenic cultures are maintained in the China Forestry Culture Collection Centre (CFCC).

Morphology

Descriptions were performed based on morphological features of the ascomata or conidiomata from infected host materials. The macro-morphological photographs were captured using a Leica stereomicroscope (M205 FA) (structure and size of stromata, structure and size of ectostromatic disc and ostioles). Micro-morphological observations (shape and size of conidiophores, asci and conidia/ascospores) were determined under a Nikon Eclipse 80i microscope equipped with a Nikon digital sight DS-Ri2 high definition colour camera, using differential interference contrast (DIC) illumination and the Nikon software NIS-Elements D Package v. 3.00. Adobe Bridge CS v. 6 and Adobe Photoshop CS v. 5 were used for the manual editing. Over 10 conidiomata/ascomata, 10 asci and 30 conidia/ascospores were measured to calculate the mean size/length and respective standard deviations (SD). Colony diameters were measured and the colony features were described using the color charts of Rayner (1970). Nomenclatural novelties and descriptions were deposited in MycoBank (Crous et al. 2004).

DNA isolation, amplification and sequencing

Genomic DNA was extracted from colonies grown on cellophane-covered PDA using a modified CTAB method (Doyle and Doyle 1990). The primers and PCR conditions are listed in Table 2. DNA sequencing was performed using an ABI PRISM 3730XL
Table 1. Isolates and GenBank accession numbers obtained from Mount Dongling in the current study. (NA – not applicable).

| Species                  | Strain    | Host                        | GenBank accession numbers |
|--------------------------|-----------|-----------------------------|---------------------------|
|                          |           |                             | ITS: MW2656206            |
|                          |           |                             | LSU: MW265880             |
|                          |           |                             | Cal: NA                   |
|                          |           |                             | his3: NA                  |
|                          |           |                             | rpb2: MN315491            |
|                          |           |                             | tef1-α: MN315480          |
|                          |           |                             | tub2: NA                  |
| Dendrostoma donglinensis | CFCC 53148| Quercus mongolica           | 266206                    |
|                          | CFCC 53149| Quercus mongolica           | 266207                    |
|                          | CFCC 53150| Quercus mongolica           | 266208                    |
| Diaporthe betulina       | CFCC 53144| Betula davurica             | 266200 MN315462           |
| Diaporthe rostrata       | CFCC 53142| Juglans regia               | 266204 MN315467           |
|                          | CFCC 53143| Juglans mandshurica         | 266205 MN315464           |
|                          | CFCC 53145| Prunus davidiana            | 266202 MN315465           |
|                          | CFCC 53146| Prunus davidiana            | 266201 MN315466           |
|                          | CFCC 53147| Juglans regia               | 266203 MN315467           |
|                          | CFCC 53148| Quercus mongolica           | 266206 MN315463           |
|                          | CFCC 53149| Quercus mongolica           | 266207 MN315464           |
|                          | CFCC 53150| Quercus mongolica           | 266208 MN315465           |
|                          | CFCC 53151| Juglans mandshurica         | 266209 MN315466           |
|                          | CFCC 53152| Juglans mandshurica         | 266210 MN315467           |
|                          | CFCC 53153| Betula davurica             | 266211 MN315468           |
|                          | CFCC 53154| Betula davurica             | 266212 MN315469           |
|                          | CFCC 53155| Betula sp.                  | 266213 MN315470           |
|                          | CFCC 53156| Betula sp.                  | 266214 MN315471           |
|                          | CFCC 53157| Betula sp.                  | 266215 MN315472           |
|                          | CFCC 53158| Betula sp.                  | 266216 MN315473           |
|                          | CFCC 53159| Betula sp.                  | 266217 MN315474           |
|                          | CFCC 53160| Betula sp.                  | 266218 MN315475           |
|                          | CFCC 53161| Betula sp.                  | 266219 MN315476           |
|                          | CFCC 53162| Betula sp.                  | 266220 MN315477           |
|                          | CFCC 53163| Betula sp.                  | 266221 MN315478           |
|                          | CFCC 53164| Betula sp.                  | 266222 MN315479           |
|                          | CFCC 53165| Betula sp.                  | 266223 MN315480           |
|                          | CFCC 53166| Betula sp.                  | 266224 MN315481           |
|                          | CFCC 53167| Betula sp.                  | 266225 MN315482           |
|                          | CFCC 53168| Betula sp.                  | 266226 MN315483           |
|                          | CFCC 53169| Betula sp.                  | 266227 MN315484           |
|                          | CFCC 53170| Betula sp.                  | 266228 MN315485           |
|                          | CFCC 53171| Betula sp.                  | 266229 MN315486           |
|                          | CFCC 53172| Betula sp.                  | 266230 MN315487           |
|                          | CFCC 53173| Betula sp.                  | 266231 MN315488           |
|                          | CFCC 53174| Betula sp.                  | 266232 MN315489           |
|                          | CFCC 53175| Betula sp.                  | 266233 MN315490           |
|                          | CFCC 53176| Betula sp.                  | 266234 MN315491           |
|                          | CFCC 53177| Betula sp.                  | 266235 MN315492           |
|                          | CFCC 53178| Betula sp.                  | 266236 MN315493           |
|                          | CFCC 53179| Betula sp.                  | 266237 MN315494           |
|                          | CFCC 53180| Betula sp.                  | 266238 MN315495           |
|                          | CFCC 53181| Betula sp.                  | 266239 MN315496           |
|                          | CFCC 53182| Betula sp.                  | 266240 MN315497           |
|                          | CFCC 53183| Betula sp.                  | 266241 MN315498           |
|                          | CFCC 53184| Betula sp.                  | 266242 MN315499           |
|                          | CFCC 53185| Betula sp.                  | 266243 MN315500           |

Table 2. Genes used in this study with PCR primers, primer DNA sequence, optimal annealing temperature and corresponding references.

| Locus | Definition         | Primers | Primer DNA sequence (5’–3’) | Optimal annealing temp (°C) | References of primers used |
|-------|--------------------|---------|-----------------------------|-----------------------------|---------------------------|
| ITS   | internal transcribed spacer of ribosomal RNA | ITS1    | TCCGTAGGATGAACCTGCGG       | 51                          | White et al. 1990         |
|       |                    | ITS4    | TCACTCCGCTTTGATATGC        | 55                          | Vilgalys and Hester 1990  |
| LSU   | large subunit of ribosomal RNA | LR0R    | ACCCGGTGAACACCTAACG        | 55                          | Carbone and Kohn 1999     |
|       |                    | LR7     | TACTACACCAAGATCTG          | 55                          | Carbone and Kohn 1999     |
| cal   | Calmodulin         | CAL-228F| GATGTCAAGGAGGCCTTCTCC      | 55                          | Carbone and Kohn 1999     |
|       |                    | CAL-737R| CATCTTTCTGGCGCATG          | 55                          | Carbone and Kohn 1999     |
| rpb2  | RNA polymerase II second largest subunit | RP2B-5F | GA(T/C)GA(T/C)A/GA(T/C)GATAC(T/G)TT(T/C)GG | 52 | Liu et al. 1999 |
|       |                    | RP2B-7cR| CGCTCCAACCTGCTATGGCATCATG  | 55                          | Crous et al. 2004         |
|       |                    |         | ACCCAT(A/G)GTATGTTG       | 52                          | Glass and Donakson 1995   |
| his3  | histone H3         | H3-1b   | GCGGGGAGCGGCTGATGCTCT      | 55                          | Alves et al. 2008         |
|       |                    | H3-1f   | AGGTGCCACTGCTGTTGA         | 58                          | Glass and Donakson 1995   |
| tef1-α| translation elongation factor 1-alpha | EF1-668F| CGTCAACTGATCTCATGGA         | 55                          | Glass and Donakson 1995   |
|       |                    | EF1-1251R| CTCGAGAATCCCAACTGCTGTC    | 55                          | Glass and Donakson 1995   |
| tub2  | beta-tubulin       | B2a     | GCTAACCACATCGCTGCTG       | 55                          | Glass and Donakson 1995   |
|       |                    | B2b     | ACCCTACATGTAAGCTGGCC       | 55                          | Glass and Donakson 1995   |
DNA Analyser with a BigDye Terminator Kit v.3.1 (Invitrogen, USA) at the Shanghai Invitrogen Biological Technology Company Limited (Beijing, China). The DNA sequences obtained from forward and reverse primers were combined using SeqMan v. 7.1.0 in the DNASTAR Lasergene Core Suite software (DNASTAR Inc., Madison, WI, USA). Reference sequences were selected based on ex-type or ex-epitype sequences available from relevant recently published literature (Rossman et al. 2007, Suetrong et al. 2015, Norphanphoun et al. 2016, Hongsanan et al. 2017, Senanayake et al. 2017, Voglmayr et al. 2017, Yang et al. 2018, Fan et al. 2018a, b, 2020) (Table 1). Subsequent alignments for each gene were generated using MAFFT v.7 (Katoh and Standley 2013) and manually improved where necessary using MEGA v. 6 (Tamura et al. 2013). Novel sequences generated in the current study were deposited in GenBank (Table 1, Suppl. materials 1–3: Tables S1–S3) and the aligned matrices used for phylogenetic analyses were submitted to TreeBASE (www.treebase.org; accession number: S24893).

**Phylogenetic analyses**

To infer the first phylogenetic relationships at the family level, an initial alignment combining the here generated and available ITS, LSU, rpb2 and tef1-α sequences was compiled following Fan et al. (2018a). This alignment was analyzed based on Maximum Parsimony (MP), Maximum Likelihood (ML), and Bayesian Inference (BI) methods.

The MP analysis was conducted using a heuristic search (1,000 bootstrap) by PAUP v. 4.0b10 (Swofford 2003). The MP analysis was conducted with random sequence additions as option to stepwise-addition (1,000 bootstrap replicates and one tree held at each addition step), and maxtrees limited to 100 by replicate. The tree bisection and reconnection (TBR) was selected as option to the branch swapping algorithm (Swofford 2003). The branches of zero length were collapsed and all equally most parsimonious trees were saved. Other calculated parsimony scores were tree length (TL), consistency index (CI), retention index (RI) and rescaled consistency (RC). The ML analysis was performed using a GTR site substitution model, including a gamma-distributed rate heterogeneity and a proportion of invariant sites in PhyML v. 3.0 (Guindon et al. 2010). The BI analysis was conducted using the best-fit evolutionary models for each partitioned locus estimated in MrModeltest v. 2.3 (Posada and Crandall 1998) following the Akaike Information Criterion (AIC), with a Markov Chain Monte Carlo (MCMC) algorithm in MrBayes v. 3.1.2 (Ronquist and Huelsenbeck 2003). Two MCMC chains were run from random trees for 10 million generations and terminated when the average standard deviation of split frequencies dropped below 0.01. Trees were saved in each 1,000 generations. The first 25 % of trees were discarded at the burn-in phase of each analysis, and the Bayesian posterior probabilities (BPP) were calculated to assess the remaining trees (Rannala and Yang 1996). The MP bootstrap support (BS) equal to or above 50 were shown at the first and second position in branches. The branches with significant BPP equal to or above 0.95 were thickened in the phylogram.
In addition to the above analyses, we provided separate phylogenetic trees for two additional genera (Dendrostoma and Diaporthe) in Diaporthales, based on various gene regions (see below) including the same parameters as in the analyses described above. The branch support from MP and ML analyses was evaluated with a bootstrap support (BS) method of 1,000 replicates (Hillis and Bull 1993). Phylograms were plotted in Figtree v. 1.4.4 (http://tree.bio.ed.ac.uk/software/figtree) and edited in Adobe Illustrator CS6 v.16.0.0 (https://www.adobe.com/cn/products/illustrator.html).

Results

Phylogenetic analysis

The combined matrix (ITS, LSU, rpb2 and tef1-α) of Diaporthales included 198 ingroup accessions (15 from the current study and 183 retrieved from GenBank) and two outgroup taxa. The aligned matrix comprised 4,047 characters including gaps (773 characters for ITS, 1,190 for LSU, 1,114 for rpb2 and 970 for tef1-α), of which 2,002 characters were constant, 158 variable characters were parsimony-uninformative and 1,887 characters were variable and parsimony-informative. MP analyses generated 100 parsimonious trees of which the first tree is presented in Fig. 2 (TL = 12,631, CI = 0.313, RI = 0.792, RC = 0.248). The tree topologies of ML and BI analyses were mostly similar to the generated MP tree. The 15 isolates obtained in this study were clustered within the families Diaporthaceae, Erythrogloeaceae, Juglanconidaceae and Melanconidaceae in Diaporthales (Fig. 2). To delimitate to the species level, phylogenetic trees for Dendrostoma and Diaporthe were constructed separately based on different DNA datasets.

For the genus Diaporthe (Diaporthaceae), a concatenated ITS, cal, his3, tef1-α and tub2 matrix was produced with 201 ingroup accessions (6 from this study and 195 retrieved from GenBank). The combined matrix comprised 3,237 characters including gaps (544 characters for ITS, 593 for cal, 587 for his3, 645 for tef1-α and 868 for tub2) of which 1,330 characters were constant, 442 variable characters parsimony-uninformative and 1,465 characters variable and parsimony-informative. The MP analysis generated 100 parsimonious trees and the first tree is presented in Fig. 3 (TL = 12,978, CI = 0.280, RI = 0.712, RC = 0.199). The isolates of Diaporthe clustered in three different clades, corresponding to the three known species in this genus. The second combined matrix (cal, tef1-α and tub2) focusing on the Diaporthe eres complex included 56 ingroup accessions (4 from this study and 52 retrieved from GenBank). The concatenated matrix comprised 1,198 characters including gaps (405 for cal, 363 for tef1-α and 430 for tub2) of which 933 characters were constant, 112 variable characters parsimony-uninformative and 153 characters variable and parsimony-informative. The MP analysis generated 100 parsimonious trees of which the first is presented in Fig. 4 (TL = 415, CI = 0.701, RI = 0.882, RC = 0.618). The tree topologies of the ML and BI analyses were almost similar to the MP tree.
Diaporthalean fungi causing canker and dieback from Mount Dongling in China

Figure 2. Phylogram of Diaporthales based on combined ITS, LSU, rpb2 and tef1-α genes. The MP and ML bootstrap support values above 50% are shown at the first and second position, respectively. Thickened branches represent posterior probabilities above 0.95 from the BI. Ex-type strains are in bold. Strains from the current study are in blue.
For the genus *Dendrostoma* (Erythrogloeaceae), ITS, *rpb2* and *tef1*-α alignments were concatenated, including 42 ingroup accessions (three from this study and 39 retrieved from GenBank) was produced. The full matrix comprised 2,400 characters including gaps (561 characters for ITS, 1,078 for *rpb2* and 761 for *tef1*-α), of which 1,486 characters are constant, 231 variable characters are parsimony-uninformative and 683 characters are variable and parsimony-informative. The only parsimonious tree generated in MP analyses is presented in Fig. 5 (TL = 1,691, CI = 0.707, RI = 0.835, RC = 0.591). Tree topologies of ML and BI analyses were mostly similar to the MP tree. Three isolates of *Dendrostoma* represented a monophyletic clade with high support value (MP/ML/BI = 99/99/1) (marked in blue in Fig. 5).
Diaporthalean fungi causing canker and dieback from Mount Dongling in China

Taxonomy

Diaporthaceae Höhn. ex Wehm., Am. J. Bot. 13: 638 (1926)

Type genus. *Diaporthe* Nitschke, Pyrenomyc. Germ. 2: 240 (1870).

Notes. Diaporthaceae was introduced by von Höhnel (1917) and subsequently involved in confusing the taxonomy due to many genera with wide variation of morphological characters and the majority without culture or DNA phylogeny. Senanayake et al. (2017, 2018) accepted 14 genera in Diaporthaceae, including *Allantoporthe*,...
Figure 3. Phylogram of Diaporthe based on combined ITS, tef1-α, tub2, cal and his3 genes. The MP and ML bootstrap support values above 50% are shown at the first and second positions, respectively. Thickened branches represent posterior probabilities above 0.95 from the BI. Ex-type strains are in bold. Strains from the current study are in blue.
Diaporthalean fungi causing canker and dieback from Mount Dongling in China

Figure 3. Continued.
Figure 3. Continued.
Phylogram of *Diaporthe* complex based on combined *caL, tef1-α* and *tub2* genes. The MP and ML bootstrap support values above 50% are shown at the first and second positions, respectively. Thickened branches represent posterior probabilities above 0.95 from BI. Ex-type strains are in bold. Strains from the current study are in blue.

**Figure 4.**
**Figure 5.** Phylogram of *Dendrostoma* based on combined ITS, *rpb2* and *tef1-α* genes. The MP and ML bootstrap support values above 50% are shown at the first and second positions, respectively. Thickened branches represent posterior probabilities above 0.95 from the BI. Ex-type strains are in bold. Strains from the current study are in blue.
Diaporthalean fungi causing canker and dieback from Mount Dongling in China

Apioportella, Chaetoconis, Chiangraiomycetes, Diaportha, Hyaliappendispora, Leucodiaportha, Mazzantia, Ophiodiaportha, Paradiaportha, Phaeocytopora, Phaeodiaportha, Pustulomyces, and Stenocarpella.

Diaportha Nitschke, Pyrenomyc. Germ. 2: 240 (1870)

Type species. Diaportha eres Nitschke, Pyrenomyc. Germ. 2: 245 (1870).

Notes. The genus Diaportha (syn. Phomopsis) was established by Nitschke (1870). The identification of Diaportha was confused due to the historical species recognition criteria based on overlapped morphology, culture characteristics and host affiliation (Dissanayake et al. 2017). The phylogenetic analysis recommended to delimitate taxa to the species level was first proposed by Udayanga et al. (2012) and later modified to include concatenated alignments of ITS, cal1, his3, tef1-a, tub2 (Gomes et al. 2013). More than 1,050 epithets for Diaportha and 950 for Phomopsis are listed in Index Fungorum (August 2019). Dissanayake et al. (2017) provided most type/ex-type species details and phylogenetic frame with 172 species in this genus. Yang et al. (2018) summarized 15 species of Diaportha associated with dieback disease of tree hosts in China and introduced 12 new species.

Diaportha betulina C.M. Tian & Q. Yang, Mycokeys 39: 97 (2018)

Description. See Yang et al. (2018).

Material examined. CHINA, Beijing City, Mentougou District, Mount Dongling, Xiaolongmen Forestry Centre (39°59’23.58”N, 115°27’05.00”E), from branches of Betula dahurica Pall., 17 Aug. 2017, H.Y. Zhu & X.L. Fan, deposited by X.L. Fan, CF 2019831, living culture CFCC 53144.

Notes. Yang et al. (2018) described Diaportha betulina from cankers of Betula spp. in Heilongjiang Province. The only strain CFCC 53144 representing D. betulina clusters in a well-supported clade and appear most closely related to D. betulae, which was also isolated from Betula platyphylla in Sichuan Province (Du et al. 2016). Diaportha betulina (strain CFCC 52562) differs from D. betulae by its slender alpha conidia (2.5–3 vs. 3–4 μm) (Du et al. 2016), and 13 bp for ITS, 7 bp for cal, 19 bp for his, 12 bp for tef and 6 bp for tub2 based on alignment of the concatenated five-gene deposited in TreeBASE (S24893). Both morphology and sequence data confirmed that our isolates belong to this species.

Diaportha eres Nitschke, Pyrenomyc. Germ. 2: 245 (1870)

Fig. 6

Description. Sexual morph: not observed. Asexual morph: Pycnidial stromata immersed in bark, scattered, slightly erumpent through the bark surface, unilocular,
with a conspicuous central column. Central column beneath the disc more or less conical, pale grey with yellow. Ectostromatic disc orange, elliptical, 160–300 μm in diam., with one ostiole per disc. Ostiole dark brown to black, at the same level as or slightly above the disc surface, 70–80 μm in diam. Locule single, 210–260 μm in diam. Conidiophores cylindrical, hyaline, unbranched, straight or slightly curved, tapering towards the apex, 12–13.5 × 2–3 μm. Conidiogenous cells enteroblastic, phialidic. Alpha conidia hyaline, aseptate, smooth, ellipsoidal, biguttulate, rounded at both ends, 6.5–8.5 × 2.5–3 (av. = 7.3 ± 0.5 × 2.8 ± 0.3, n = 30) μm. Beta conidia were not observed.

**Culture characteristics.** Cultures on PDA are initially white, growing up to 4 cm in diam. after 3 days, and becoming yellow green to brown after 7–10 days. Colonies are flat felty with a thick texture at the marginal area, with a thin texture at the center, abundant aerial mycelium, sterile.
**Material examined.** CHINA, Beijing City, Mentougou District, Mount Dongling, Xiaolongmen Forestry Centre (39°58'06.45"N, 115°26'48.36"E), from branches of *Prunus davidiana* (Carr.) Franch., 20 Aug. 2017, H.Y. Zhu & X.L. Fan, deposited by X.L. Fan, CF 2019808, living culture CFCC 53146; *ibid.* CF 2019858, living culture CFCC 53145. CHINA, Beijing City, Mentougou District, Mount Dongling, Xiaolongmen Forestry Centre (39°57'47.49"N, 115°29'20.52"E), from branches of *Juglans regia* L., 20 Aug. 2017, H.Y. Zhu & X.L. Fan, deposited by X.L. Fan, CF 2019801, living culture CFCC 53147.

**Notes.** *Diaporthe eres* is the type species of *Diaporthe*, and is also the most common species causing canker disease on a wide range of hosts (Gomes et al. 2013, Udayanga et al. 2014, Dissanayake et al. 2017, Yang et al. 2018). Our isolates are associated with canker disease of *Prunus davidiana* in China, which belong to the *Diaporthe eres* species complex (Fig. 4). Fan et al. (2018c) treated many *Diaporthe* species as *D. eres*, and showed the combined cal, tef1-α and tub2 genes provide a better topology than the combined five-gene phylogeny for the *D. eres* complex. Both sequence data and morphology confirm that our isolates belong to this species (Fig. 4).

*Diaporthe rostrata* C.M. Tian, X.L. Fan & K.D. Hyde, Mycological Progress 14: 82 (2015)

≡ *Diaporthe juglandicola* C.M. Tian & Q. Yang. Mycosphere 8(5): 821 (2017)

**Description.** See Fan et al. (2015).

**Material examined.** CHINA, Beijing City, Mentougou District, Mount Dongling, Xiaolongmen Forestry Centre (39°57'54.68"N, 115°27'45.27"E), from branches of *Juglans mandshurica* Maxim., 22 Aug. 2017, H.Y. Zhu & X.L. Fan, deposited by X.L. Fan, CF 2019807, living culture CFCC 53142; *ibid.* CF 2019910, living culture CFCC 53143.

**Notes.** Fan et al. (2015) introduced *Diaporthe rostrata* from *Juglans mandshurica* causing walnut dieback in China. Yang et al. (2017) introduced *D. juglandicola* as a sister clade with *D. rostrata*, but it has no conspicuous rostrate necks on the bark. However, we recommend to treat *D. juglandicola* as a synonym of *D. rostrata*, based on the same host species, and lacking of phylogenetic support to separate them after involving our current materials (CF 2019807 and CF 2019910) with conspicuous rostrate necks.

**Erythrogloeaceae** Senan., Maharachch. & K.D. Hyde, Stud. Mycol. 86: 258 (2017)

**Type genus.** *Erythrogloeum* Petr. Sydowia 7: 378 (1953).

**Notes.** The family *Erythrogloeaceae* was recently introduced by Senanayake et al. (2017) based on ITS, LSU, rpb2 and tef1-a, and included four genera (*Chrysocrypta, Dendrostoma, Disculoides* and *Erythrogloeum*) (Fan et al. 2018a, Senanayake et al. 2018).
**Dendrostoma** X.L. Fan & C.M. Tian, Persoonia 40: 124 (2018)

**Type species.** *Dendrostoma mali* X.L. Fan & C.M. Tian, Persoonia 40: 124 (2018).

**Notes.** *Dendrostoma* was introduced by Fan et al. (2018a) as a phytopathogenic genus, causing canker diseases on several economic hardwoods such as *Malus spectabilis*, *Osmanthus fragrans* and *Quercus acutissima*. Jiang et al. (2019b) accepted 14 species of *Dendrostoma* using a concatenated matrix of four genes (ITS, LSU, *rpb2* and *tef1*-α), including 10 new species associated with chestnut and oak canker disease in China. Here we recommend a set of three genes (ITS, *rpb2* and *tef1*-α) to separate species of this genus.

**Dendrostoma donglinensis** H.Y. Zhu & X.L. Fan, sp. nov.
MycoBank No: 832194
Fig. 7

**Etymology.** Named after the location where it was collected, Mount Dongling.

**Holotype.** CHINA, Beijing City, Mentougou District, Mount Dongling, Xiaolongmen Forestry Centre (39°58′19.62″N, 115°26′51.27″E), from branches of *Quercus mongolica* Fisch. ex Ledeb., 18 Aug. 2017, H.Y. Zhu & X.L. Fan, deposited by X.L. Fan, holotype CF 2019903, ex-type living culture CFCC 53148.

**Description.** Sexual morph: not observed. Asexual morph: Pycnidial stromata immersed in the bark, scattered, erumpent through the surface of bark, unilocular, with a conspicuous central column. Central column beneath the disc more or less conical, yellow. Conceptacle absent. Ectostromatic disc hyaline, circular to ovoid, 750–1190 μm in diam., with a single ostiole per disc. Ostiole grey to black, at the same level as the disc surface, 240–270 μm in diam. Locule single, circular to irregular, undivided, 550–750 μm in diam. Conidiophores hyaline, unbranched, approximately cylindrical. Conidiogenous cells enteroblastic, phialidic. Conidia hyaline, fusoid, acute at each end, smooth or occasional not smooth, aseptate, 16.5–20.5 × 2–3.5 (av. = 18 ± 1.1 × 3 ± 0.3, n = 30) μm.

**Culture characteristics.** Cultures on PDA are initially white, growing slowly to 2 cm in diam. after 3 days and 4 cm after 14 days, becoming salmon in the center after 7–10 days. Growth stops when colony reaches 8 cm and cultures becoming salmon to honey after the 30 days. Colonies are felty with a uniform texture; sterile.

**Additional material examined.** CHINA, Beijing City, Mentougou District, Mount Dongling, Xiaolongmen Forestry Centre (39°58′19.62″N, 115°26′51.27″E), from branches of *Quercus mongolica* Fisch. ex Ledeb., 18 Aug. 2017, H.Y. Zhu & X.L. Fan, deposited by X.L. Fan, CF 2019887, living culture CFCC 53149; *ibid.* CF 2019805, living culture CFCC 53150.

**Notes.** *Dendrostoma donglinensis* is associated with canker disease of *Quercus mongolica* in China. It can be distinguished from its closest relative *D. parasiticum* by its
Figure 7. Morphology of *Dendrostoma donglinensis* from *Quercus mongolica* (CF 2019903). A–E Habit of conidiomata on twig F transverse section of conidioma G longitudinal section through conidioma H conidiophores and conidiogenous cells I conidia J colonies on PDA at 3 days (left) and 30 days (right). Scale bars: 1 mm (A); 500 μm (B–G); 10 μm (H, I).
fusoid, acute at each end and larger conidia (16.5–20.5 × 2–3.5 vs. 9.3–11.7 × 2.8–3.3 μm). The isolates are phylogenetically distinct from all other available strains of *Dendrostoma* included in this study and we therefore describe this species as new, based on DNA sequence data and morphology.

**Juglanconidaceae Voglmayr & Jaklitsch, Persoonia 38: 142 (2017)**

*Type genus.* *Juglanconis* Voglmayr & Jaklitsch, Persoonia 38: 142 (2017).

*Notes.* Juglanconidaceae was introduced by Voglmayr et al. (2017), including a single genus *Juglanconis*.

**Juglanconis Voglmayr & Jaklitsch, Persoonia 38: 142 (2017)**

*Type species.* *Juglanconis juglandina* (Kunze) Voglmayr & Jaklitsch, Persoonia 38: 144 (2017).

*Notes.* *Juglanconis* was introduced by Voglmayr et al. (2017) to accommodate previous *Melanconium juglandinum, M. oblongum* and *M. pterocaryae* based on morphology and DNA data of type materials. The genus is restricted to one host in Juglandaceae, which is identified by having perithecial ascomata, 8-spored asci with an apical ring, hyaline, bicelled ascospores in the sexual morph; and acervular conidiomata, brown conidia with gelatinous sheaths in asexual morph (Voglmayr et al. 2017). *Juglanconis* includes five species (*J. appendiculata, J. japonica, J. juglandina, J. oblonga* and *J. pterocaryae*) (Voglmayr et al. 2019), of which *J. juglandina* and *J. oblonga* are common pathogens in *Juglans* spp. in China (Fan et al. 2018b).

**Juglanconis oblonga** (Berk.) Voglmayr & Jaklitsch Persoonia 38: 147 (2017)

≡ *Melanconium oblongum* Berk., Grevillea 2 (22): 153 (1874)
≡ *Diaporthe juglandis* Ellis & Everh., Proc. Acad. Nat. Sci. Philadelphia 45: 448 (1893)
≡ *Melanconis juglandis* (Ellis & Everh.) A.H. Graves, Phytopathology 13: 311 (1923)

*Description.* See Fan et al. (2018b).

*Material examined.* CHINA, Beijing City, Mentougou District, Mount Dongling, Xiaolongmen Forestry Centre (39°57′54.68″N, 115°27′45.27″E), from branches of *Juglans mandshurica* Maxim., 22 Aug. 2017, H.Y. Zhu & X.L. Fan, deposited by X.L. Fan, CF 2019906, living culture CFCC 53151; *ibid.* CF 2019909, living culture CFCC 53152.

*Notes.* *Juglanconis oblonga* (previous *Melanconium oblongum*) is associated with canker disease of Juglandaceae hosts in North America and Southeast Asia (Graves 1923, Voglmayr et al. 2017, Fan et al. 2018b). This species is similar to *J. juglandina* in disease symptoms but can be distinguished by its longer conidia (22 × 12.5 compared...
to 20 × 13 μm) and DNA sequence data (Fan et al. 2018b). This species is a common pathogen causing walnut canker in China (Fan et al. 2018b).

**Melanconidaceae**

G. Winter, Rabenh. Krypt. -Fl., Edn 2 (Leipzig) 1(2): 764 (1886)

**Type genus.** *Melanconis* Tul. & C. Tul., Select. Fung. Carpol. (Paris) 2: 115 (1863).

**Notes.** Melanconidaceae was introduced by Winter (1886) and has been subject to some confusion due to the overlap in morphological characters between genera and the absence of DNA sequence data supporting the family concept (Barr 1978). Castlebury et al. (2002) and Rossman et al. (2007) restricted this family to a single genus *Melanconis* based on LSU rDNA sequences, which was adapted by recent studies (Senanayake et al. 2017, Fan et al. 2018b).

*Melanconis stilbostoma* (Fr.) Tul. & C. Tul., Select. Fung. Carpol. (Paris) 2: 115 (1863)

**Type species.** *Melanconis stilbostoma* (Fr.) Tul. & C. Tul., Select. Fung. Carpol. (Paris) 2: 115 (1863).

**Notes.** *Melanconis* was established by Tulasne & Tulasne (1863) based on *Sphaeria stilbostoma*. *Melanconis* has approximately 105 species epithets recorded in Index Fungorum (August 2019), but for most species no living cultures or DNA sequence data are available. Rossman et al. (2007) suggested that many of the species previously residing in *Melanconis* may belong elsewhere. *Melanconis* includes five species (*Melanconis alni, Ms. betulae, Ms. marginalis, Ms. itoana* and the type species *Ms. stilbostoma*), which were all restricted to the hosts in Betulaceae (Fan et al. 2016, 2018b).

**Material examined.** CHINA, Beijing City, Mentougou District, Mount Dongling, Xiaolongmen Forestry Centre (39°59'23.58"N, 115°27'05.00"E), from branches of *Betula daurica* Pall., 22 Aug. 2017, H.Y. Zhu & X.L. Fan, deposited by X.L. Fan, CF 2019832, living culture CFCC 53128; *ibid.* CF 2019833, living culture CFCC 53129. CHINA, Beijing City, Mentougou District, Mount Dongling, Xiaolongmen Forestry Centre (39°59'23.58"N, 115°27'05.00"E), from branches of *Betula* sp., 21 Aug. 2017, H.Y. Zhu & X.L. Fan, deposited by X.L. Fan, CF 2019871, living culture CFCC 53130; *ibid.* CF 2019911, living culture CFCC 53131.

**Notes.** *Melanconis stilbostoma* is the type species of *Melanconis* and is thus far only known to occur on *Betula* spp. with a global distribution (Fan et al. 2016). *Betula daurica, B. pendula, B. rotundifolia, B. tianschanica* and *B. platyphylla* are recorded as hosts for *Melanconis stilbostoma* in China (Zhuang 2005, Fan et al. 2016, 2018b).
Discussion

In the present work six diaporthalean species were identified residing in four families (Diaporthaceae, Erythrogloeaceae, Juglanconidaceae and Melanconidaceae) in the order Diaporthales. These include five known species (*Diaporthe betulina, D. eres, D. rostrata, Juglanconis oblonga* and *Melanconis stilbostoma*), and one new species (*Dendrostoma donglinensis*). All specimens in the current study were collected from symptomatic branches and twigs associated with canker or dieback diseases. *Dendrostoma* (Erythrogloeaceae) species were isolated from *Quercus mongolica* (Fagaceae). *Juglanconis* (Juglanconidaceae) species were isolated from *Juglans mandshurica* (Juglandaceae) and *Melanconis* (Melanconidaceae) species were isolated from *Betula dahurica* (Betulaceae), which suggests these fungi are host specific. *Diaporthe* (Diaporthaceae) species were isolated from *Betula dahurica* (Betulaceae), *Juglans regia, J. mandshurica* (Juglandaceae), *Prunus davidiana* (Rosaceae) and *Quercus mongolica* (Fagaceae). This might indicate that *Diaporthe* species are less host specific.

The classification of Diaporthales presented here follows the previous studies (Castlebury et al. 2002, Rossman et al. 2007) and discoveries of new taxa from many other works (Suetrong et al. 2015, Dissanayake et al. 2017, Voglmayr et al. 2017, Senanayake et al. 2017, 2018). We performed frequently and used four genes (ITS, LSU, *rpb2* and *tef1*-α) to evaluate the 30 families in this order, but it was found to be confusing in some taxa such as *Apoharknessia* and *Lasmenia* in Apoharknessiaceae (Fig. 2). It suggests that more studies using a multiphasic approach are still needed to clarify some issues in this order. Diaporthales includes many phytopathogenic genera such as *Dendrostoma, Diaporthe, Melanconis* and *Juglanconis*, which have been reported causing canker disease of tree hosts in China (Fan et al. 2016, 2018b, Yang et al. 2018, Jiang et al. 2019b). The current study focuses on diaporthalean fungi in Mount Dongling of Beijing, which is considered as a biodiversity hotspot with a high diversity for fungal species and (Guo et al. 2008, Zhu et al. 2018). We hope that the descriptions and molecular data of diaporthalean fungi in this study could provide a resource for future studies in this region.

Acknowledgements

This study is financed by the Fundamental Research Funds for the Central Universities (2019ZY23), the National Natural Science Foundation of China (31670647) and the College Student Research and Career-creation Program of Beijing (S201910022007).

References

Alvarez LV, Groenewald JZ, Crous PW (2016) Revising the Schizoparmaceae: *Coniella* and its synonyms *Pilidiella* and *Schizoparme*. Studies in Mycology 85: 1–34. https://doi.org/10.1016/j.simyco.2016.09.001
Diaporthalean fungi causing canker and dieback from Mount Dongling in China

Alves A, Crous PW, Correia A, Phillips AJL (2008) Morphological and molecular data reveal cryptic speciation in *Lasiodiplodia theobromae*. Fungal Diversity 28: 1–13. https://doi.org/10.1002/yea.1554

Barr ME (1978) The Diaporthales in North America with emphasis on *Gnomonia* and its segregates. Mycologia Memoir 7: 1–232.

Braun U, Nakashima C, Crous PW, Groenewald JZ, Moreno-Rico O, Rooney-Latham S, Blomquist CL, Haas J, Marmolejo J (2018) Phylogeny and taxonomy of the genus *Tubakia* s. lat. Fungal Systematics and Evolution 1: 41–99. https://doi.org/10.3114/fuse.2018.01.04

Carbone I, Kohn LM (1999) A method for designing primer sets for speciation studies in filamentous ascomycetes. Mycologia 91: 553–556. https://doi.org/10.2307/3761358

Castlebury LA, Rossman AY, Jaklitsch WJ, Vasilyeva LN (2002) A preliminary overview of the Diaporthales based on large subunit nuclear ribosomal DNA sequences. Mycologia 94: 1017–1031. https://doi.org/10.1080/15572536.2003.11833157

Crous PW, Gams W, Stalpers JA, Robert V, Stegehuis G (2004) MycoBank: an online initiative to launch mycology into the 21st century. Studies in Mycology 50: 19–22.

Crous PW, Schumacher RK, Akulov A, Thangavel R, Hernández-Restrepo M, Carnegie A, Cheewangkoon R, Wingfield MJ, Summerell B, Quaedvlieg W, Coutinho TA, Roux J, Wood AR, Giraldo A, Groenewald JZ (2019) New and Interesting Fungi. 2. Fungal Systematics and Evolution 3: 57–134. https://doi.org/10.3114/fuse.2019.03.06

Crous PW, Summerell BW, Shivas RG, Burgess TI, Decock CA, Dreyer LL, Granke LL, Guest DI, Hardy GESTJ, Hausbeck MK, Hüberli D, Jung T, Koukol O, Lennox CL, Liew ECY, Lombard L, McTaggart AR, Pryke JS, Roets F, Saude C, Shuttleworth LA, Stukely MJC, Váňky K, Webster BJ, Windstam ST, Groenewald JZ (2012) Fungal Planet description sheets: 107–127. Persoonia 28: 138–182. https://doi.org/10.3767/003158512X652633

Dissanayake AJ, Phillips AJL, Hyde KD, Yan JY, Li XH (2017) The current status of species in *Diaporthe*. Mycosphere 8: 1106–1156. https://doi.org/10.5943/mycosphere/8/5/5

Doyle JJ, Doyle JL (1990) Isolation of plant DNA from fresh tissue. Focus 12: 13–15. https://doi.org/10.2307/2419362

Du Z, Fan XL, Hyde KD, Yang Q, Liang YM, Tian CM (2016) Phylogeny and morphology reveal two new species of *Diaporthe* from Betula spp. in China. Phytotaxa 269: 90–102. https://doi.org/10.11646/phytotaxa.269.2.2

Fan XL, Bezerra JDP, Tian CM, Crous PW (2020) *Cytospora* (Diaporthales) in China. Persoonia 45: 1–45. https://doi.org/10.3767/persoonia.2020.45.01

Fan XL, Bezerra JDP, Tian CM, Crous PW (2018a) Families and genera of diaporthalean fungi associated with canker and dieback of tree hosts. Persoonia 40: 119–134. https://doi.org/10.3767/persoonia.2018.40.05

Fan XL, Du Z, Bezerra JDP, Tian CM (2018b) Taxonomic circumscription of melanconis-like fungi causing canker disease in China. MycoKeys 42: 89–124. https://doi.org/10.3897/mycokeys.42.29634

Fan XL, Yang Q, Bezerra JDP, Alvarez LV, Tian CM (2018c) *Diaporthe* from walnut tree (*Juglans regia*) in China, with insight of the *Diaporthe orhei* complex. Mycological Progress 17: 841–853. https://doi.org/10.1007/s11557-018-1395-4
Fan XL, Du Z, Liang YM, Tian CM (2016) Melanconis (Melanconidaceae) associated with Betula spp. in China. Mycological Progress 15: 1–9. https://doi.org/10.1007/s11557-016-1163-2
Fan XL, Hyde KD, Udayanga D, Wu XY, Tian CM (2015) Diaporthe rostrata, a novel ascomycete from Juglans mandshurica associated with walnut dieback. Mycological Progress 14: 1–8. https://doi.org/10.1007/s11557-015-1104-5
Glass NL, Donaldson GC (1995) Development of primer sets designed for use with the PCR to amplify conserved genes from flamentous ascomycetes. Applied and Environmental Microbiology 61: 1323–1330.
Gomes RR, Glienke C, Videira SIR, Lombard L, Groenewald JZ, Crous PW (2013) Diaporthe: a genus of endophytic, saprobic and plant pathogenic fungi. Persoonia 31: 1–41. https://doi.org/10.3767/003158513X666844
Graves AH (1923) The Melanconis disease of the butternut (Juglans cinerra L.). Phytopathology 13: 411–435.
Guarnaccia V, Crous PW (2017) Emerging citrus diseases in Europe caused by species of Diaporthe. IMA Fungus 8: 317–334. https://doi.org/10.5598/imafungus.2017.08.02.07
Guindon S, Dufayard JF, Lefort V, Anisimova M, Hordijk W, Gascuel O (2010) New algorithms and methods to estimate maximum-likelihood phylogenies: assessing the performance of PhyML 3.0. Systematic Biology 59: 307–321. https://doi.org/10.1093/sysbio/syq010
Guo LD, Huang GR, Wang Y (2008) Seasonal and tissue age influences on endophytic fungi of Pinus tabulaeformis (Pinaceae) in the Dongling Mountains, Beijing. Journal of Integrative Plant Biology 50: 997–1003. https://doi.org/10.1111/j.1744-7909.2008.00394.x
Guterres DC, Galvão-Elias S, Santos MDM, Souza BCP, Almeida CP, Pinho DB, Miller RNG, Dianese JC (2019) Phylogenetic relationships of Phaeochorella parinarii and recognition of a new family, Phaeochorellaceae (Diaporthales). Mycologia. https://doi.org/10.1080/00275514.2019.1603025
Hillis DM, Bull JJ (1993) An empirical test of bootstrapping as a method for assessing confidence in phylogenetic analysis. Systematic Biology 42: 182–192. https://doi.org/10.1093/sysbio/42.2.182
Hongsanan S, Maharachchikumbura SSN, Hyde KD, Samarakoon MC, Jeewon R, Zhao Q, Al-Sadi AM, Bahkali AH (2017) An updated phylogeny of Sordariomycetes based on phylogenetic and molecular clock evidence. Fungal Diversity 84: 1–17. https://doi.org/10.1007/s13225-017-0384-2
Jiang N, Fan XL, Tian CM (2019a) Identification and pathogenicity of Cryphonectriaceae species associated with chestnut canker in China. Plant Pathology 68: 1132–1145. https://doi.org/10.1111/ppl.13033
Jiang N, Fan XL, Crous PW, Tian CM (2019b) Species of Dendrostoma (Erythrogloeaceae, Diaporthales) associated with chestnut and oak canker diseases in China. MycoKeys 48: 67–96. https://doi.org/10.3897/mycokeys.48.31715
Katoh K, Standley DM (2013) MAFFT multiple sequence alignment software version 7: improvements in performance and usability. Molecular biology and evolution 30: 772–780. https://doi.org/10.1093/molbev/mst010

Guo LD, Huang GR, Wang Y (2008) Seasonal and tissue age influences on endophytic fungi of Pinus tabulaeformis (Pinaceae) in the Dongling Mountains, Beijing. Journal of Integrative Plant Biology 50: 997–1003.
Guindon S, Dufayard JF, Lefort V, Anisimova M, Hordijk W, Gascuel O (2010) New algorithms and methods to estimate maximum-likelihood phylogenies: assessing the performance of PhyML 3.0. Systematic Biology 59: 307–321.
Hillis DM, Bull JJ (1993) An empirical test of bootstrapping as a method for assessing confidence in phylogenetic analysis. Systematic Biology 42: 182–192.
Hongsanan S, Maharachchikumbura SSN, Hyde KD, Samarakoon MC, Jeewon R, Zhao Q, Al-Sadi AM, Bahkali AH (2017) An updated phylogeny of Sordariomycetes based on phylogenetic and molecular clock evidence. Fungal Diversity 84: 1–17.
Jiang N, Fan XL, Tian CM (2019a) Identification and pathogenicity of Cryphonectriaceae species associated with chestnut canker in China. Plant Pathology 68: 1132–1145.
Jiang N, Fan XL, Crous PW, Tian CM (2019b) Species of Dendrostoma (Erythrogloeaceae, Diaporthales) associated with chestnut and oak canker diseases in China. MycoKeys 48: 67–96.
Katoh K, Standley DM (2013) MAFFT multiple sequence alignment software version 7: improvements in performance and usability. Molecular biology and evolution 30: 772–780.

Liu YJ, Whelen S, Hall BD (1999) Phylogenetic relationships among ascomycetes: evidence from an RNA polymerase II subunit. Molecular Biology and Evolution 16: 1799–1808. https://doi.org/10.1093/oxfordjournals.molbev.a026092

Ma KP, Guo XM, Yu SL (1995) On the characteristics of the flora of Mount Dongling area and its relationship with a number of other mountainous floras in China. Bulletin Botanical Research 15: 501–515.

Nitschke T (1870) Pyrenomycetes Germanici 2. Eduard Trewendt, Breslau.

Norphanphoun C, Hongsanan S, Doilom M, Bhat DJ, Wen TC, Bulgakov TS, Hyde KD (2016) Lamproconiaceae fam. nov. to accommodate Lamproconium desmazieri. Phytotaxa 270: 89–102. https://doi.org/10.11646/phytotaxa.270.2.2

Posada D, Crandall KA (1998) Modeltest: testing the model of DNA substitution. Bioinformatics 14: 817–818. https://doi.org/10.1093/bioinformatics/14.9.817

Rannala B, Yang Z (1996) Probability distribution of molecular evolutionary trees: a new method of phylogenetic inference. Journal of Molecular Evolution 43: 304–311. https://doi.org/10.1007/BF02338839

Rayner RW (1970) A Mycological Colour Chart. Commonwealth Mycological Institute, Kew.

Ronquist F, Huelsenbeck JP (2003) MrBayes 3: Bayesian phylogenetic inference under mixed models. Bioinformatics 19: 1572–1574. https://doi.org/10.1093/bioinformatics/btg180

Rossman AF, Farr DF, Castlebury LA (2007) A review of the phylogeny and biology of the Diaporthales. Mycyscience 48: 135–144. https://doi.org/10.1007/S10267-007-0347-7

Senanayake IC, Crous PW, Groenewald JZ, Maharachchikumbura SSN, Jeewon R, Phillips AJL, Bhat DJ, Perera RH, Li QR, Li WJ, Tanghirasunun N, Norphanphoun C, Karunarathna SC, Erio C, Manawasighe IS, Al-Sadi AM, Hyde KD (2017) Families of Diaporthales based on morphological and phylogenetic evidence. Studies in Mycology 86: 217–296. https://doi.org/10.1016/j.simyco.2017.07.003

Senanayake IC, Jeewon R, Chomnunti P, Wanasinghe DN, Norphanphoun C, Karunarathna A, Pem D, Perera RH, Cameroesi E, McKenzie EHC, Hyde KD, Karunarathna SC (2018) Taxonomic circumscription of Diaporthales based on multigene phylogeny and morphology. Fungal Diversity 93: 241–443. https://doi.org/10.1007/s13225-018-0410-z

Suetrong S, Klaysuban A, Sakayaroj J, Preedanon S, Ruang-Areeate P, Phongpaichit S, Pang KL, Jones EBG (2015) Tirisporellaceae, a new family in the order Diaporthales (Sordariomycetes, Ascomycota). Cryptogamie Mycologie 36: 319–330. https://doi.org/10.7872/crym/v36.iss3.2015.319

Sun X, Guo LD, Hyde KD (2011) Community composition of endophytic fungi in Acer truncatum and their role in decomposition. Fungal diversity 47: 85–95. https://doi.org/10.1007/s13225-010-0086-5

Swofford DL (2003) PAUP*: Phylogenetic Analysis Using Parsimony, * and Other Methods, Version 4.0b10, Sinauer Associates, Sunderland.

Tamura K, Stecher G, Peterson D, Filipski A, Kumar S (2013) MEGA6: molecular evolutionary genetics analysis version 6.0. Molecular Biology and Evolution 30: 2725–2729. https://doi.org/10.1093/molbev/mst197

Tulasne LR, Tulasne C (1863) Selecta Fungorum Carpologia, Vol. 2. Paris.
Udayanga D, Castlebury LA, Rossman AY, Chukeatirote E, Hyde KD (2014) Insights into the genus *Diaporthe*: phylogenetic species delimitation in the *D. eres* species complex. Fungal Diversity 67: 203–229. https://doi.org/10.1007/s13225-014-0297-2

Udayanga D, Liu XZ, Crous PW, McKenzie EHC, Chukeatirote E, Hyde KD (2012) A multi-locus phylogenetic evaluation of *Diaporthe* (*Phomopsis*). Fungal Diversity 56: 157–171. https://doi.org/10.1007/s13225-012-0190-9

Vilgalys R, Hester M (1990) Rapid genetic identification and mapping of enzymatically amplified ribosomal DNA from several Cryptococcus species. Journal of Bacteriology 172: 4238–4246. https://doi.org/10.1128/jb.172.8.4238-4246.1990

White TJ, Bruns T, Lee S, Taylor J (1990) Amplification and direct sequencing of fungal ribosomal RNA genes for phylogenetics. PCR Protocols: a guide to methods and applications 18: 315–322. https://doi.org/10.1016/B978-0-12-372180-8.50042-1

Winter G (1886) Fungi Australienses. Revue Mycologique Toulouse 8: 207–213.

Xavier KV, KC AN, Crous PW, Groenewald JZ, Vallad GE (2019) *Dwiroopa punicae* sp. nov. (Dwiroopaceae fam. nov., Diaporthales), associated with leaf spot and fruit rot of pomegranate (*Punica granatum*). Fungal Systematics and Evolution 4: 33–41. https://doi.org/10.3114/fuse.2019.04.04

Yang Q, Fan XL, Du Z, Tian CM (2017) *Diaporthe juglandicola* sp. nov. (Diaporthales, Ascomycetes), evidenced by morphological characters and phylogenetic analysis. Mycosphere 8: 817–826. https://doi.org/10.5943/mycosphere/8/5/3

Yang Q, Fan XL, Guaraccia V, Tian CM (2018) High diversity of *Diaporthe* species associated with dieback diseases in China, with twelve new species described. MycoKeys 39: 97–149. https://doi.org/10.3897/mycokeys.39.26914

Zhu HY, Tian CM, Fan XL (2018) Studies of botryosphaeriaceous fungi associated with canker and dieback of tree hosts in Dongling Mountain of China. Phytotaxa 348: 63–76. https://doi.org/10.11646/phytotaxa.348.2.1

Zhuang WY (2005) Fungi of northwestern China. Ithaca, New York.
Supplementary material 1

Table S1. Isolates and GenBank accession numbers used in the phylogenetic analyses of Diaporthales
Authors: Haiyan Zhu, Meng Pan, Guido Bonthond, Chengming Tian, Xinlei Fan
Data type: molecular data
Copyright notice: This dataset is made available under the Open Database License (http://opendatacommons.org/licenses/odbl/1.0/). The Open Database License (ODbL) is a license agreement intended to allow users to freely share, modify, and use this Dataset while maintaining this same freedom for others, provided that the original source and author(s) are credited.
Link: https://doi.org/10.3897/mycokeys.59.38055.suppl1

Supplementary material 2

Table S2. Isolates and GenBank accession numbers used in the phylogenetic analyses of Diaporthe
Authors: Haiyan Zhu, Meng Pan, Guido Bonthond, Chengming Tian, Xinlei Fan
Data type: molecular data
Copyright notice: This dataset is made available under the Open Database License (http://opendatacommons.org/licenses/odbl/1.0/). The Open Database License (ODbL) is a license agreement intended to allow users to freely share, modify, and use this Dataset while maintaining this same freedom for others, provided that the original source and author(s) are credited.
Link: https://doi.org/10.3897/mycokeys.59.38055.suppl2

Supplementary material 3

Table S3. Isolates and GenBank accession numbers used in the phylogenetic analyses of Diaporthe erez complex
Authors: Haiyan Zhu, Meng Pan, Guido Bonthond, Chengming Tian, Xinlei Fan
Data type: molecular data
Copyright notice: This dataset is made available under the Open Database License (http://opendatacommons.org/licenses/odbl/1.0/). The Open Database License (ODbL) is a license agreement intended to allow users to freely share, modify, and use this Dataset while maintaining this same freedom for others, provided that the original source and author(s) are credited.
Link: https://doi.org/10.3897/mycokeys.59.38055.suppl3
Supplementary material 4

Table S4. Isolates and GenBank accession numbers used in the phylogenetic analyses of *Dendrostoma*

Authors: Haiyan Zhu, Meng Pan, Guido Bonthond, Chengming Tian, Xinlei Fan

Data type: molecular data

Copyright notice: This dataset is made available under the Open Database License (http://opendatacommons.org/licenses/odbl/1.0/). The Open Database License (ODbL) is a license agreement intended to allow users to freely share, modify, and use this Dataset while maintaining this same freedom for others, provided that the original source and author(s) are credited.

Link: https://doi.org/10.3897/mycokeys.59.38055.suppl4