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Full Length Article

Powdery-Fruit Disease of *Cinnamomum burmannii* and its Influence on Fruit Essential Oil

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Abstract

Powdery-fruit disease is a notorious disease of *Cinnamomum burmannii*, causing severe loss to fruit production. In this study, the pathogen causing powdery-fruit disease of *C. burmannii* was identified based on morphological characters and 18S rDNA gene sequences. Three essential oils samples from the leaves and normal and infected fruits of *C. burmannii* were obtained by hydro-distillation and analyzed by GC-MS. The composition of the infected fruits was strongly affected by the pathogen of powdery-fruit disease. This study provides new knowledge for the change in chemicals before and after pathogen infection in the fruits of *C. burmannii*, paving the way to further reveal the unknown mechanism of pathogen infection in *C. burmannii*. © 2020 Friends Science Publishers

Keywords: Powdery-fruit disease; *Cinnamomum burmannii*; Essential oils; GC-MS

Introduction

Cinnamomum burmannii (Nees and Nees) Blume is a cost-effective and significant evergreen tree from the Lauraceae family, widely distributed in Vietnam, Indonesia, Philippines, Myanmar, India and south of the Yangtze River in China (Yang *et al.* 2019). The Lauraceae family has essential ecological and economic value particularly in the provisioning of non-timber resources. In addition, extracted oils are widely used in perfumery, food additives, and medicine (Reis-Avila and Oliveira 2017). *Cinnamomum* is regarded as one of the most commercially valuable genera among the Lauraceae family owing to the abundance of volatile oils (Jayaprakasha and Rao 2011). Essential oils are complex mixtures of natural compounds produced by plant metabolism and are responsible for the distinctive aroma of the plants (Pavela 2015). Over the past decades, essential oils derived from many medicinal plant species have been analyzed for their chemical components and prominent biological activities (Dra *et al*. 2017; Osanloo *et al*. 2017; Nascimento *et al*. 2018). The bark of *C. burmannii* has been used to treat rheumatism, diarrhea and abdominal pain in Chinese medicine (Tan *et al*. 2011). The roots, bark, and leaves of this plant can be used to extract aromatic oils or pigments that are widely used in the spice and pharmaceutical industries (Shan *et al*. 2007; Huang *et al*. 2011). In the meantime, *C. burmannii* is environmentally friendly and is also good for human health, as *C. burmannii* has long been used as a precious perfume material (Wang *et al*. 2019). In the previous phytochemical investigations, cinnamaldehyde, eugenol, coumarin, citronellol, borneol, eucalyptol and various alkenes have been identified from the essential oils derived from the leaves and stems of *C. burmannii* (Liu *et al*. 2007; Wang *et al*. 2009; AI-Dhubiab 2012; Li *et al*. 2016; Kumar and Kumari 2019). However, most of the essential oils tested in the previous studies were extracted from the *C. burmannii* leaves and bark. The chemical composition and characterizations of volatile oils from the fruits of *C. burmannii* have not been studied.

The fruits of *C. burmannii* can also be potentially used as a source of pigments for the exploitation of neoteric dyes (Tan *et al.* 2011). Surprisingly, fungal infection of fruits has rarely attracted attention, compared to the detailed studies on leaves, twigs and stems of *Cinnamomum* (Liu and Xu 2014; Shan *et al.* 2014; Jiang and Kirschner 2016). Powdery-fruit disease is a common and severe fruit disease of *C. burmannii*, *C. cassia* and *C. camphora*, and the pathogen can vary widely in different hosts (Chen *et al*. 2013; Shan *et al.* 2014). Previous reports have shown that powdery-fruit disease of *C. burmannii* and *C. camphora*

was caused by the plant pathogenic fungus *Clinoconidium cinnamomi* and *Clinoconidium sawadae*, respectively (Jiang and Kirschner 2016). The plant pathogenic fungus *Clinoconidium cinnamomi* was first described as *Elaeodema cinnamomi*, while *C. sawadae* was firstly recorded as *Exobasidium sawadae* (Guo *et al*. 1991). Fruits of *Cinnamomum* species were mainly infected by *Elaeodema* and *Exobasidium* species, which occurred in East Asia. These pathogen species were not conspecific, and the differentiation was spore production. The spores were produced by basidia in *Exobasidium* and hyphae in *Elaeodema*. In addition, *Elaeodema* is an endemic genusin China (Guo *et al*. 1991). Taking these perplexing literature reports into consideration, it is important to reinvestigate the taxonomic affiliations by sampling different types of infected fruits of *C. burmannii* for further analysis.

To the best of our knowledge, the distinction of the volatile oils between normal and infected fruits of *C. burmannii* has not been studied. In addition, there are only a few reports on the pathogen of powdery-fruit disease of *C. burmannii*. This study focuses on identifying the pathogen and investigating the compositions of the volatile oils derived from the leaves and normal and infected fruits of *C. burmannii*, aiming to reveal the effects of powdery-fruit disease on the fruits of *C. burmannii*.

Materials and Methods

Plant material

Fresh leaves and normal and infected fruits of *C. burmannii* (2 kg) were collected from Huolu Mountain, Guangzhou, China, in November 2018, and the ripe spores were collected from the same spots in March 2019. The voucher specimens were deposited in the College of Forestry and Landscape Architecture, South China Agricultural University. The leaves and normal and infected fruits were stored at −20°C before use.

Morphological identification

The identification of the pathogen causing powdery-fruit disease was performed by observing morphological characters such as macroscopic traits of the infected fruits and the microscopic appearance of spores (Guo *et al*. 1991).

DNA extraction, NS-rDNA amplification and sequence analysis

Fresh spore samples 50–100 mg in size were harvested from the fresh infected fruits and were used for genomic DNA extraction. The genomic DNA extraction, polymerase chain reaction and sequencing of internal transcribed spacer (ITS, primer ITS 1: 5'-TCCGTAGGTGAACCTGCGG-3'; primer ITS 4: 5'-TCCTCCGCTTATTGATATGC-3') and 18S rDNA sequences (primer NS1: 5'-GTATCATATGCTTGTCTC-3'; primer NS8: 5'-TCCGCAGGTTCACCTACGGA-3') from the infected fruits were operated as described in procedures from our previous reports (Shan *et al*. 2019). The sequences were first edited by the BLASTN program against the database (NCBI), and were then submitted to GenBank to obtain the accession numbers.

Phylogenetic analysis

To construct a phylogeny of the pathogen, we retrieved sequences of the close hits from the NCBI (http://www.ncbi.nlm.nih.gov/) and performed multiple sequence alignment using Clustal W. The aligned files were then exported in mega format. A neighbor-joining tree was constructed in MEGA 6.0 using default parameters and bootstrap values calculated from 1,000 replications (Shan *et al*. 2019).

Preparation of theessential oils

The volatile oils of leaves and normal and infected fruits were isolated by hydro-distillation using a Clevenger-type apparatus for 6 h at 100°C. Distillates were further extracted with diethyl ether, and extractions were dried over anhydrous sodium sulfate, followed by filtration. All the essential oils were stored at 4°C in sealed dark glass vials for further use (Shan *et al.* 2016).

Essential oils analysis

The chemical composition of the volatile oils was analyzed by GC-MS according to Shan *et al.* (2016). Identical column and conditions were used for both GC and GC-MS. The components were indicated by comparison of their mass with NIST 2011 library data. The relative amount (RA) of each individual component from the volatile oils was expressed as the percentage of the peak area corresponding to the total peak area.

Results

Morphological identification and characterization of powdery-fruit disease pathogeny

The outward appearances of normal and infected fruits were shown in Fig. 1. The infected fruits displayed as woolen, flat spherical or irregular flat spherical shape, with diameters reaching 1.0 ~ 2.5 cm. The fruits of *C. burmannii* transformed into galls with reddish-brown, slightly scaly skin that fractured and peeled off, releasing buff ochre masses of spores when they were mature. During the process, the core was still the host organization. Interestingly, the powdery-fruit disease only affects the fruits of *C. burmannii*. Coincidentally, the time when the pathogen spores become mature is identical to *C. burmanni*

(**a**) (**b**) (**c**)

Fig. 1: The outward appearance of powdery-fruit disease in *C. burmannii*. (**a**) Normal and infected fruits in the growing condition. (**b**) Collected normal fruits. (**c**) Collected infected fruits

Fig. 2: Morphological characteristics of the spores

flowering. Therefore, it is suspected that the pathogen spores probably invade the host through the flowers.

The spores shown in Fig. 2 were long ellipsoidal or oval, unicellular, and pistac with several hyaline oil globules. The spores measured up to 11.6 to 15.2 μm long and 5.2 to 8.1 μm wide (n = 30).

Molecular identification and phylogenetic analysis

From the amplification of the 18S rDNA region, an approximately 2, 071-bp PCR product was obtained for the pathogen CBd1. The sequence of the pathogen CBd1 was uploaded to GenBank, with accession number MH378887.

For phylogenetic analysis, a total of nineteen sequences among four type strains were obtained from GenBank. The 18S rDNA (NS) sequence was used to construct the phylogenetic tree. The phylogenetic tree revealed that the pathogen CBd1 formed a clade with members of the genus *Acaromyces* (Fig. 3). *Acaromyces ingoldii* (accession number NG061199) was a type strain of *Acaromyces* sp. and the pathogen CBd1 sequence had 99.63% similarity with the *Acaromyces ingoldii* sequence (accession number NG061199 and KP866248).

Essential oil analysis

By hydro-distillation, the isolated yields of the volatile oils from the fresh leaves and fruits of both normal and infected of *C. burmannii* were 0.160, 0.084 and 0.044% (w/w), respectively. These results revealed that the content of essential oil in leaves was significantly higher than that in fruits, with the content in normal fruits being nearly 2-fold that of infected fruits. The results data from GC-MS analysis were shown in Table 1. The chemical components of the essential oils from three different samples indicated significant variations in type and quantity. To evaluate leaf oil, at least sixty-four compounds were authenticated, which accounted for 96.20% of the total contents. Twenty-eight compounds were identified from the volatile oil of normal fruits, in contrast to fifty-two compounds in infected fruits, which constituted 89.33 and 91.48% of the total oils, respectively. These results indicated that the numbers of chemical components in infected fruits were almost twice those from normal fruits.

Additionally, the types and relative percentages of chemical components in the three essential oils also vary greatly. Borneol (24.99%), *D*-limonene (11.16%), *L*-(-)-borneol (10.51%), α-pinene (4.73%), bornyl acetate (4.56%), terpineol (3.66%) and β-pinene (3.18%) were major components in the leaf oil, with a total percentage of 62.79%. α-Caryophyllene (32.94%), *L*-caryophyllene (17.75%), 1, 5, 5, 8-tetramethyl-3, 7-cycloundecadien-1-ol (5.83%), epizonarene (4.82%), caryophyllene oxide (4.63%) and *o*-menth-8-ene (4.42%) were major components in normal fruits, with a total percentage of 70.39%. In contrast, in infected fruits, the major components were β-guaiene (10.01), (-)-β-cadinene (8.64%), l-caryophyllene (5.78%), pinene (5.68%) and α-caryophyllene (5.25%), with a total percentage of 35.36% in the oils. β-Guaiene was the most abundant ingredient in infected fruits and no other components were above 10%. Other components were present at less than 4% in infected fruits.

In addition, four common components, *D*-limonene, l-caryophyllene, α-caryophyllene and δ-cadinene were found in all the three essential oils. Peruviol was detected in both leaves and normal fruits but not in infected fruits. Ten common constituents were found in both normal and infected fruits: *D*-limonene, l-caryophyllene, α-caryophyllene, epizonarene, γ-selinene, δ-cadinene, 10s,11s-himachala -3(12), 4-diene, (+) -α-elemene, β-guaiene and longifolene, though they were variable in contents. For example, the relative content of α-caryophyllene in normal fruits was 32.94%, while that in infected fruits was 5.25%. In addition, l-caryophyllene accounted for 17.75% in normal fruits and 5.78% in infected fruits. Oppositely, the relative content of *D*-limonene, γ-selinene, δ-cadinene, 10s, 11s-himachala -3 (12), 4-diene, (+) -α-elemene and β-guaiene were richer in infected fruits.

Discussion

A distinct mycelium is an important characteristic of a pathogen. Unfortunately, it was challenging to obtain the CBd1pathogen from Basidiomycota since it cannot be cultured on an artificial medium. Multigene sequencing has

Table 1: Chemical compositions of the essential oils from the leaves, normal and infected fruits of *C. burmannii*

No.	Compound	Molecular formula	Peak area (%)		
			Leaves	Normal fruits	Infected fruits
1	3-Thujene	$C_{10}H_{16}$	0.76	–[a]	–
2	*α*-Pinene	$C_{10}H_{16}$	4.73	–	5.68
3	Camphene	$C_{10}H_{16}$	2.95	–	1.93
4	Sabenene	$C_{10}H_{16}$	0.50	–	–
5	(*E*)-1,1-Dimethyl-2-(3-methylbuta-1,3-dien-1-yl) cyclopropane	$C_{10}H_{16}$	–	–	0.49
6	*β*-Pinene	$C_{10}H_{16}$	3.18	–	–
7	Myrcene	$C_{10}H_{16}$	2.41	–	0.22
8	*α*-Phellandrene	$C_{10}H_{16}$	1.71	–	–
9	*p*-Cymol	$C_{10}H_{14}$	–	–	0.47
10	*α*-Terpinene	$C_{10}H_{16}$	0.14	–	–
11	*D*-Limonene	$C_{10}H_{16}$	11.16	0.21	2.10
12	Eucalyptol	$C_{10}H_{18}O$	2.08	–	1.21
13	Salicylic aldehyde	$C_7H_6O_2$	0.19	–	–
14	Ocimene	$C_{10}H_{16}$	0.21	–	0.14
15	*γ*-Terpinene	$C_{10}H_{16}$	0.84	–	–
16	Terpinolene	$C_{10}H_{16}$	1.85	–	–
17	Cyclohexene, 3-methyl-6-(1-methylethylidene)-	$C_{10}H_{16}$	–	–	0.35
18	Linalool	$C_{10}H_{18}O$	0.86	–	–
19	Fenchol	$C_{10}H_{18}O$	–	–	0.52
20	(+)-2-bornanone	$C_{10}H_{16}O$	0.88	–	–
21	Borneol	$C_{10}H_{18}O$	24.99	–	0.81
22	*L*-(-)-Borneol	$C_{10}H_{18}O$	10.51	–	–
23	(-)-4-Terpineol	$C_{10}H_{18}O$	0.68	–	–
24	*α*-Terpinylisovalerate	$C_{15}H_{26}O_2$	–	0.14	–
25	2-(4-Methylphenyl)propan-2-ol	$C_{10}H_{14}O$	0.09	–	–
26	Terpineol	$C_{10}H_{18}O$	3.66	–	–
27	Decanal	$C_{10}H_{20}O$	0.27	–	–
28	Octyl acetate	$C_{10}H_{20}O_2$	0.18	–	–
29	*α*-Bornene	$C_{10}H_{16}$	–	–	2.96
30	(z)-3,7-dimethylocta-2,6-dienal	$C_{10}H_{16}O$	0.06	–	–
31	Geraniol	$C_{10}H_{18}O$	1.23	–	–
32	Citral	$C_{10}H_{16}O$	0.09	–	–
33	2,4,6-Trimethyl-1,3,6-heptatriene	$C_{10}H_{16}$	–	–	0.11
34	1-Decanol	$C_{10}H_{22}O$	0.35	–	–
35	Bornyl acetate	$C_{12}H_{20}O_2$	4.56	–	0.37
36	*γ*-Pyronene	$C_{10}H_{16}$	0.83	–	–
37	Isoterpinolene	$C_{10}H_{16}$	–	–	0.32
38	Neryl acetate	$C_{12}H_{20}O_2$	0.23	–	–
39	1, 2-Dimethylspiro [4.4]nona-1, 3-diene	$C_{11}H_{16}$	–	0.26	–
40	Ylangene	$C_{15}H_{24}$	0.18	–	–
41	Clovene	$C_{15}H_{24}$	–	0.29	–
42	*α*-Copaene	$C_{15}H_{24}$	0.19	–	–
43	Isolongifolene	$C_{10}H_{16}$	–	–	0.27
44	Geranyl acetate	$C_{12}H_{20}O_2$	0.87	–	–
45	*α*-Funebrene	$C_{15}H_{24}$	-	–	0.55
46	(-)-*β*-Elemene	$C_{15}H_{24}$	0.19	–	–
47	3-Acetyl-4-ethenylphenyl acetate	$C_{12}H_{12}O_3$	–	–	0.24
48	Cyclodecane	$C_{10}H_{20}$	0.08	–	–
49	l-Caryophyllene	$C_{15}H_{24}$	2.81	17.75	5.78
50	Calarene	$C_{15}H_{24}$	0.10	–	–
51	1*H*-Benzocycloheptene,2,4a,5,6,7,8-hexahydro-3,5,5,9-tetramethyl-, (4a*R*)-	$C_{15}H_{24}$	–	0.19	–
52	Coumarin	$C_9H_6O_2$	0.47	–	–
53	Isoledene	$C_{15}H_{24}$	0.09	–	2.62
54	(+)-Aromadendrene	$C_{15}H_{24}$	0.45	–	–
55	Cinnamyl acetate	$C_{15}H_{24}$	1.35	–	–
56	*β*-Copaene	$C_{15}H_{24}$	–	0.57	–
57	*α*-Caryophyllene	$C_{15}H_{24}$	0.67	32.94	5.25
58	a-Gurjunene	$C_{15}H_{24}$	0.10	–	–
59	*α*-Guaiene	$C_{15}H_{24}$	–	–	0.15
60	Alloaromadendrene	$C_{15}H_{24}$	0.14	–	–
61	*trans*-Calamenene	$C_{15}H_{22}$	-	–	0.41
62	*γ*-Muurolene	$C_{15}H_{24}$	0.28	–	–
63	3,4-Dihydrocoumarin, 4,4-dimethyl-6-ethyl-	$C_{13}H_{16}O_2$	–	0.20	–
64	Germacrene D	$C_{15}H_{24}$	0.20	–	–
65	Cedrene-V6	$C_{15}H_{24}$	–	–	1.16

Table 1: Continued

Table 1: Continued

66	*β*-Eudesmene	$C_{15}H_{24}$	0.10	–	–
67	2-Isopropenyl-4a,8-dimethyl-1,2,3, 4,4a,5,6,7-octahydronaphthalene	$C_{15}H_{24}$	–	1.55	–
68	Epizonarene	$C_{15}H_{24}$	–	4.82	1.85
69	(-)-*β*-Cadinene	$C_{15}H_{24}$	–	–	8.64
70	Azulene,1,2,3,3a,4,5,6,7-octahydro-1,4-dimethyl-7-(1-methylethenyl)-, (1*R*,3a*R*,4*R*,7*R*)	$C_{15}H_{24}$	–	–	3.17
71	(+)-Valencene	$C_{15}H_{24}$	–	0.58	–
72	*δ*-Selinene	$C_{15}H_{24}$	0.15	–	–
73	(-)-*α*-Selinene	$C_{15}H_{24}$	–	2.73	–
74	*γ*-Selinene	$C_{15}H_{24}$	–	1.38	2.97
75	Bicyclogermacrene	$C_{15}H_{24}$	1.74		
76	*α*-Gurjunene	$C_{15}H_{24}$	0.12	–	3.29
77	(-)-*γ*-Cadinene	$C_{15}H_{24}$	0.06		
78	*δ*-Cadinene	$C_{15}H_{24}$	0.19	0.17	0.36
79	Naphthalene,1,2,4a,5,6,8a-hexahydro-4,7-dimethyl-1-(1-methylethyl)-	$C_{15}H_{24}$	–	0.66	–
80	1-tert-Butyl-3,5-dimethylbenzene	$C_{12}H_{18}$	–	–	1.00
81	Alloaromadendrene	$C_{15}H_{24}$	–	–	0.79
82	Eudesma-3,7(11)-diene	$C_{15}H_{24}$	–	–	0.42
83	*α*-Farnesene	$C_{15}H_{24}$	–	0.28	–
84	Neoisolongifolene	$C_{15}H_{24}$	–	–	0.29
85	Calarene	$C_{15}H_{24}$	–	–	0.81
86	*α*-Elemol	$C_{15}H_{26}O$	0.21	–	–
87	Peruviol	$C_{15}H_{26}O$	0.35	1.02	–
88	Eudesma-3,7(11)-diene	$C_{15}H_{24}$	0.20	–	–
89	*β*-Neoclovene	$C_{15}H_{24}$	–	–	0.63
90	Caryophyllenyl alcohol	$C_{15}H_{26}O$	–	–	3.26
91	*β*-Patchoulene	$C_{15}H_{24}$	–	–	0.52
92	Espatulenol	$C_{15}H_{24}O$	0.43	–	–
93	Aromadendrene oxide-(1)	$C_{15}H_{24}O$	–	–	0.88
94	Caryophyllene oxide	$C_{15}H_{24}O$	–	4.63	–
95	6-Isopropyl-4,8*α*-dimethyl-1,2,3,7,8,8*α*-hexahydronaphthalene	$C_{15}H_{24}$	–	–	3.45
96	*β*-Vatirenene	$C_{15}H_{24}$	–	–	0.52
97	10s,11s-Himachala-3(12),4-diene	$C_{15}H_{24}$	–	0.40	2.01
98	Cycloheptane, 4-methylene-1-methyl-2-(2-methyl-1-propen-1-yl)-1-vinyl-	$C_{15}H_{24}$	–	–	0.91
99	1,5,5,8-Tetramethyl-3,7-cycloundecadien-1-ol	$C_{15}H_{26}O$	–	5.83	–
100	(-)-Globulol	$C_{15}H_{26}O$	0.70	–	–
101	(+)-Viridiflorol	$C_{15}H_{26}O$	0.38	–	–
102	(+)-*α*-Elemene	$C_{15}H_{24}$	–	0.97	3.01
103	5-Cyclohexyl-1-pentene	$C_{11}H_{20}$	–	–	0.92
104	Tetradecanal	$C_{14}H_{28}O$	0.13	–	–
105	*o*-Menth-8-ene	$C_{10}H_{18}$	–	4.42	–
106	Machilol	$C_{15}H_{26}O$	0.24	–	–
107	*γ*-Maaliene	$C_{15}H_{24}$	–	–	0.84
108	Spathulenol	$C_{15}H_{24}O$	0.28	–	–
109	*β*-Guaiene	$C_{15}H_{24}$	–	2.35	10.01
110	*β*-Eudesmol	$C_{15}H_{26}O$	0.16	–	–
111	*α*-Eudesmol	$C_{15}H_{26}O$	0.28	–	–
112	*γ*-Eudesmol	$C_{15}H_{26}O$	–	–	2.11
113	Isocamphane	$C_{10}H_{18}$	–	1.62	–
114	Naphthalene, 1,2,4a,5,8,8a-hexahyd ro-4,7-dimethyl-1-(1-methylethyl)- , (1*α*,4a *β*,8a*α*)-(. +/-.)-	$C_{15}H_{24}$	–	–	2.29
115	Eudesm-7(11)-en-4-ol	$C_{15}H_{26}O$	–	1.46	–
116	1*H*-Cyclopropa[a]naphthalene, decahydro-1,1,3a-trimethyl-7-methylene-, [1a*S*-(1a*α*,3a*α*,7a*β*,7b*α*)]-	$C_{15}H_{24}$	–	–	1.52
117	Longifolene	$C_{15}H_{24}$	–	1.34	0.44
118	Nerolidol	$C_{15}H_{26}O$	–	0.57	–
119	(E)-*β*-Farnesene	$C_{15}H_{24}$	–	–	0.46
120	Phytol	$C_{20}H_{40}O$	0.13	–	–

a: not detected (< 0.1%)

been commonly used for the identification of species level. In this study, the primers pair ITS 1 and ITS 4 and NS 1 and NS 8 were selected to amplify the internal transcribed spacer region of the ribosomal RNA. Unfortunately, the sequence based on the primers ITS 1 and ITS 4 was not successfully acquired, consistent with a literature report (Jiang and Kirschner 2016). Jiang and Kirschner (2016) reported that powdery-fruit disease of *C. burmannii* was caused by the fungus *C. cinnamomi* based on the morphological characters and partial nuclear ribosomal large subunit RNA (LSU) gene sequencing data. In our work, the pathogen CBd1was shown to maintain 99.63% sequence similarity to that of *Acaromyces ingoldii* (accession number NG061199 and KP866248) according to the morphological characteristics and 18S rDNA (NS) gene

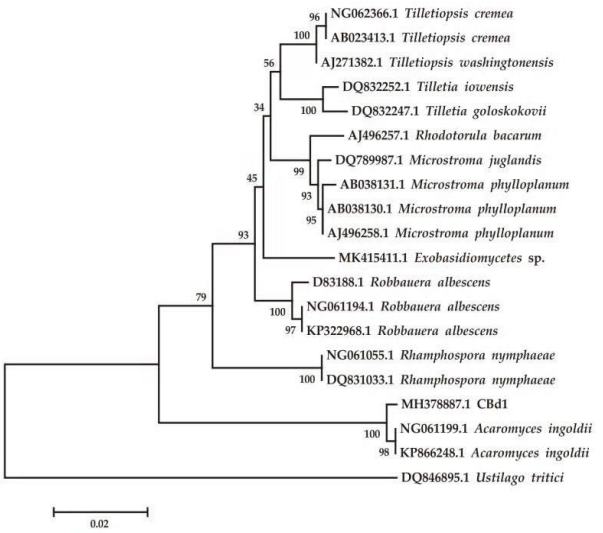

Fig. 3: Molecular phylogenetic analysis of pathogen CBd1 with nineteen strains obtained from GenBank. The tree was constructed by the neighbor-joining method based on 18S rDNA (NS) sequences. Bootstrap values after 1000 replicates were expressed as percentages at branching points

sequence. Multigene sequencing will be necessary to accurately identify the species in the future.

Essential oils are complicated mixtures of natural products consisting of aromatic volatile components such as hydrocarbons, lipids, phenols, terpenes, ketones and their derivatives, showing various biological activities such as anti-inflammatory, antimicrobial, anti-proliferative and insecticidal properties (Taga *et al.* 2012; Sharifi-Rad *et al.* 2017). There have been many reports about the composition of essential oils from the leaves and bark of *C. burmannii*. The chemical profile of essential oil from the leaves of *C. burmannii* growing in Kunming (China) was mainly composed of linalool (54.93%) (Ding *et al.* 1994). Interestingly, trans-cinnamaldehyde (60.17%), eugenol (17.62%), coumarin (13.39%) and borneol (6.79%) were the major volatile components in the leaf oil of *C. burmannii* collected from Guangzhou, China (Wang *et al.* 2009). Thirty-three compounds were obtained from the leaf essential oil of *C. burmannii* by supercritical CO_2 extraction technology, and among them, borneol (47.23%) was the predominant volatile component (Li *et al*. 2016). In this study, we found that borneol (24.99%), D-limonene (11.16%) and L-(-)-borneol (10.51%) were the major components with borneol being the most abundant compound in the leaf oil. Overall, borneol existed in all the samples, while the relative amount of this component varied greatly. Such variability in different specimens could be related to geographical locations. In addition, the chemical compositions also varied by a large extent in different plant organs. For example, coumarin was the most abundant compound in the *C. burmannii* bark, while borneol was the predominant compound in the leaves (Wang *et al.* 2013). The bark oil of *C. burmannii* from Guangxi (China) was rich in eucalyptol (30.93%) and borneol (18.31%) (Li *et al.* 2015). To the best of our knowledge, there is no report analyzing the fruit essential oils of *C. burmannii*, and only one study has shown that eucalyptol, *α*-terpineol, *α*-phellandrene, *D-limonene*, *α*-pinene and *β*-pinene were the main components in the fruit oil of *C. migao*, which is in the same genus as *C. burmannii* (Zhang *et al.* 2011). We found that *α*-caryophyllene (32.94%) and *l*-caryophyllene (17.75%) were the main components, with a negligible amount of *D*-limonene (0.21%) in normal fruits. In contrast, a lower content of α-pinene (5.48%) and a higher content of *D*-limonene (2.10%) were found in the infected fruits. Overall, the chemical composition of essential oils not only correlates to plant species and organs, but also to the growth environment and extraction methods and techniques. These factors are responsible for the deviations in the oil components in terms of quality and quantity (Wang *et al.* 2019). It should be noted that the chemical composition and percentage differences between the normal and infected fruits may relate to the production of phytotoxic substances by the pathogen (Shan *et al*. 2012; Sun *et al*. 2017; Meng *et al*. 2019). The potential chemical structures and biological activities of the phytotoxins due to pathogen infection will be studied in the future.

Conclusion

The 18S rDNA (NS) gene sequence of the pathogen CBd1 (accession number MH378887) exhibited 99.63% similarity to that of *Acaromyces ingoldii* (accession numbers NG061199 and KP866248). The compositions and ratios of the essential oils from normal and infected fruits of *C. burmannii* varied greatly. The differences could be attributed to the pathogen infection in the fruits of *C. burmannii*, although the mechanism remains unknown, which will require further exploration.

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