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Phylogenetic analysis of major molds inhabiting woods and their discoloration characteristics. Part 2. Genus *Penicillium*

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Abstract

In this study, the diversity of wood inhabiting *Penicillium* species in Korea was investigated through the analysis of their phenotypic traits and phylogeny. Moreover, the discoloration and growth patterns of these species on the surfaces of two commercially important conifers were examined. From a collection of 137 *Penicillium* isolates from various wood species, 37 *Penicillium* species have been identified including six unknown *Penicillium* species. The occurrence of six *Penicillium* species in Korea was demonstrated for the first time: *P. biourgeianum*, *P. decaturense*, *P. kloeckeri*, *P. meleagrinum* var. *viridiflavum*, *P. ochrochloron*, and *P. sumatrense*. *P. funiculosum* was the most prevalent species (15.4%). Sapwood blocks of Japanese red pine (*Pinus densiflora*) and radiata pine (*Pinus radiata*) were exposed to each *Penicillium* species to examine their discoloration and growth patterns on these woods. The blocks of Japanese red pine were more susceptible to *Penicillium* growth, and the highest discoloration rate on both sapwood blocks was achieved by one of the recently described species *P. decaturense*.

Keywords: β-tubulin; internal transcribed spacer (ITS); large subunit (LSU); mold; *Penicillium*; phylogeny; wood discoloration.

Introduction

Forests play an important role in ecosystems by providing habitat for numerous organisms depending on it. This is also true for wood as the main biomass produced in forest (Stevens 1997). Wood is an organic material mainly consisting of cellulose, hemicelluloses, and lignin (Panshin and De Zeeuw 1980). The supramolecular architecture of these compounds was reviewed recently by Salmén and Burgert (2008). Molds and stain fungi colonize sapwood of fresh fallen dead wood rapidly (Robbins and Morrell 2002). Stored sugars in living cells and other nutrients on the wood surface are helpful in this regard (Carll and Highley 1999). Molds derogate the appearance of wood and lower its economic value. Moreover, they also adversely affect human health (Bush et al. 2006).

Penicillium Link, a genus of deuteromycetous fungi with more than 225 recognized species (Samson and Pitt 2000), is one of the most well-known molds. It is commonly isolated from forest woods (Crawford et al. 1990), and many studies describe it as one of the most frequently encountered genera in sawmills (Halpin et al. 1994; Simeray et al. 1997; Sivrikaya and Kara 2009).

Currently, there are more than 60 *Penicillium* species in Korea (Kim et al. 2007), most of which are described as soil-inhabiting fungi, and some as plant pathogenic fungi. Usually, they are identified only by their phenotypic characteristics. Little is known about the diversity of wood inhabiting species and it is not known how fast they grow, even though prevention of mold growth by fungicides has been extensively studied (Kim et al. 2002).

In this study, the diversity of *Penicillium* species inhabiting woods has been investigated by means of phenotypic and phylogenetic analysis. The discoloration and colonization patterns typical for these molds were also measured on two commercially important conifers.

Materials and methods

Fungal isolation

Investigated trees included Douglas-fir (*Pseudotsuga menziesii*), Japanese red pine (*Pinus densiflora*), Korean pine (*Pinus koraiensis*), larch (*Larix kaempferi*), Norway spruce (*Picea abies*), and radiata pine (*Pinus radiata*). The stems were taken from sawmills, log yards, and from the beams of cooling towers, including chromated copper arsenate (CCA)- and creosote-treated woods in Korea in the time range between 2000 and 2007 (Table 1). Fungi were isolated and purified as described by Huh et al. (2010).

Cultures

Spore suspensions obtained from each isolate were inoculated in three points onto malt extract agar (MEA, malt extract 20 g, peptone 1 g, glucose 20 g, agar 15 g, distilled water 1000 ml; pH 5.5), 25% glycerol nitrate agar (G25N, glycerol, analytical grade 250 g, $NaNO_3$ 3 g, K_2HPO_4 1 g, KCl 0.5 g, $MgSO_4 \cdot 7\ H_2O$ 0.5 g, $FeSO_4 \cdot 7\ H_2O$ 0.01 g, yeast extract 5 g, agar 15 g, distilled water 750 ml; pH 6.5) and Czapek yeast autolysate agar (CYA, $NaNO_3$ 3 g, K_2HPO_4 1 g, KCl 0.5 g, $MgSO_4 \cdot 7\ H_2O$ 0.5 g, $FeSO_4 \cdot 7\ H_2O$ 0.01 g, sucrose 30 g, yeast extract 5 g, agar 15 g, distilled water 1000 ml; pH 6.5) in 9 cm Petri dishes and incubated at 25°C for 7 days. Phenotypes were examined following the method described by Pitt (1979). All isolates were preserved in 20% glycerol at -80°C as well as in sterile

Table 1 *Penicillium* strains isolated from woods (identified based on ITS, LSU rDNA, and β-tubulin sequences and on their phenotypic features) and their discoloration rate on woods of Japanese pine (JP) and radiata pine (RP).

Isolate number[a]	GenBank acc. no.[b]	No. of isolates (% frequency)[c]	Source[d]	Sample location[c]	Closet fungal match (acc. no.)[f]	S (%)[g]	Fungus	Discoloration rate[h]	
								JP	RP
								7d/14d/21d	7d/14d/21d
KUC1432	HM469395	1 (0.7)	JP (1)	BH (1)	*P. biourgeianum* (AY484911)	100.0	*P. biourgeianum*	1/2/4	1/1/2
KUC1433	HM469396	2 (1.5)	JP (1), RP (1)	BH (1), BS (1)	*P. brasilianum* (AB455514)	99.6	*P. brasilianum*	5/5/5	3/3/4
KUC1628-1	HM469408	2 (1.5)	LA (1), RP (1)	IC (1), YJ (1)	*P. brevicompactum* (EF634407)	99.8	*P. brevicompactum*	3/3/5	1/1/1
KUC3084	HM469428	2 (1.5)	CCALA (1), CTW (1)	GM (1), SU (1)	*P. citrinum* (EF634428)	100.0	*P. citrinum*	4/5/5	1/1/1
KUC3029	HM469426	1 (0.7)	CCARP (1)	IC (1)	*P. commune* (EF200099)	100.0	*P. commune*	4/5/5	2/3/3
KUC3019	HM469424	7 (5.1)	CCARP (1), KP (4), RP (2)	IC (2), IS (1), GP (4)	*P. crustosum* (EF634415)	100.0	*P. crustosum*	5/5/5	3/3/4
KUC1522	HM469399	4 (2.9)	KP (4)	GP (4)	*P. decaturense* (AF125946)	100.0	*P. decaturense*	5/5/5	4/5/5
KUC1284	HM469392	6 (4.4)	JP (5), RP (1)	BH (5), IC (1)	*P. diversum* (DQ308554)	100.0	*P. diversum*	5/5/5	3/4/4
KUC1909	HM469423	1 (0.7)	DF (1)	YI (1)	*P. expansum* (DQ339556)	100.0	*P. expansum*	1/3/4	1/2/2
KUC3020	HM469425	8 (5.8)	CCARP (1), JP (1), LA (2), NS (1), RP (3)	BH (1), IC (4), IS (1), YJ (2)	*P. fellutanum* (EF200082)	100.0	*P. fellutanum*	5/5/5	2/3/5
KUC3055	HM469427	21 (15.3)	CCARP (1), JP (5), KP (11), LA (2), RP (2)	BH (5), BS (1), GP (1), IC (1), IS (1), YJ (12)	*P. funiculosum* (GQ221866)	100.0	*P. funiculosum*	4/5/5	3/4/4
KUC1553	HM469402	15 (10.9)	JP (4), KP (1), LA (3), RP (7)	BH (4), BS (3), GP (1), IC (3), IS (1), YJ (3)	*P. glabrum* (AF033407)	100.0	*P. glabrum*	4/5/5	2/3/4
KUC1619-2	HM469406	1 (0.7)	LA (1)	YJ (1)	*P. implicatum* (AF033428)	100.0	*P. implicatum*	2/4/4	2/3/3
KUC5018	HM469429	1 (0.7)	CTW (1)	GM (1)	*P. janthinellum* (AY373921)	100.0	*P. janthinellum*	4/5/5	2/3/3
KUC1286	HM469393	2 (1.5)	JP (1), LA (1)	BH (1), YJ (1)	*Talaromyces wortmannii* (AY533693)	100.0	*P. kloeckeri*	3/5/5	2/3/3
KUC1678	HM469412 *HM469411*	8 (5.8)	JP (5), KP (2), LA (1)	BH (5), GP (1), YJ (2)	*P. meleagrinum* var. *viridiflavum* (EF198531/*EF198510*)	100.0 *100.0*	*P. meleagrinum* var. *viridiflavum*	3/5/5	1/1/2
KUC1551	HM469400	2 (1.5)	JP (1), KP (1)	BH (1), GP (1)	*P. miczynskii* (AF033416)	99.3	*P. miczynskii*	4/4/5	2/2/2
KUC1680	HM469414	13 (9.5)	CCAKP (1), CCALA (1), JP (6), KP (3), LA (2)	BH (6), GP (2), SU (2), YJ (3)	*P. minioluteum* (PECRR58SC)	99.8	*P. minioluteum*	3/5/5	1/2/2
KUC1626	HM469407	1 (0.7)	LA (1)	YJ (1)	*P. multicolor* (EU427298)	99.6	*P. multicolor*	4/5/5	3/3/5
KUC1348-1	HM469394	2 (1.5)	JP (1), KP (1)	BH (1), YJ (1)	*P. ochrochloron* (AF033441)	99.8	*P. ochrochloron*	4/5/5	1/1/1
KUC1674	HM469410	2 (1.5)	CCALA (1), KP (1)	GP (1), SU (1)	*P. oxalicum* (DQ123663)	100.0	*P. oxalicum*	3/4/5	2/2/2
KUC1758	HM469418	1 (0.7)	RP (1)	IS (1)	*P. pinophilum* (GU566216)	100.0	*P. pinophilum*	1/3/4	0/1/2
KUC1788	HM469419	2 (1.5)	CCARP (1), RP (1)	BS (1), IC (1)	*P. purpurescens* (AF033408)	100.0	*P. purpurascens*	2/2/4	1/1/2
KUC1679	HM469413	8 (5.8)	JP (3), RP (5)	BH (3), BS (2), GP (1), IC (2)	*P. radicum* (EU262660)	100.0	*P. radicum*	0/1/3	0/0/1
KUC1729	HM469416	1 (0.7)	JP (1)	BH (1)	*P. raistrickii* (AF033491)	99.6	*P. raistrickii*	1/1/4	1/1/1
KUC5153	HM469430	3 (2.2)	JP (1), NS (1), RP (1)	BH (1), IC (2)	*P. simplicissimum* (DQ026013)	100.0	*P. simplicissimum*	5/5/5	3/3/4
KUC1617	HM469405	1 (0.7)	LA (1)	YJ (1)	*P. spinulosum* (AF033410)	100.0	*P. spinulosum*	3/4/4	1/3/3
KUC1681-1	HM469415	2 (1.5)	KP (1), LA (1)	GP (1), YJ (1)	*P. steckii* (EF634431)	100.0	*P. steckii*	3/5/5	2/2/3
KUC1613	HM469404 *HM469403*	2 (1.5)	LA (2)	YJ (2)	*P. sumatrense* (AF033424/*EF198503*)	99.8 *98.6*	*P. sumatrense*	3/5/5	2/3/4
KUC1476	HM469398	2 (1.5)	JP (1), RP (1)	BH (1), IS (1)	*P. variabile* (AY373936)	99.6	*P. variabile*	4/5/5	2/3/4
KUC1794	HM469420	2 (1.5)	KP (1), RP (1)	BS (1), YJ (1)	*P. verruculosum* (AF510496)	98.9	*P. verruculosum*	1/4/4	2/3/4
KUC1434	HM469397	3 (2.2)	JP (3)	BH (3)	*P. citreonigrum* (AY157489)	98.7	*Penicillium* species 1	2/3/4	3/3/4
KUC1552	HM469401	2 (1.5)	KP (2)	GP (1), YJ (1)	*P. purpurogenum* (DQ681328)	97.8	*Penicillium* species 2	2/3/4	1/1/2

Table 1 (Continued)

Isolate number[a]	GenBank acc. no.[b]	No. of isolates (% frequency)[c]	Source[d]	Sample location[e]	Closet fungal match (acc. no.)[f]	S (%)[g]	Fungus	Discoloration rate[h] JP 7d/14d/21d	Discoloration rate[h] RP 7d/14d/21d
KUC1795-3	HM469421	1 (0.7)	RP (1)	BS (1)	*P. sclerotiorum* (AF033404)	96.3	*Penicillium* species 3	3/5/5	2/4/4
KUC1907	HM469422	1 (0.7)	DF (1)	YI (1)	*P. verruculosum* (AF510496)	96.3	*Penicillium* species 4	2/4/4	0/1/3
KUC1754	HM469417	3 (2.2)	RP (3)	IS (3)	*P. brasilianum* (AB455514)	98.9	*Penicillium* species 5	2/3/4	2/3/4
KUC1651	HM469409	1 (0.7)	RP (1)	IC (1)	*P. simplicissimum* (AF203084)	98.9	*Penicillium* species 6	2/3/4	2/3/4

[a]KUC: Korea University Culture Collection, Seoul, Korea.
[b]Accession numbers of rDNA (ITS and LSU) sequences are in regular font and β-tubulin gene sequences are in italic font.
[c]Frequency=(number of isolates/total isolates)×100.
[d]CCAKP, CCA-treated Korean pine; CCALA, CCA-treated larch; CCARP, CCA-treated radiata pine; CTW, creosote-treated wood; DF, Douglas-fir; JP, Japanese red pine; KP, Korean pine; LA, larch; NS, Norway spruce; RP, radiata pine.
[e]BH, Bonghwa; BS, Busan; GP, Gapyeong; GM, Gwangmyeong; IS, Iksan; IC, Incheon; SU, Seoul; YI, Yongin; YJ, Yeoju.
[f]Accession numbers of ITS sequences are in regular font and β-tubulin gene sequences are in italic font.
[g]Similarity scores from pairwise alignments of sample sequences with closest BLAST match or reference strains; similarity of ITS sequences are in regular font and similarity of β-tubulin sequences are in italic font.
[h]Discoloration rate: 0 to 5, with 5 corresponding to the highest growth rate and largest discolored area; JP, Japanese red pine; RP, radiata pine.

water at 4°C in Korea University Culture Collection (KUC), Korea University, Korea.

DNA extraction, PCR, DNA sequencing, and phylogenetic analysis

DNA extractions are described in detail in Part 1 of this publication (Huh et al. 2010). The nuclear internal transcribed spacer (ITS), including 5.8S ribosomal DNA (rDNA), as well as a phylogenetically informative segment of the nuclear ribosomal large subunit (LSU) rDNA, were amplified with primers ITS1F (5′-CTTGGTCATTTAGAGGAAGTAA-3′) and LR5 (5′-TCCTGAGGGAAACTTCG-3′) or ITS1F/LR3 (5′-CCGTGTTTCAAGACGGG-3′) primer set (Hopple and Vilgalys 1994) by means of the *Accupower* PCR premix kit (Bioneer, Seoul, Korea). When identification of fungi with ITS and LSU rDNA was not possible, the partial β-tubulin gene was also amplified with Bt2a (5′-GGTAACCAAATCGGTGCTGCTTTC-3′) and Bt2b (5′-ACCCTCAGTGTAGTGACCCTTGGC-3′) primer set (Glass and Donaldson 1995) using the Takara ex Taq polymerase (Takara, Otsu, Shiga, Japan). PCR reactions were carried out in a Bio-Rad MyCycler (Bio-Rad, Hercules, CA, USA). PCR cycling conditions of the ITS and LSU rDNA regions were as follows: an initial denaturation step of 95°C for 7 min, followed by 30 cycles of 95°C for 40 s, 51°C for 40 s, and 72°C for 1 min 20 s. An elongation step of 72°C for 7 min was performed in the end. The amplification of the partial β-tubulin gene followed the conditions described by Samson et al. (2004). PCR amplicons were detected, purified, and sequenced as described by Huh et al. (2010). The amplicons of the Bt2a/Bt2b, ITS1F/LR3, and ITS1F/LR5 primers were approximately 400, 1100, and 1400 bp, respectively. The sequences obtained in this study were deposited in GenBank under accession numbers HM469392–HM469430 (Table 1). The procedures for phylogenetic analyses are also described in detail in Part 1 of this publication (Huh et al. 2010).

Wood discoloration characteristics

For the discoloration test, fresh sapwood blocks of two commercially important conifers, Japanese red pine and radiata pine, were prepared as described by Huh et al. (2010).

Penicillium strains grown on MEA were inoculated on both ends of the wood block. They were incubated for 3 weeks at 25°C and the discoloration degrees were measured every 7 days, being scored from 0 to 5 based on the intensity of growth and discolored area of the wood blocks. Discoloration characteristics such as colonizing patterns and color changes were also observed.

Results and discussion

Fungal identification and phylogenetic analysis

A total of 137 isolates of *Penicillium* were collected during the investigation of six wood species. The 37 species were identified by examining their morphological and molecular traits. Six unknown *Penicillium* species (Table 1) were also found, referred to here as species 1–6. *P. funiculosum* (15.4%) was the most prevalent, followed by *P. glabrum* (10.9%), and *P. minioluteum* (9.5%). Together, these three species represent more than one-third of the total number of isolates collected. Lee et al. (2003) reported that *P. funiculosum* and *P. glabrum* were commonly found in forest and

paddy field soil in Korea. Kim (2005) isolated *P. funiculosum* and *P. minioluteum* as plastic degrading fungi.

Most species identified in this study have been previously described as soil-inhabiting fungi except *P. fellutanum*, which was isolated from pitch pine (*Pinus rigida*) as an endophyte (Kim et al. 2008), and *P. citrinum*, *P. crustosum*, *P. expansum*, and *P. oxalicum*, which have been reported as causing plant diseases (The Korean Society of Plant Pathology 2009). Interestingly, *P. biourgeianum*, *P. decaturense*, *P. kloeckeri*, *P. meleagrinum* var. *viridiflavum*, *P. ochrochloron*, and *P. sumatrense* had not been previously reported in Korea.

The distribution of *Penicillium* might also be influenced by the specificity of the species with their hosts in a special geographic region. For example in Bonghwa, *P. biourgeianum* (0.7%) was found exclusively on Japanese red pine. Similarly, *P. decaturense* (2.9%) and *P. sumatrense* (1.5%) were isolated only from Korean pine in Gapyeong and from larch in Yeoju, respectively. By contrast, *P. citrinum*, *P. commune*, *P. crustosum*, *P. fellutanum*, *P. funiculosum*, *P. janthinellum*, *P. minioluteum*, *P. oxalicum*, and *P. purpurascens* were found on treated woods. Among them, *P. citrinum* and *P. commune* were found exclusively on woods treated with both CCA and/or creosote, indicating that these two species might be more resistant to wood preservatives than the other *Penicillium* species studied.

The evolutionary relationships within this genus as inferred from ITS sequences are depicted in Figure 1a. The phylogenetic resolution was generally strong and most *Penicillium* species formed distinct clades with strong or moderate maximum parsimony bootstrap proportions (MPBP)/ neighbor-joining bootstrap proportions (NJBP) values. However, among the sequences examined, *P. meleagrinum* var. *viridiflavum* and *P. sumatrense* were clustered in the same clade (MPBP = 94%, NJBP = 100%), but their phylogenies were resolved in a study involving other loci as proposed by Serra et al. (2008). As shown in Figure 1b, these two *Penicillium* species were identified by β-tubulin gene sequences.

Regarding the unknown species, *Penicillium* species 1 KUC1434 and *P. citreonigrum* CBS 454.93 were strongly clustered in the same clade (MPBP = 99%, NJBP = 100%). However, they had a similarity of 98.7% and the LSU region sequence from *Penicillium* species 1 KUC1434 did not match to any of *P. citreonigrum* sequences available in GenBank. *Penicillium* species 2 KUC1552 was grouped with *P. purpurogenum* (MPBP = 100%, NJBP = 100%), but they are readily distinguishable by the presence of purple pigment. *Penicillium* species 3 KUC1795-3 was in the same clade as *P. sclerotiorum* NRRL 2074 (MPBP = 66%, NJBP = 51%), but their similarity was rather low (96.3%). *Penicillium* species 4 KUC1907 also showed a low similarity value (96.3%) with *P. verruculosum* ATCC 62396. *Penicillium* species 5 KUC1754 and *Penicillium* species 6 KUC1651 were most closely matched to *P. brasilianum* FKI-3368 (similarity of 98.9% in the ITS region) and to *P. simplicissimum* IBT 15303 (similarity of 98.9%, ITS region), respectively, but their LSU region sequences were identical and most closely matched to *P. skrjabinii* (99.8%). Identification of these unknown *Penicillium* species will therefore require a thorough investigation with type strains and additional loci.

Wood discoloration characteristics

Sapwood blocks of Japanese red pine and radiata pine were inoculated with each of the 37 *Penicillium* species identified (Table 1). These two species exhibited a different degree of susceptibility to each *Penicillium* species, and vice versa the *Penicillium* species also differed in their ability to attack the wood specimens. Overall, blocks of Japanese red pine were more susceptible to *Penicillium* growth than blocks of radiata pine. Within 7 days, approximately half of the *Penicillium* species showed a discoloration score higher than 3 on Japanese red pine blocks, whereas only *P. decaturense* KUC1522 reached the same score on blocks of radiata pine. The entire surface area of Japanese red pine blocks was occupied after 21 days by most *Penicillium* species, but only three species – *P. decaturense* KUC1522, *P. fellutanum* KUC3020, and *P. multicolor* KUC1626 – achieved a discoloration rate of 5 on radiata pine within the same period. These results might be due to the different abundance of low-molecular weight sugars or nitrogenous compounds in the sapwoods (Terziev and Nilsson 1999), which means that Japanese red pine might be rich in those nutrients compared to radiata pine.

In terms of the discoloration patterns, most *Penicillium* species showed lumpy or sparse patterns with greenish or brownish colors on woods. However, some species also exhibited different patterns. For example, *P. minioluteum* KUC1680 discolored the sapwood specimens sparsely into pinkish, whereas *P. radicum* KUC1679 and *P. raistrickii* KUC1729 showed no obvious spore colors due to little sporulation. Interestingly, *P. multicolor* KUC1626 not only discolored the blocks greenish by spores but also stained them yellowish by soluble pigment. Similarly, *P. verruculosum* KUC1794 stained the wood blocks into a brownish tone.

The three predominant species, *P. funiculosum*, *P. glabrum*, and *P. minioluteum*, exhibited a high discoloration rate on Japanese red pine. Their growth rate was similar and the maximum discoloration degree was achieved after 7–14 days. The highest discoloration rate on radiata pine was that of *P. funiculosum*, followed by *P. glabrum* and *P. minioluteum*. Of all species, the newly recorded *P. decaturense* KUC1522 had the highest discoloration rate on both types of sapwood blocks, covering the entire surface of the sapwood blocks and discoloring them in 7 days. The other newly recorded species showed relatively low discoloration rates on radiata pine. Consequently, we suggest that the most abundant species as well as the species showing the highest discoloration rate should be considered as target mold species by future studies to prevent mold growth on woods.

Conclusions

In this study, we describe the diversity of *Penicillium* species inhabiting woods and their discoloration patterns on commercially important wood species. Among the 37 *Penicillium*

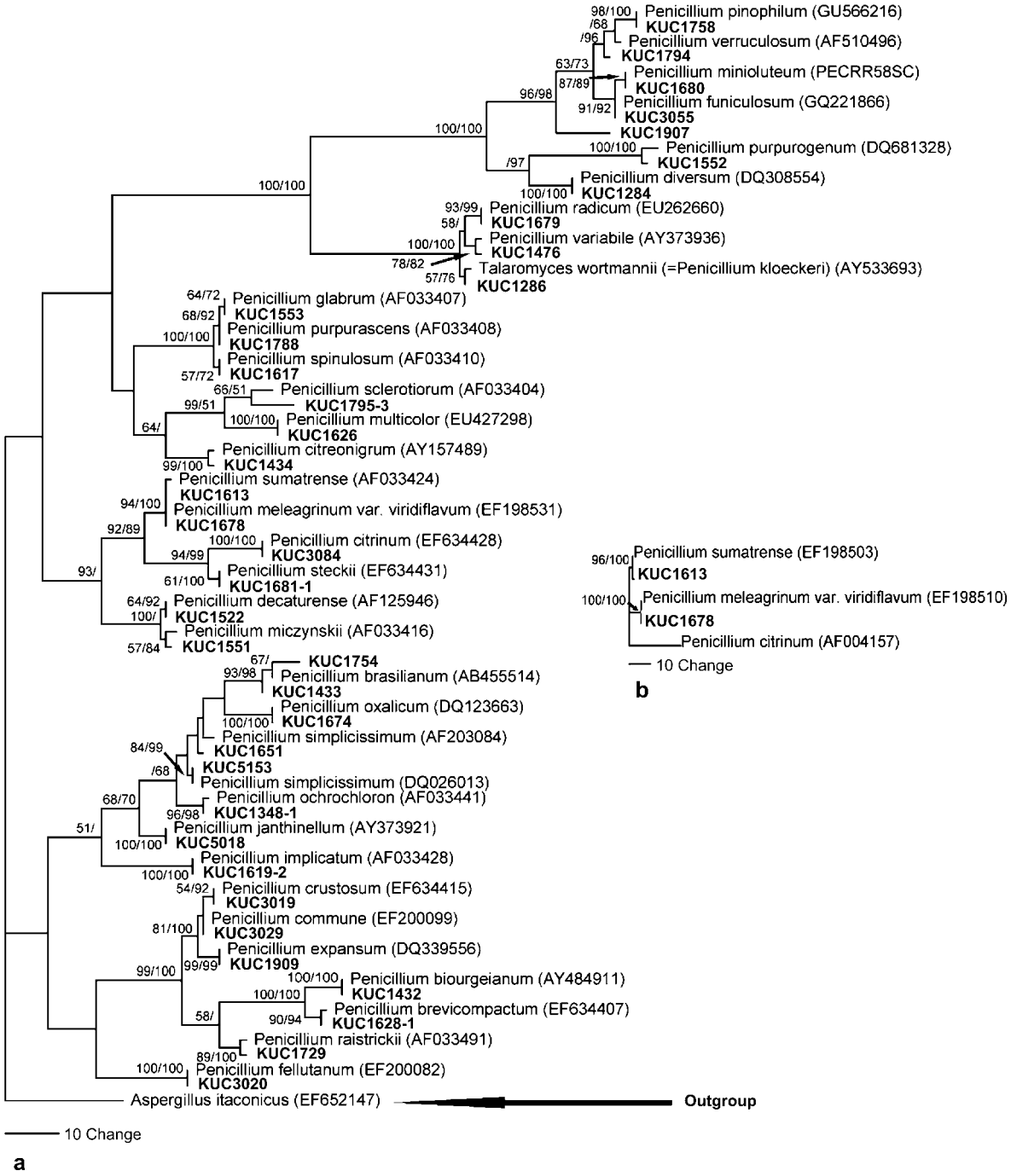

Figure 1 (a) Maximum parsimonious tree based on ITS region sequences. The dataset comprised 528 characters, of which 329 were constant, 12 were parsimony uninformative, and 187 were parsimony informative. The tree length was 565 steps with consistency index (CI) = 0.5611 and retention index (RI) = 0.9005. (b) Maximum parsimonious tree based on the β-tubulin region. The dataset comprised 389 characters, of which 330 were constant, 48 were parsimony uninformative, and 11 were parsimony informative. Tree length was 64 steps with CI = 1.0000 and RI = 1.0000. Bootstrap values are shown above branches (MP bootstrap proportions/NJ bootstrap proportions). Strains found in this study are in bold font. References taken from GenBank are in regular font. Genetic distances between strains are represented by branch length and accession numbers are indicated in parentheses.

species identified, six species had their first record of occurrence in Korea and six other species were previously unknown. Further studies with additional loci should help understand their relative positions in the phylogeny of the group.

As a matter of fact, similar to *Trichoderma* species described in Part 1 of this publication (Huh et al. 2010), the species of *Penicillium* are found worldwide and most of these species dominantly cause damage in damp building materials, including logs and lumber products.

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References

Bush, R.K., Portnoy, J.M., Saxon, A., Terr, A.I., Wood, R.A. (2006) The medical effects of mold exposure. J. Allergy Clin. Immunol. 117:326–333.

Carll, C., Highley, T. (1999) Decay of wood and wood-based products above ground in buildings. J. Test. Eval. 27:150–158.

Crawford, R., Carpenter, S., Harmon, M. (1990) Communities of filamentous fungi and yeast in decomposing logs of *Pseudotsuga menziesii*. Mycologia 82:759–765.

Glass, N., Donaldson, G. (1995) Development of primer sets designed for use with the PCR to amplify conserved genes from filamentous ascomycetes. Appl. Environ. Microbiol. 61:1323–1330.

Halpin, D., Graneek, B., Lacey, J., Nieuwenhuijsen, M., Williamson, P., Venables, K., Newman Taylor, A. (1994) Respiratory symptoms, immunological responses, and aeroallergen concentrations at a sawmill. Occup. Environ. Med. 51:165–172.

Hopple, J. Jr., Vilgalys, R. (1994) Phylogenetic relationships among coprinoid taxa and allies based on data from restriction site mapping of nuclear rDNA. Mycologia 86:96–107.

Huh, N., Jang, Y., Lee, J., Kim, G.-H., Kim, J.-J. (2010) Phylogenetic analysis of major molds inhabiting woods and their discoloration characteristics. Part 1. Genus *Trichoderma*. Holzforschung: 65:257–263.

Kim, M.N. (2005) Morphological characteristics and identification of plastic degrading fungi. J. Natural Sci. 15:1–9.

Kim, J.-J., Ra, J.-B., Kim, H.-J., Kim, K.-H. (2002) Sapstain and mold control on radiata pine lumber – laboratory and field tests of selected fungicides. Mycobiology 30:37–40.

Kim, W.K., Sang, H.K., Woo, S.K., Park, M.S., Narayan Chandra, P., Yu, S.H. (2007) Six species of *Penicillium* associated with blue mold of grape. Mycobiology 35:180–185.

Kim, C.S., Park, M.S., Yu, S.H. (2008) Two species of endophytic *Penicillium* from *Pinus rigida* in Korea. Mycobiology 36:222–227.

Lee, S., Hong, S.-B., Kim, C.-Y. (2003) Contribution to the checklist of soil-inhabiting fungi in Korea. Mycobiology 31:9–18.

Panshin, A., De Zeeuw, C. Textbook of Wood Technology: Structure, Identification, Properties, and Uses of the Commercial Woods of the United States and Canada. McGraw-Hill Book Co., New York, 1980.

Pitt, J. The Genus *Penicillium* and its Teleomorphic States *Eupenicillium* and *Talaromyces.* Academic Press, London, 1979.

Robbins, C., Morrell, J. Mold, Housing, and Wood. Western Wood Products Association, Portland, OR, 2002.

Salmén, L., Burgert, I. (2008). Cell wall features with regard to mechanical performance. A review. COST Action E35 2004–2008: Wood machining – micromechanics and fracture. Holzforschung 63:121–129.

Samson, R., Pitt, J. Integration of Modern Taxonomic Methods for *Penicillium* and *Aspergillus* Classification. Harwood Academic Publishers, Amsterdam, 2000.

Samson, R.A., Seifert, K.A., Kuijpers, A.F.A., Houbraken, J., Frisvad, J.C. (2004) Phylogenetic analysis of *Penicillium* subgenus *Penicillium* using partial β-tubulin sequences. Stud. Mycol. 49:175–200.

Serra, R., Peterson, S., Venancio, A. (2008) Multilocus sequence identification of *Penicillium* species in cork bark during plank preparation for the manufacture of stoppers. Res. Microbiol. 159:178–186.

Simeray, J., Mandin, D., Chaumont, J. (1997) An aeromycological study of sawmills: effects of type of installation and timber on mycoflora and inhalation hazards for workers. Int. Biodeterior. Biodegrad. 40:11–17.

Sivrikaya, H., Kara, O. (2009) Airborne fungi in wood and wood based board factories. Indoor Built Environ. 18:265–269.

Stevens, V. (1997) The ecological role of coarse woody debris: an overview of the ecological importance of CWD in BC forests. Research Program, BC Ministry of Forests, Victoria, BC, Canada. Working Paper 30/97.

Terziev, N., Nilsson, T. (1999) Effect of soluble nutrient content in wood on its susceptibility to soft rot and bacterial attack in ground test. Holzforschung 53:575–579.

The Korean Society of Plant Pathology. (2009) List of Plant Diseases in Korea, 5th edn. The Korean Society of Plant Pathology, Suwon, Korea. pp. 15–214.

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