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FUNGAL DISEASES

Short communication

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Anthracnose of *Sansevieria trifasciata* caused by *Colletotrichum sansevieriae* sp. nov.

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Abstract A *Colletotrichum* sp. was isolated from water-soaked lesions on sansevieria (*Sansevieria trifasciata* Prain cv. Laurentii) in Japan. Classifying the species only from the morphology of the fungus was difficult; therefore, host range was tested and the ribosomal DNA ITS2 region was phylogenetically analyzed. The fungus was pathogenic only on sansevieria among 20 test plants belonging to 11 families. In a phylogenetic analysis with the neighbor-joining method, the two isolates used formed a single-isolate clade. The fungus is thus proposed to be a new species, *Colletotrichum sansevieriae*. This report is the first of anthracnose on sansevieria.

Key words *Colletotrichum sansevieria* · *Sansevieria* · New species · Anthracnose

Sansevieria (*Sansevieria trifasciata* Prain cv. Laurentii) is cultivated in subtropical regions in Japan and is used as a potted ornamental and for cut leaves (Kawase and Tsukamoto 1994). Since 1996, water-soaked lesions and leaf blight have been observed on sansevieria on Yoron Island, Kagoshima Prefecture, Japan. From the lesions, a *Colletotrichum* sp. was mainly isolated. To date, only *Fusarium moniliforme* (Kagiwatari 1985) and *Pythium spinosum* (Takeuchi et al. 2002) have been reported as pathogens of sansevieria in Japan. In this article, we describe a new fungal disease on sansevieria caused by *Colletotrichum* sp., which is proposed as a new species.

Symptoms. Round, water-soaked lesions were first found on leaves, after which the lesions rapidly enlarged and coalesced, resulting in blight of the leaves (Fig. 1A–C). Lesions were observed on all parts of both young and mature leaves. On blighted leaves, black and crustose cankers, typical symptoms of anthracnose, formed. On these cankers, abundant cylindrical conidia were observed (Fig. 1D).

Taxonomy. *Colletotrichum sansevieriae* M. Nakamura & M. Ohzono, sp. nov. (Fig. 1E–I). Coloniae in agaro decocto tuberosum, griseo-albus et partim cremea vel persicinum, pannosae cum mycelio aerio, raro setosae; reversum griseae vel atro-olivaceo griseae et partim cremea vel persicinum. Raro scletotia. Appressoria sepiacea vel fusco-brunneo, ovatae, unicellularia, 6.3–(7.7)–8.8 × 6.3–(7.3)–7.5 µm. Conidia recta cylindrical, ad apicem obtuse, ad basim leviter acuta; basis truncata ad locum cui affixa est, 12.5–(18.4)–32.5 × 3.8–(6.4)–8.8 µm. Habitat: *Sansevieria trifasciata* Prain cv. *Laurentii.* Holotypus: Cultura (Sa-1-2:MAFF239721), isolata e foliis morbo affectis *Sansevieria trifasciata* Prain cv. Laurentii, Yoron insulae, Kagoshima, Japonia, 1997.

Colonies on potato dextrose agar (PDA) grayish-white and partly cream to pink, felted with aerial mycelium, reverse grey to dark olivaceous grey and partly cream to pink. Setae rare. Sclerotia rare. Appressoria sepia to dark brown, ovate, one-celled, 6.3–(7.7)–8.8 × 6.3–(7.3)–7.5 µm. Conidia straight, cylindrical, obtuse at the apex, and slightly acute at the base, base with truncate attachment point, 12.5–(18.4)–32.5 × 3.8–(6.4)–8.8 µm. On and from *Sansevieria trifasciata* Prain cv. Laurentii. Etymology: sansevieriae from sansevieria, the host of the fungus; the fungus is pathogenic only on sansevieria. Holotype: a culture (Sa-1-2:MAFF239721) isolated from *Sansevieria trifasciata* Prain cv. Laurentii, Yoron Island, Kagoshima, Japan, 1997. The culture was deposited in the NIAS Genebank, National Institute of Agrobiological Sciences as a frozen culture in a vapor phase of liquid nitrogen.

This description is based on the isolate (Sa-1-2:MAFF239721) grown on PDA in the dark at 25°C. Conidia from 10-day-old cultures were measured. Conidia from lesions are also shown in Fig. 1D. Setae formed on lesions were not observed. Appressoria were produced on a potato–carrot agar slide culture. The optimum growth temperature ranged from 25° to 28°C and at 25°C mycelia grew 22 mm during 7 days. Because the fungus was difficult to

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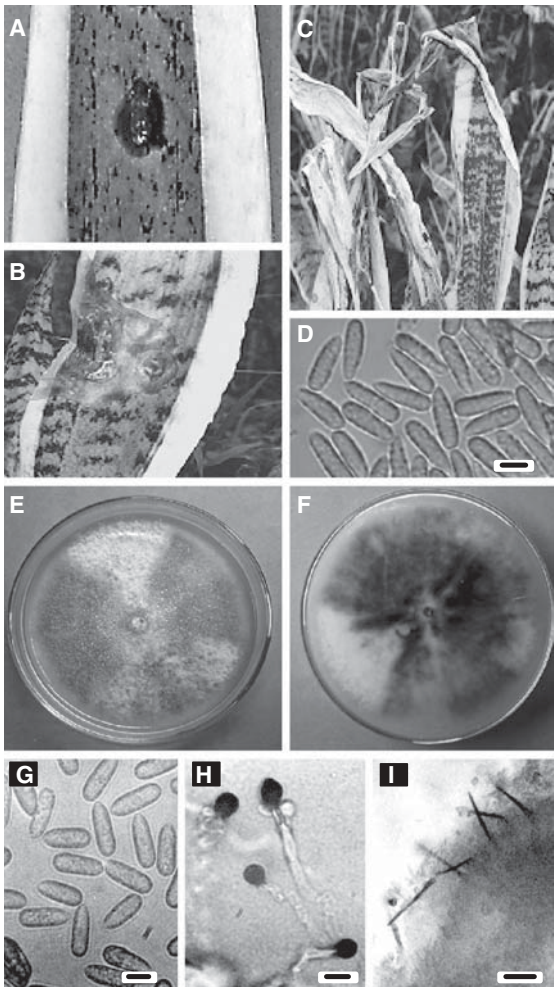

Fig. 1. A–C. Symptoms of anthracnose on *Sansevieria trifasciata* cv. Laurenii by *Colletotrichum sanseveriae*. **D** Conidia formed on *S. trifasciata* cv. Laurenii. *Bar* 10 µm. **E, F** Colony surface (**E**) and reverse (**F**) of *C. sanseveriae* cultured on potato dextrose agar (PDA) at 25°C for 14 days. **G** Conidia formed on PDA. *Bar* 10 µm. **H** Appressorium formed on a potato–carrot agar slide culture. *Bar* 10 µm. **I** Setae formed on PDA at 25°C in darkness. *Bar* 50 µm. All morphology is based on *C. sanseveriae* (Sa-1-2:MAFF239721)

identify only from its morphology, most of which overlapped with those of species such as *Colletotrichum gloeosporioides*, *Colletotrichum acutaum*, and *Colletotrichum orbiculare* (von Arx 1957; Sato 1996; Sutton 1980), we tested the host range and analyzed the ribosomal DNA ITS2 region phylogenetically.

Pathogenicity and host range. For the pathogenicity test, wounded (with needle) and unwounded leaves of potted sansevieria cv. Laurentii were inoculated with cotton (10 × 10 mm) soaked in a conidial suspension (1×10^6 conida/ml in sterilized distilled water). Inoculated plants were kept in a humidified chamber at 27°C under 12 h fluorescent light/12 h darkness. Six days after inoculation, water-soaked lesions appeared on only wounded leaves, and the lesions enlarged and had coalesced within 2 weeks. The fungus was readily reisolated from the inoculated leaves. This is the first report of a disease caused by a *Colletotrichum* sp. on

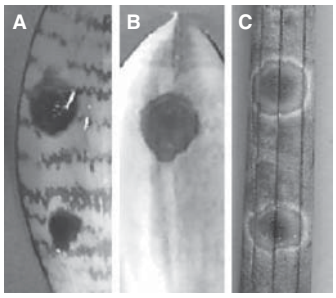

Fig. 2A–C. Symptoms on *S. trifasciata* cv. Hanii (**A**), cv. Golden Hanii (**B**), and *Sansevieria stuckyi* (**C**) inoculated with *C. sanseveriae* (Sa-1-2:MAFF239721)

Table 1. Results of inoculation tests with *Colletotrichum sansevieriae*

Plants	Pathogenicity
Phaseolus vulgaris	–
Cucumis melo	–
Cucumis sativus	–
Fragaria chiloensis	–
Spinacia oleracea	–
Capsicum annuum	–
Lycopersicon esculentum	–
Lilium Oriental Group (cv. Casablanca)	–
Corchorus olitorius	–
Yucca elephantips	–
Agave americana	–
Dracaena sanderiana	–
Dracaena concinna	–
Cordyline terminalis cv. Red Edge	–
Sansevieria trifasciata Prain cv. Laurentii	+
Sansevieria trifasciata Prain cv. Hanii	+
Sansevieria trifasciata Prain cv. Golden Hanii	+
Sansevieria stuckyi	+
Carica papaya (fruit)	–
Mangifera indica (fruit)	–

+, pathogenic; –, nonpathogenic

sansevieria in Japan. We propose the name anthracnose of sansevieria (*sanseberia-tanso-byo* in Japanese). Previously, *C. gloeosporioides* was isolated from *Sansevieria trifasciata* Prain cv. Laurentii (Horie et al. 1990), but its pathogenicity was not confirmed. We also isolated *C. gloeosporioides* and tested its pathogenicity on sansevieria; but it was not pathogenic to the plant (data not shown). *Colletotrichum gloeosporioides* was thus thought to be a secondary saprophyte.

To investigate the host range of the fungus, 20 plants belonging to 11 families were used (Table 1), most of which are the host plants of *Colletotrichum* species that are morphologically similar to the fungus. Plants were inoculated with a conidial suspension or with colony discs. The fungus proved to be pathogenic only on sansevieria plants (Table 1, Fig. 2), indicating that the fungus has a high host specificity.

Phylogenetic analysis. The ITS2 regions of two isolates (Sa-1-2:MAFF239721 and Sa-1-1 preserved in our laboratory) were analyzed phylogenetically with those of other *Colletotrichum* species. ITS1 regions are often used for

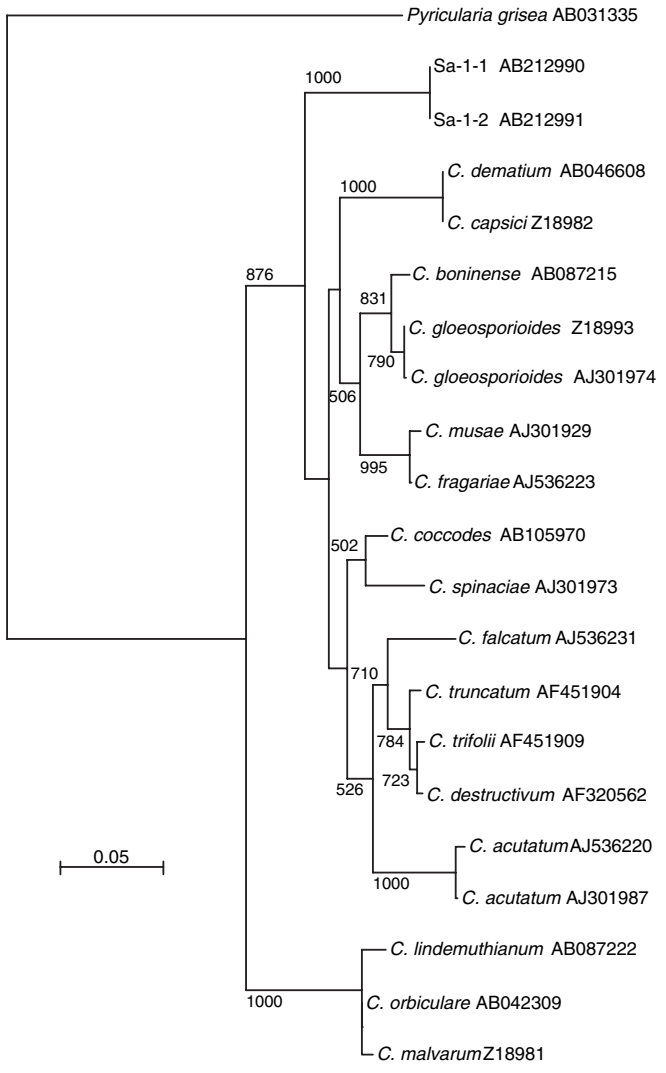

Fig. 3. A phylogenetic tree of *Colletotrichum* species based on ITS2 regions by neighbor-joining analysis. *Numbers* on the branches are the frequency with which a cluster appears in a bootstrap test of 1000 runs. Bootstrap values greater than 500 are shown. Designations to the *right* of the species names are the accession numbers in the DNA Database of Japan (DDBJ)

phylogenetic analysis; however, we chose the ITS2 regions to clearly distinguish the isolates from other *Colletotrichum* species because ITS2 regions are more conservative (less variable) than ITS1 (Sreenivasaprasad et al. 1996; Martinez-Culebras et al. 2003). The ITS2 regions were amplified with primers Pn3 and Pn8 (Bailey et al. 1996) and direct-sequenced with primer Pn8/10 (Bailey et al. 1996). The ITS2 regions of the two isolates had 159 bp and 100% homology. The regions were compared with those of other *Colletotrichum* species registered in the DNA Database of Japan (DDBJ), most of which are morphologically similar to the isolates. The highest homology that the two isolates showed was only 90.6% with *Colletotrichum boninense* (Table 2) and in a neighbor-joining tree, the two isolates made a single-isolated clade at a high bootstrap value (Fig. 3), indicating that the two isolates are clearly distinct from other *Colletotrichum* species.

Table 2. Sequence similarities (%) of the ITS2 region among *Colletotrichum* species

No.	Isolates (accession no.)	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16	17	18	19	20
1	*C. acutatum* (AJ301987)	–																			
2	*C. acutatum* (AJ536220)	99.3	–																		
3	*C. boninense* (AB087215)	90.3	90.7	–																	
4	*C. capsici* (Z18982)	87.2	88.5	92.9	–																
5	*C. coccodes* (AB105970)	92.2	92.7	93.7	93.6	–															
6	*C. dematium* (AB046608)	87.2	88.5	92.9	100[a]	93.6	–														
7	*C. destrictivum* (AF320562)	92.2	91.5	91.6	89.9	94.3	89.9	–													
8	*C. falcatum* (AJ536231)	91.4	90.7	91.4	90.1	92.7	90.1	92.9	–												
9	*C. fragariae* (AJ536223)	91.4	90.7	94.7	90.9	94.0	90.9	92.9	89.5	–											
10	*C. gloeosporioides* (Z18993)	91.4	90.7	98.7	94.3	92.3	94.3	90.5	91.4	95.4	–										
11	*C. gloeosporioides* (AJ301974)	91.4	90.7	98.7	94.4	92.3	94.4	90.7	91.4	95.5	100	–									
12	*C. lindemuthianum* (AB087222)	85.3	85.5	85.8	83.0	84.0	83.0	82.9	83.6	84.9	84.9	84.8	–								
13	*C. malvarum* (Z18981)	86.0	85.6	86.2	82.9	84.4	82.9	84.0	83.9	85.2	85.4	84.9	98.1	–							
14	*C. musae* (AJ301929)	91.4	90.7	94.2	90.4	91.7	90.4	90.5	89.4	99.3	95.5	95.5	82.9	83.0	–						
15	*C. orbiculare* (AB042309)	86.6	86.3	86.9	83.5	85.0	83.5	84.7	84.5	85.8	86.0	85.5	98.8	99.4	83.6	–					
16	*C. spinaciae* (AJ301973)	90.3	89.9	90.8	92.7	95.5	92.7	91.8	88.7	92.2	90.9	90.9	85.9	86.6	91.6	87.3	–				
17	*C. trifolii* (AF451909)	93.5	92.8	92.8	89.9	94.3	89.9	98.8	94.2	92.8	92.0	91.7	82.4	83.5	91.1	84.1	91.1	–			
18	*C. truncatum* (AF451904)	92.9	92.7	92.3	89.4	94.9	89.4	97.5	95.4	93.5	93.5	93.4	82.9	84.0	94.0	84.7	91.7	98.1	–		
19	Sa-1-1 (AB212990)	88.0	89.0	90.6	86.2	86.2	86.2	89.1	85.2	88.9	89.9	89.9	82.0	82.7	87.4	83.3	84.6	89.1	87.4	–	
20	Sa-1-2 (AB212991)	88.0	89.0	90.6	86.2	86.2	86.2	89.1	85.2	88.9	89.9	89.9	82.0	82.7	87.4	83.3	84.6	89.1	87.4	100	–

Similarities were calculated by GENETYX-MAC Network version 13. 0. 3 (GENETYX Corp., Tokyo, Japan)
[a] *Colletotrichum dematium* was molecularly identified as *C. capsici* (Sreenivasaprasad et al. 1996)

On these grounds (host-range test and phylogenetic analysis), we have come to the conclusion that the fungus is a new species, *Colletotrichum sansevieriae*.

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