

First Report of *Diaporthe actinidiae*, the Causal Organism of Stem-end Rot of Kiwifruit in Korea

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Post-harvest diseases of kiwifruit caused severe damages on the fruits during storage, transportation, marketing and consumption. *Phomopsis* sp. was reported to be one of the major causal organisms of post-harvest fruit rots of kiwifruit. Symptoms of stem-end rot caused by *Phomopsis* sp. appeared at the stem-end area of the fruit as it ripened. The brown pubescent skin at the area became soft and lighter in color than the adjacent firm healthy tissues. A watery exudate and white mycelial mats were frequently visible at the stem-end area forming a water-drop stain down the sides on the dry brown healthy skin. When the skin was peeled back, the affected flesh tissue was usually water-soaked, disorganized, soft and lighter green than the healthy tissue. *Phomopsis* sp. was consistently isolated from the diseased fruits, and its pathogenicity was confirmed by an artificial inoculation test on healthy fruit of kiwifruits. The mycological characteristics of the teleomorph state of the fungus produced on potato-dextrose agar were in accordance with those of *Diaporthe actinidiae*. This is the first report on the occurrence of a teleomorph state of *D. actinidiae* as the causal organism of stem-end rot of kiwifruit in Korea.

Keywords : *Diaporthe actinidiae*, kiwifruit, *Phomopsis* sp., stem-end rot.

Kiwifruit (Chinese gooseberry, *Actinidia deliciosa* (A. Chev.) C. F. Liang et A. R. Ferguson) has been grown in southern regions of Korea since the early 1980s. Kiwifruit is currently cultivated on about 1,500 ha in Korea and great amounts of the fruits have been imported from foreign countries due to the increasing domestic demand (Koh, 1995). Kiwifruit can be maintained more than 4 months when stored at $0 \pm 1^\circ\text{C}$ (Schroeder and Fletcher, 1967). However, fruit rot diseases caused a severe loss of kiwifruits during storage, transportation, marketing and con-

sumption after harvest. More than 7 fungi were reported to be associated with post-harvest fruit rots of kiwifruit (Hawthorne et al., 1982; Pennycook, 1985). Among the fungi, *Phomopsis* sp. and *Botryosphaeria* sp. were reported to be the major causal organisms of post-harvest fruit rots of kiwifruit (Beraha, 1970; Chung, 1997; Lee et al., 1998; Park et al., 1994; Sommer and Beraha, 1975). Fruit rot of kiwifruit caused by *Phomopsis* sp. was named as stem-end rot by Beraha (1970). Sommer and Beraha (1975) reported its teleomorph state as *Diaporthe actinidiae*. In Korea, Park et al. (1994) reported *Phomopsis* sp. as one of the major fruit rot pathogens of kiwifruit. Recently, Yi and Lee (1998) identified *Phomopsis mali* as the species of *Phomopsis* causing fruit decay of kiwifruit as well as Japanese apricot and apple, but the teleomorph state of the fungus has not been reported until now.

Diseased fruits were collected from kiwifruit orchards in Cheju and Chonnam provinces in 1999. Symptoms of stem-end rot appeared at the stem-end area of the fruit as it ripened. The brown pubescent skin at the area became soft and lighter in color than the adjacent firm healthy tissues. A watery exudate and white mycelial mats were frequently visible at the stem-end area forming a water-drop stain down the sides on the dry brown healthy skin (Fig. 1A). The affected flesh tissue visible when the skin was peeled back was usually water-soaked, disorganized, soft and lighter green than the healthy tissue (Fig. 1B). A sour or fermented odor occurs frequently from severely decayed fruits. Decayed fruits were much softer than ripe healthy fruits and often a bitter taste was associated with the decay.

A species of *Phomopsis* was consistently isolated from the lesions showing typical symptoms of stem-end rot. Chalk white-colored aerial mycelial mats were formed with a circular shape on potato-dextrose agar (PDA) plates after incubation at 25°C over 7 days (Fig. 1F). After 4 weeks of incubation, black, spherical or bluntly conical pycnidia bearing α - and β -conidia were formed all over the mycelial mats and measured $230 \times 500 \mu\text{m}$ in size (Fig. 1K). A stock culture was prepared from the colony by transferring a single α -conidium with a sharp needle under a dissecting

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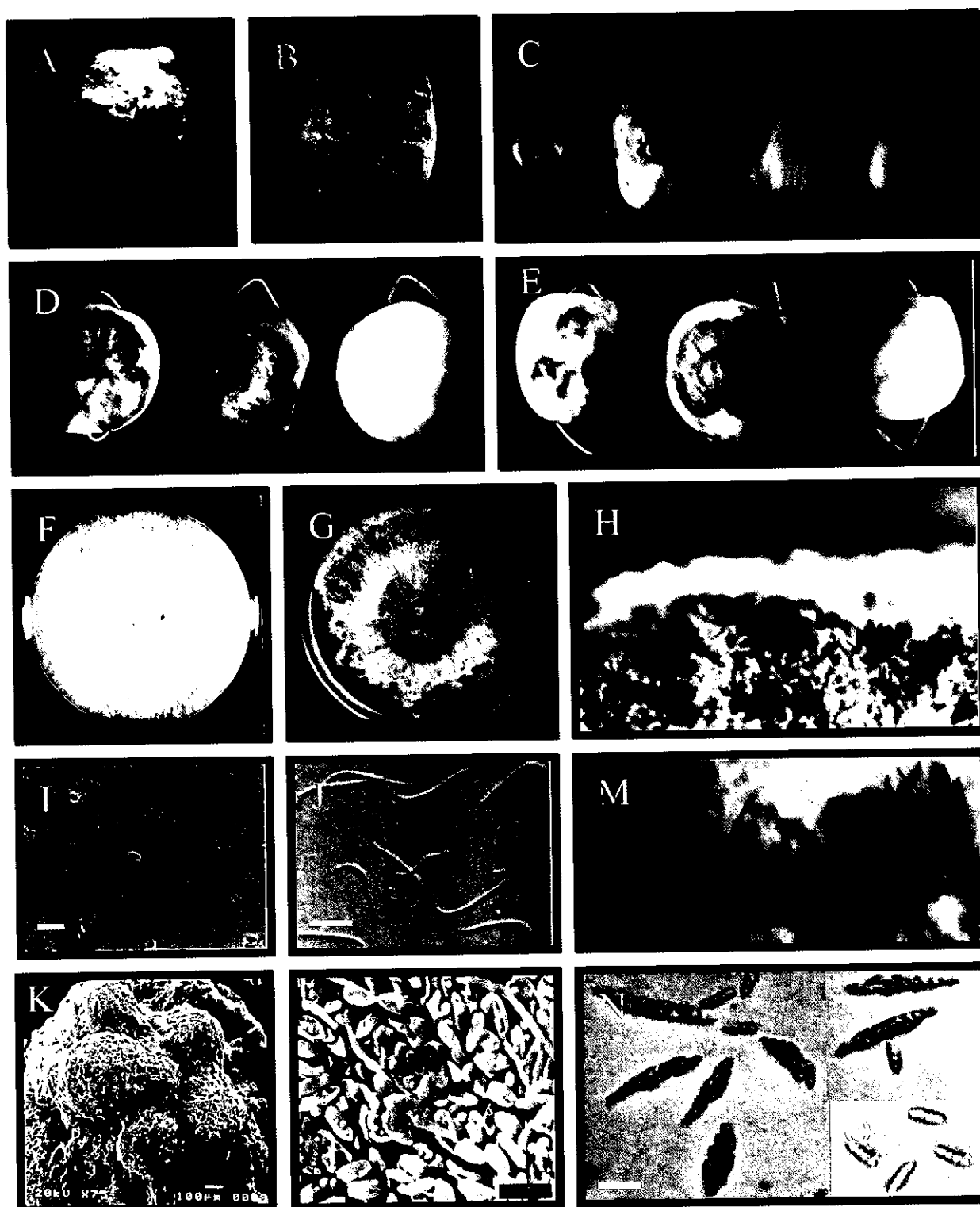


Fig. 1. Symptoms of stem-end rot of kiwifruits and mycological characteristics of *Diaporthe actinidiae*. (A) External symptoms on naturally infected kiwifruit. (B) Internal symptoms on naturally infected kiwifruit. Note disorganized soft decay tissue of kiwifruit. (C) Internal symptoms on artificially infected kiwifruits. (D) Internal symptoms on artificially infected pears. (E) Internal symptoms on artificially infected apples. (F) Mycelial colony of 7-day-old culture of *D. actinidiae* on PDA. (G) Mycelial colony of 45-day-old culture of *D. actinidiae* on PDA. (H) Margins of the older culture showing abundant perithecia. (I) Micrograph of α -conidia. Bar=10 μ m. (J) Micrograph of β -conidia. Bar=10 μ m. (K) Scanning electron micrograph of pycnidia produced on PDA. Bar=100 μ m. (L) Scanning electron micrograph of α - and β -conidia placed on the surface of PDA. Bar=5 μ m. (M) Prithecia on stroma. (N) Micrographs of asci. Bar=10 μ m. (O) Micrographs of ascospores. Bar=10 μ m.

Table 1. Morphological characteristics of the present isolates compared with *Diaporthe actinidiae* previously described

Characteristics		Present isolates	<i>D. actinidiae</i> ^a
Conony on PDA	Mycelia	chalk white	chalk white
	Older cultures	dark gray	tan to light brown
	Margins	white at surface and deep in agar	black at surface and deep on agar
Pycnidia	Shape	black, spherical, bluntly conical	black, flask shaped, bluntly conical
	Size (µm)	230 × 500	250 × 1000
α-Conidia	Shape	hyaline, unicellular, ovate	hyaline, unicellular, ovoid
	Size (µm)	4.5-7.5 × 2.5-3.5	6.9 × 2.4
β-Conidia	Shape	hyaline, filiform, curved at one end	hyaline, filiform, curved at one end
	Size (µm)	14.5-30.0 × 1.0-2.5	20.5 × 1.6
Perithecia	Shape	irregular clusters, black, globose, ostiolate, with necks sinuous, filiform	irregular clusters, black, globose, ostiolate, with necks sinuous, filiform
	Size (µm)	550-840 × 50-95	470-900 × 50-120
Asci	Shape	clavate, sessile	clavate, sessile
	Size (µm)	27.5-40.0 × 7.5-12.0	29-40 × 5.2-7.3
Ascospores	Shape	biseriate, hyaline, 2-celled, constricted at the septum, fusoid to ellipsoid	biseriate, hyaline, 2-celled, constricted at the septum, fusoid to ellipsoid
	Size (µm)	8.0-12.5 × 2.5-3.0	8.9-9.4 × 3.1

^aData from Sommer and Beraha (1975).

microscope to a PDA slant for further study. On PDA plates, α-conidia were hyaline, unicellular, and fusiform and 1.6-2.6 µm wide × 4.3-7.5 µm long (Fig. 1I) and β-conidia were hyaline, unicellular, filiform to hamate and 0.8-1.5 µm wide × 18.2-37.5 µm long (Fig. 1J) (Table 1).

Abundant perithecia embedded in usually distinct, black, elevated stroma in irregular clusters were yielded on PDA plates more than 8 weeks after incubation at room temperature (Fig. 1H). Perithecia were black, globose, 200-500 µm in diameter with necks sinuous, filiform, 50-95 × 550-840 µm in size (Fig. 1L). Asci were released from crushed stroma containing perithecia and were clavate, sessile and 27.5-40.0 × 7.5-12.0 µm in size (Fig. 1M), which contained biseriate, hyaline, 2-celled, constricted at the septum, fusoid to ellipsoid ascospores of 8.0-12.5 × 2.5-3.0 µm in size (Fig. 1N). The mycological characteristics of the teleomorph state of *Phomopsis* sp. were in accordance with those of *D. actinidiae* (Table 1) (Sommer and Beraha, 1975). Of 17 isolates, Sommer and Beraha (1975) found 5 isolates forming a teleomorph state on PDA plates 6-8 weeks after incubation at 20-25°C. In this experiment, only one isolate developed perithecia from hundreds of isolates incubated on PDA plates but neither perithecia nor pycnidia were produced on host fruits.

For pathogenicity test, inoculum of *D. actinidiae* was prepared by inoculating mycelial plugs on PDA plates and culturing at 25°C for 6 weeks. Twenty near-ripe healthy fruits were surface-disinfected with 70% ethanol for 2 min, rinsed in sterile water and air dried. The conidial suspension (10⁶ α-conidia/ml) harvested from the culture was sprayed

onto the healthy fruits of kiwifruit with or without wounding 2-3 mm deep by 10 sterile pins. All the fruits inoculated were maintained in a polyethylene bag at a dark incubator (25°C) for 3 or 7 days. Five fruits were served as controls and were treated with sterilized water and maintained under the same conditions. The wound-inoculated fruits produced droplets of liquid within 2 days and mycelial growth was visible at the stem-end area 3 days after inoculation. Disorganized soft decay was observed in the affected flesh tissue when the skin was peeled back 5 days after inoculation, but no visible symptoms were observed on the fruits without wounding (Fig. 1C). The flesh of the inoculated fruits became decayed drastically with a sour or fermented odor, as the disease progressed. The control fruits did not produce any manifestation of decay. Pieces of the decayed tissue were transferred to fresh PDA plates from the margin of the lesion on the inoculated fruits. All yielded the causal *D. actinidiae* organism.

The same methods were applied to the healthy pear and apple fruits in order to confirm the host range of *D. actinidiae*. Similarly, the typical symptoms were observed on the wounded fruits of pear and apple, respectively, but the severities were somewhat milder than those on kiwifruits (Fig. 1D, E) (Table 2). The result suggests that *D. actinidiae* can hardly infect healthy fruits and that only wounded fruits are vulnerable to the infection of the fungus. In fact, the kiwifruits are believed to be infected by *D. actinidiae* through wounds, since there are numerous chances to be wounded inevitably during harvest, selection, storage, packing, transportation, and marketing of kiwifruits. There-

Table 2. Pathogenicity of *Diaporthe actinidiae* isolated from kiwifruit to fruits of kiwifruit, apple and pear by artificial inoculation

Host	Pathogenicity ^a	
	Wounded	Not wounded
Kiwifruit	++	-
Apple	+	-
Pear	+	-

^aPathogenicity was evaluated by the severity of fruit rot 5 days after inoculation.

++: Severe symptoms, +: Mild symptoms, -: No symptom.

fore, the kiwifruit should be carefully handled in order to prevent wounds on fruits. This is the first report on the occurrence of a telemorph state of *D. actinidiae* as the causal organism of post-harvest stem-end rot of kiwifruit in Korea.

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