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Note

Occurrence of Stem Rot of *Disporum smilacinum* Caused by *Sclerotium rolfsii* in Korea

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In 2005 and 2007, a basal stem rot of *Disporum smilacinum* caused by *Sclerotium rolfsii* occurred sporadically in a herb farm at Hamyang, Korea. The symptom initiated with water-soaking lesion and progressed into stem rot and wilt of a whole plant. Severely infected plants were blighted and died eventually. White mycelial mats appeared on the lesion at early stage and a number of sclerotia were formed on the stem near the soil line. The sclerotia were globoid in shape, 1-3 mm in size and white to brown in color. The optimum temperature for the growth and sclerotia formation was 30 on PDA and the hyphal width was measured 3-8 μm. The typical clamp connections were observed in the hyphae of the fungus grown on PDA. On the basis of symptom, mycological characteristics and pathogenicity to the host plant, this fungus was identified as *Sclerotium rolfsii* Saccardo. This is the first report on the stem rot of *D. smilacinum* caused by *S. rolfsii* in Korea.

Keywords **:** *Disporum smilacinum*, *Sclerotium rolfsii*, Stem rot

Disporum smilacinum belonging to *Liliaceae* is a perennial wild plant grown in the mountainous area. However, the plant is cultivated in some areas because its shoots are used as a medicinal herb or vegetable in Korea (Lee, 2003).

A destructive stem rot of *Disporum smilacinum* caused by *Sclerotium rolfsii* was sporadically observed in the experimental field at Hamyang, Gyeongsangnam-do Agricultural Research and Extension Services in 2005 and 2007. The disease mainly occurs on stems, sometimes on leaves, seemed important for the safe cultivation of the plant, however, only a leaf spot caused by *Cercospora dispori* has been recorded on the plant in Korea (The Korean Society of Plant Pathology, 2004).

The sclerotial diseases caused by *S. rolfsii* primarily occur in warm climates, especially under a high temperature with humid condition. The pathogen of sclerotial diseases causes damping-off of seedlings, stem canker, crown blight, and rots on root, crown, bulb, tuber and fruit of various plant groups (Gobayashi et al., 1992). Sclerotial diseases frequently affect a wide variety of plants, including most vegetables, flowers, legumes, cereals, forage plants and eeds (Agrios, 2005).

Sclerotium rolfsii that had not been considered as a serious pathogen in the country became one of the most important soil-borne pathogens in recent years, especially in Gyeongnam province. Newly recorded hosts of the pathogen by the authors are *Tawny daylily*, *Capsicum annuum* (Kwon and Park, 2004), Strawberry (Kwon et al., 2004), *Aster glehni* and *Hosta longipes* (Kwon and Park, 2005).

Symptom and disease occurrence. In general, white mycelial mats were appeared on the surface of basal stems (Fig. 1A, B). The white mycelia were readily visible nearby soil surface and grew inside of stem tissues. The fungus produced numerous small globoid sclerotia of uniform size which were white at first and turned brown on both PDA and the host in the field. The heavily infected plants became water-soaked, rotted, wilted, blighted, and died eventually (Fig. 1C). In Gyeongnam area, the *Disporum smilacinum* is cultivated in the fields over the years. Cultivation condition of the plant during the summer maintained high temperature and humidity around the plant because of dense planting and cannopy. Consequently, the cultivation practice favored an outbreak of the disease on stems of *D. smilacinum*. The infection rate of surveyed areas reached 36% in July of 2005. Abundant sclerotia of the pathogen were often produced on the surfaces of infected stems and near the soil-line in the fields, which play an important role of secondary inoculum in the fields.

Mycological characteristics. Freshly infected stems were collected from the fields and cut into small pieces for isolation of the causal pathogen. The small pieces of 5×5 mm in size, were disinfected in 1% NaOCl solution for 30 seconds and washed in distilled water for 3 times. The causal fungus was easily isolated on water agar (WA) and

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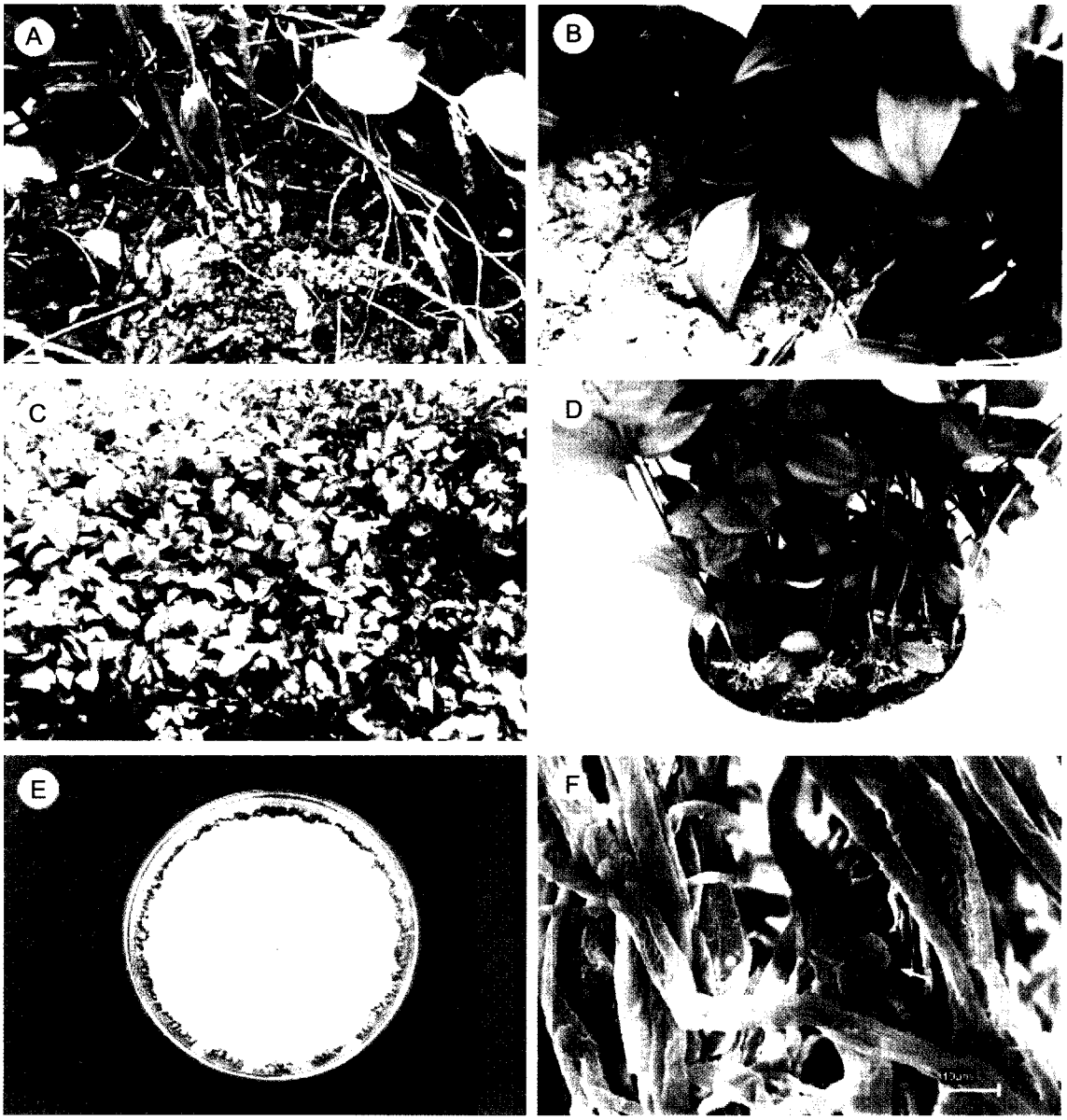

Fig. 1. Symptoms of stem rot of *Disporum smilacinum* and mycological characteristics of the pathogenic fungus, *Sclerotium rolfsii.* A and B: typical symptoms occurred on stems and near soil line in the field, C: infected plants wilted, blighted and died eventually, D: symptoms after artificial inoculation, E: mycelial mat and sclerotia grown on PDA after 18 days, F: clamp connection (arrow).

grew well on potato dextrose agar (PDA). After incubation for 72 hrs at 25°C, mycelial tips growing out the tissues were cut and transferred to PDA for further study. The cultural pattern of the fungus was examined after incubation for 18 days at 25-30°C on PDA. For the scanning electiron microscopy, the culture was cut into pieces 5×5 mm with a surgical blade and fixed with 2.5% Karnovsky solution for 24 hrs at 4°C. After fixing the specimens were rinsed carefully with 0.05 M cacodylate buffer (pH 7.2) for 10 min 3 times. Post fixing, the specimens were soaked in 1% osmium tetroxide solution for 2 hrs at 4°C and rinsed carefully 3 times with the same buffer solution. They were dehydrated through a series of ethanol solution gradient (50, 75, 90, 95 and 100%) after soaking for 20 min at room temperature, and followed by isoamyl acetate 100% for 1 hr 2 times. After that, the samples were dried by critical point dryer for 1 hr and were coated by gold/palladium coating by Sputter coater.

The fungus grew between 10 and 35°C, and optimal growth was recorded at 30°C on PDA, however, it did not grow below 5°C and over 40°C. The white mycelium usually formed many narrow hyphal strands in the aerial mycelium and they were measured 3-8 μm in width. This mycelium showed typical clamp connection structure (Fig.

Table 1. Comparison of mycological characteristics between the present isolate obtained from *Disporum smilacinum* and *Sclerotium rolfsii* described previously

Characteristics		Present isolate	*S. rolfsii*[a]
Colony	color	white	white
Hyphae	diameter	3-8 μm	4.5-9 μm
	clamp connection	present	present
Sclerotium	shape	globoid	spherical
	size	1-3 mm	1-2 mm
	color	brown	brown

[a] Described by Mordue (1974)

1F). The number of sclerotia was investigated for the characteristics after mycelial growth for 18 days. Small and globoid sclerotia were produced on the surface of lesions. The sclerotia were white and turned to dark brown at maturation with relatively uniform sizes. The maximum numbers of sclerotia produced at 30°C, but the sclerotium formation was sharply reduced below 20°C or over 35°C on PDA. However, no sclerotium was observed below 10°C or over 40°C. The size of sclerotia were measured 1-3 mm and the shapes were mostly spherical (Fig. 1E, Table 1).

Pathogenicity test. Pathogenicity of the fungus to *D. smilacinum* was tested in a greenhouse at Gyeongsangnam-do Agricultural Research and Extension Services, Korea. Totally 60 plants collected in 2006 were cultivated in Wagner pots (1/5000a) filled with autoclaved field soil. Inoculum was prepared with mycelial mats grown on PDA for 4 days. The plants were inoculated with 500 g of the inoculum source near the basal stems in May to June, 2007. The first symptom was appeared 7 days after inoculation and developed to severe stem rot and blight causing eventual death (Fig. 1D). The causal pathogen was re-isolated from the lesions to prove Koch's postulation.

Based on the characteristics examined in this study, present isolates causing stem rot on *Disporum smilacium* was identified as *S. rolfsii* described by previous workers (Mordue, 1974). The type culture of the fungus is stored at the Korean Agricultural Culture Collection (KACC No. 42085), National Institute of Agricultural Biotechnology, Rural Development Administration, Suwon.

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