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Phytophthora Rot on *Luffa cylindrica* Caused by *Phytophthora nicotianae*

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In 2004 and 2005, *Phytophthora* rot on *Luffa cylindrica* which had not been reported in Korea occurred in the experimental field at Gyeongsangnam-do Agricultural Research and Extension Services. The disease initiated on leaves and fruits of the plant with small water-soaked dark brown spots and progressed rapidly. The causal pathogen isolated from diseased tissues was identified as a *Phytophthora* sp. because of aseptate mycelia and zoospores released directly from sporangia. The fungus grew well on PDA and 10% V-8 juice agar showing an arachnoid or rosaceous colony pattern. Sporangia formed abundantly in water and were conspicuously papillate, noncaducous, ovoid to globose, and sized 26~62×19~38 μm. The fungus was heterothallic as producing sexual reproduction structures only when mated with only A2 standard mating type strain. Oogonia and oospores were spherical, smooth walled, and measured as 20~28 μm and 16~24 μm, respectively. Oospores were aplerotic and antheridia were amphigynous, unicellula and spherical. Chlamydospores were globose and 20~38 μm in diameter. Optimum temperature for growth was around 28~30°C. The fungus caused similar symptoms on artificially inoculated plant and could be re-isolated thereby proving Koch's postulation. Based on the mycological criteria investigated in this study, the causal fungus of *Luffa sylindrica* rot was identified as *Phytophthora nicotianae*. This is the first report of *Phytophthora* rot of *Luffa cylindrica* caused by *P. nicotianae* in Korea.

Keywords **:** *Luffa cylindrica, Phytophthora* rot, *Phytophthora nicotianae*

Luffa cylindrica belongs to Cucurtitaceae was originated in South Asia, South Europe and Africa. *L. cylindrica* is used in medicine, make-up, slippers, hats and wallpaper (Young, 1989; Lee, 2003). Generally, *L. cylindrica* is cultivated from May to October in open fields. Since 2004 a new disease on *L. cylindrica* caused by *Phytophthora* continuously occurred in most of the cultivation area in Gyeongsangnam-do Agricultural Research and Extension Services. Only two diseases on the plant were previously recorded in Korea; Mosaic virus (*Cucumber mosaic virus, Watermelon mosaic virus*) and leaf spot caused by *Ascochyta* sp. (The Korean Society of Plant Pathology, 2004).

Phytophthora is known as one of the most destructive plant pathogen groups. It attacks most cultivation crops worldwide (Erwin and Ribeiro, 1996). Jee (1998) and Jee et al. (2000) reported that 18 species of *Phytophthora* distributed throughout Korea attack 70 host plants. However, *Luffa cylindrica* has not been listed as a host of the fungus in the country yet (The Korean Society of Plant Pathology, 2004).

In this study, the causal pathogen of *L. cylindrica* rot is identified based on mycological characteristics. Pathogenicity of the fungus to the plant was also confirmed.

Materials and Methods

Pathogen isolation. In 2004 and 2005, *Phytophthora* rot on *Luffa cylindrica* was observed in the experimental fields at Gyeongsangnam-do Agricultural Research and Extension Services. The disease mainly occurred on fruits and leaves of the plant. Freshly infected fruits were collected from the fields and cut into small pieces for isolation of the causal pathogen. The small pieces, sized 5×5 mm, were disinfected in 1% NaOCl solution for 30 seconds, washed in distilled water, and placed on both *Phytophthora* semi-selective medium and water agar (Jee et al., 1998; 2000). After incubation for 48 hr at 25°C, mycelial tips growing out from the infected tissues were cut and transferred to 10% V8 juice agar for further study.

Mycological characteristics. The cultural colony patterns of the pathogen were observed on potato dextrose agar (PDA) after incubation for 7-14 days at 25°C. Sexual and asexual reproduction structures of the pathogen were examined on 10% V8 juice agar. After incubation for 5 days at 25°C, actively growing mycelia were cut into small blocks (1.0 × 1.0 cm) and submerged in distilled water for sporulation. The sporangial features were examined under a light microscope after incubation for 24 hours under light.

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Mating types of the isolates were determined by mating with standard A1 or A2 type of *P. nicotianae*. Chlamydospores formed on the 10% V8 juice agar in old cultures were examined under a light microscope at 200 or 400×.

Pathogenicity test. Zoospores were used as an inoculum for the pathogenicity test. Mycelial agar blocks (1.0×1.0 cm) were made from 5 day old cultures grown on 10% V8 agar and transferred to new Petri dishes containing 25 ml of distilled water. The Petri dishes were incubated under a fluorescent light for 24 hr at 25°C for sporulation. The Petri dishes were chilled for 1 hour at 5°C for zoospore release from sporangium. Zoospores were collected by filtering through four layers of cheesecloth and adjusted to 10^4/ml. Fruits of *L. cylindrica* grown in a field for 140 days were used for the test. Fruits either wounded by using a pin or not wounded were fully sprayed with the inoculum and kept in a plastic growth chamber (29×22×15 cm) to supply 100% moisture for 24 hr in the laboratory.

Results and Discussion

Symptoms. *Phytophthora* rot was observed mostly on the fruits and leaves of *L. cylindrica*. On leaves, small water-soaked brownish irregular spots enlarged rapidly along veins and eventually the entire leaf was blighted (Fig. 1A). Similarly, small brownish spot developed on fruit expanded rapidly and the infected fruit dried after a few days under favorable conditions (Fig. 1B). The disease usually started with water-soaked lesions on the fruit's tip. The pathogen penetrated mainly through wounds and inner tissues of the fruit are decayed severely (Fig. 1C). Under a humid weather condition white mycelial mats were formed inside or on the surface of infected fruits in the fields. The heavily infected fruits became watery and mummified eventually (Fig. 1D, E). Abundant sporangia of the pathogen were often produced on the surfaces of infected fruits, which may play an important role as the secondary inoculum source attacking aerial parts of the plant by splash in the fields.

Environmental conditions. *L. cylindrica* was seeded in pots in late April, transplanted in early May, and cultivated in hurdle plank from May to October in the fields at Gyeongsangnam-do Agricultural Research and Extension Services. The disease started in late June and spread widely in July and August during the rainy season. The rain splash may play an important role in the rapid spread and aerial infection of the pathogen. The infected field showed about 6% infection rate on fruits and leaves. It was considered that such rainy weather conditions in the area favored the disease development in the open field and hurdle plank cultivation.

Mycological identification. Colony patterns of the isolate on PDA were arachnoid or rosaceus, which is one of the distinctive cultural characteristics of *P. nicotianae* (Fig. 2A). The fungi grew between 5°C and 40°C and optimally at 28°C. Sporangia were rarely produced on agar but abundantly in water. They were mostly ovoid to globose in shape, mainly noncaducous, conspicuously papillate, and 26~62×19~38 μm in size (Fig. 2B). Oogonium were spherical, smooth walled, and 20~28 μm in size (Fig. 2D). The isolates was heterothallic since oospores were formed

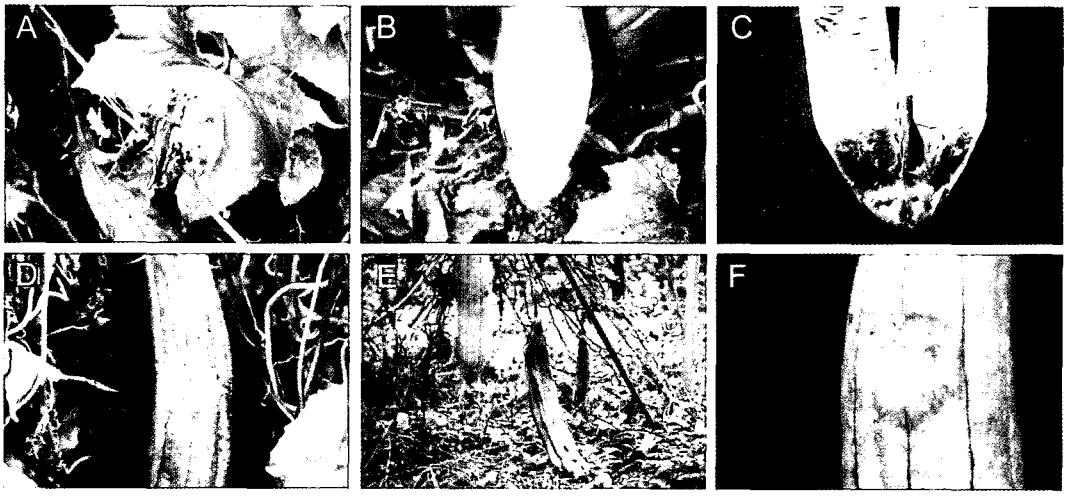

Fig. 1. Symptoms of the *Phytophthora* rot on *Luffa cylindrica* caused by *Phytophthora nicotianae*. A: Irregular lesions appeared on a leaf, B: A typical symptom on fruit, C: Longitudinal section of fruit showing symptoms caused by natural infection, D: White mycelia abundantly growing on surface of the fruit, E: A mummified fruit at late stage of infection, F: Symptoms on fruit caused by artificial inoculation.

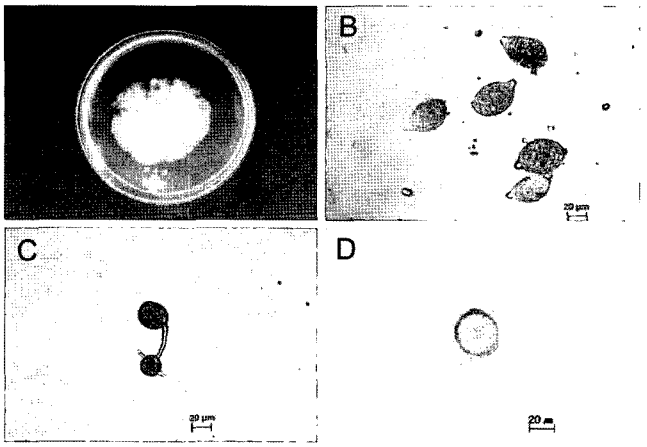

Fig. 2. Morphological characteristics of *Phytophthora* rot of *Luffa cylindrica* caused by *Phytophthora nicotianae*. A: Arachnoid colony morphology on potato dextrose agar for 8 days after incubation on PDA, B: Ovoid or globose sporangia, C: Chlamydospores, D: Oospores.

only when mated with A2 mating type strain. Hence, the isolates collected in this study were A1 mating type. Oospores were aplerotic, spherical in shape and 16~24 μm in size. Small and spherical antheridia were unicellular and amphigynous. Chlamydospores were also abundantly produced intercalary and terminal of aged culture on 10% V8 juice agar, globose in shape and sized mostly 20~38 μm in size (Fig. 2C, Table 1). Mycological characteristics of the isolate matched *Phytophthora nicotianae* as described by Erwin and Ribeiro (1996), Jee (1998) and Jee et al. (1998; 2000). Accordingly, we identified the causal pathogen of *L. cylindrica* rot as *P. nicotianae*.

Pathogenicity test. When zoosporagial suspension of a representative isolate sprayed on fruit to *L. cylindrica*, symptoms similar to those observed in the field appeared within 3-4 days on wounded fruit (Fig. 1F). Water-soaked lesions appeared on the surface of the fruit and rapidly rotted the whole fruit within 7 days. The pathogen was re-isolated from the freshly infected lesions to prove Koch's postulate. To our knowledge, this is the first report of *phytophthora* rot on *L. cylindrica* caused by *P. nicotianae* in Korea as well as in the world, since no report has been found in the fungal databases, systematic botany and mycology laboratory, U.S. Department of Agriculture, Agricultural Research Service.

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Table 1. Comparison of mycological characteristics of the *Phytophthora* causing *Luffa cylindrica* and *Phytophthora nicotianae*

Characteristics	Presented isolate	*P. nicotianea*[a]
Colony	arachnoid or rosaceous	dense or loose rosette
Sporangium		
shape	papillate, noncaducous, ovoid to globose	papillate, noncaducous, ovoid to spherical
size	26~62x19~38 μm	28~66x20~48 μm
Sporangiophore	branched	branched
Oogonium		
shape	spherical, smooth wall	spherical, smooth wall
size	20~28 μm	24~34 μm
Oospore		
shape	aplerotic, spherical	aplerotic, spherical
size	16~24 μm	18~34 μm
Antheridium	amphigynous, unicellula, spherical	amphigynous, unicellula, spherical
Chlamydospore		
shape	abundant, globose	abundant, spherical
size	20~38 μm	18~50 μm
Sexuality	heterothallic	heterothallic
Hyphal swelling	formed on agar and water	formed on agar and water

[a]Described by Breda de Haan, J. van (1896).

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