Downy Mildew of Astragalus membranaceous Burge Caused by Peronospora trifoliorum de Bary

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A severe downy mildew of Astragalus membranaceus occurred in Gangwon province in 1999 and 2000. Symptoms developed on young leaves and shoots, showing grayish white mycelium on the lower leaves. The infected plants had reduced internodes and twisted leaflets when the disease was severe. Peronospora trifoliorum was identified as the causal agent of the disease based on mycological characteristics. Pathogenicity of the fungus was confirmed by artificial inoculation. This is the first record of downy mildew on astragal plant caused by Peronospora trifoliorum in Korea.

Keywords: astragal, downy mildew, *Peronospora trifoliorum*.

Astragal (Astragalus membranaceus Burge) is one of the most important medicinal crops in Korea. Few diseases have been reported to infest this crop in the country (The Korean Society of Plant Pathology, 1998). However, it was found that many diseases on astragal plants were caused by downy mildew (Peronospora sp.), root rot (Phytophthora drechsleri), basal rot (Rhizoctonia solani), fusarium wilt (Fusarium oxysporium), and powdery mildew (Erysiphe pici) (Table 1). Among them, downy mildew was the most serious disease during the growing season, with a disease incidence ranging from 1 to 100% in the fields of the main cultivating areas in Gangwon province from 1999 to 2000. The objectives of this study were to: a) identify the causal agent of downy mildew; and b) determine the occurrence time of the disease and its impact on the root production of astragal.

Downy mildew of astragal occurred from early spring to fall, during the harvesting period, when temperature was low and humidity was high. Symptoms of the disease were chlorotic blotches on the upper leaf (Fig. 1A), and white to gray mycelium on the lower leaf (Fig. 1B). In the early stage, lesion appeared narrow, chlorotic or yellow to gray-ish yellow color on the leaves. Size of lesion was variable and deformation of leaves was observed on any part of the

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infected plant. Only young tissues were susceptible to infection. Early defoliation was observed in diseased plants and the internode was shortened with severe infection during the growing season.

The causal agent of downy mildew on astragal was identified as Peronospora trifoliorum based on mycological characteristics (Francis, 1983) (Table 2). For mycological characteristics of the pathogen, the surface of the lower infected leaves was observed by light and scanning electron microscopy (Fig. 1C-F). Coenocytic mycelium usually emerged singly through stomata. Hypha grew finger-like extension with 4-10 µm in width. Sporangiophore was dichotomously 5-7 branched and divided one-third at the top. The size of sporangiophore was measured 180-320 $\mu m \times 8-12 \,\mu m$ (avg. $250 \times 10 \,\mu m$). Sporangium was ellipsoid ovoid in shape and $24-29 \,\mu\text{m} \times 16-20 \,\mu\text{m}$ (avg. $26.5 \times 18 \,\mu\text{m}$) in size. Oospore with yellow or yellowish pink outer wall was found in diseased leaves and the shape was spherical. Oospore was 24-36 µm (in diameter) in size and had deeply wrinkled surface (Fig. 1D, F), that was distinguishable from others (Fallon and Sutherland, 1996).

For pathogenicity test, sporangia suspension was prepared by cutting off the mildewed leaves and young shoots. The materials were placed in a jar containing de-ionized water and were shaken to dislodge sporangia. Finally, sporangia suspension was harvested by passing through a five-layered cheesecloth to remove the plants (Fried and Stuteville, 1977).

The sporangia suspension adjusted to 10⁴ per ml was sprayed on astragal seedlings to run off. Then, the seedlings were immediately covered with double-layered polyethylene bags to maintain high humidity and placed in a chamber maintained at 18-20°C for 10 days. In another experiment, detached leaflets of astragal were used for pathogenicity. Leaflets were placed on water agar in plastic petri dishes (9 cm in diameter) with the abaxial surface downward. Each leaflet was inoculated with a 50 µl droplet of sporangia suspension (10⁴ per ml) chilled at 4°C for 2 h; the sporangia were obtained from diseased leaves. Then, the plates were placed in an incubator at 20°C for 10 days in the dark. All the isolates tested were pathogenic and grew vigorously on seedlings and detached leaves. The isolates

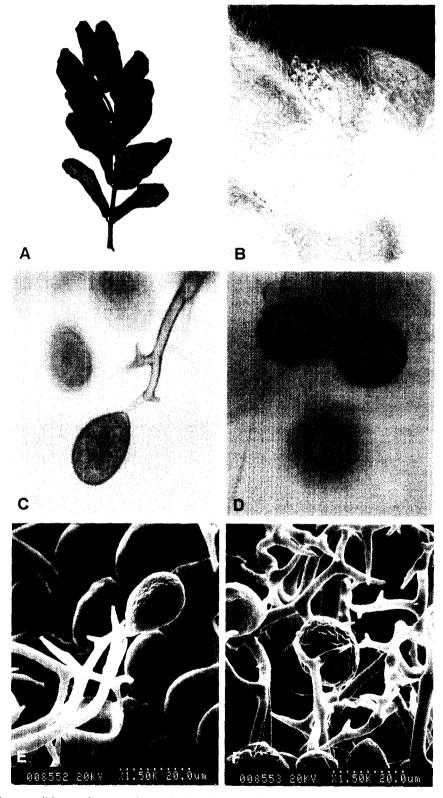


Fig. 1. Symptoms of downy mildew on leaves and mycological characteristics of *Peronospora trifoliorum*. Deformation in the margins and yellowish color on the surface of infected leaves (\mathbf{A}); white to gray fungus on the lower leaf (\mathbf{B}), sporangia and sporangiophores (\mathbf{C}) and oospores (\mathbf{D}) by light microscopy (\times 200); and scanning electron micrographs (\mathbf{E} , \mathbf{F}).

Table 1. Disease incidence of astragal plant in Gangwon province from 1999 to 2000

Disease (pathogen)	Incidence (%) ^a		A
	1999	2000	 Area with severe disease^b
Downy mildew (Peronospora trifoliorum)	1~90	1~100	JS, PC, GN, SC
Brown spot (Septoria astragalicola) ^c	0.1~5	1	JS, PC, GN, SC
Stem and basal rot (Rhizoctonia solani) ^d	0.5~1	0.1~10	JS, GN, PC
Fusarium wilt (Fusarium oxysporum) ^d	0.1~5	0.1~15	JS, GN, PC
Root rot (Phytophthora drechsleri) ^d	0.5~1	0.5~30	JS, GN, PC
Powdery mildew (Erysiphe pici) ^c	_	1~5	JS, GN, SC
Spot (Alternaria sp.) ^c	1~2	1~3	JS
Rust (<i>Uromyces</i> sp.)°	1~5	_	JS

Disease incidence was surveyed from May to September. JS: Jungsun, PC: Pyungchang, SC: Samcheok, GN: Gangnung, Percentage of diseased leaves. Percentage of diseased plants.

Table 2. Comparison of mycological characteristics of the present isolate from downy mildew on astragal plant and *Peronospora* trifoliorum

Characteristics	Present isolate	Peronospora trifoliorum"
Mycelium	Coenocytic finger-like extension 4-10 µm	Coenocytic and intercellular finger-like extension
Sporangia	Ellipsoid, ovoid a little narrow toward attached end $24-29 \times 16-20 \mu m$ (avg. $26.5 \times 18 \mu m$) pale violet to pale grayish in mass	Broadly ellipsoid brownish violet 24-29 × 18-21 µm
Sporangiophore	$180\text{-}320 \times 8\text{-}12~\mu\text{m}$ (avg. $250 \times 10~\mu\text{m}$) 5-7 dichotomously branched, nonseptate	$200-500 \times 4-8 \mu m$ branching obscurely dichotomous, 4-7 branched ends $5-9 \times 3 \mu m$ tapered rather abruptly to a blunt point
Oospore	Spherical yellow or yellowish pink outer wall, deeply wrinkled 24-36 µm in diameter	Straight or slightly curved yellow outer wall, somewhat wrinkled 20-30 μm in diameter

^aCMI Descriptions of pathogenic fungi and bacteria No. 768.

caused traces of lesion on the leaves within 24 h, which enlarged on the entire leaves 10 days after inoculation. The symptoms were similar to those of naturally infected leaves. Mycelium on the inoculated plants was highly branched with mycelial mat on the surface of lower leaves (Fig. 1B). There was no visible symptom on the plant treated with sterile water up to 10 days after inoculation.

As observed in the study, downy mildew of astragal plant may be broadly classified as primary and secondary infections. The primary infection may take place on young plants when shoots initiate growth in spring. The progress of infection on young shoot showed a characteristic symptom with chlorotic blotches and deformity in the margins of leaves. During the cultivation periods, farmers cut the shoot to induce the root growth at early or mid bud stage, and leave the shoot on the ground during the summer season. The fungus may survive on the shoot and function as a secondary inoculum for infection in newly growing shoot in fall. Even if the secondary infection does not produce severe disease, the plant may encounter other pathogens

with weak growth, resulting in reduced root production.

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