

Pathogenicity of *Didymella bryoniae* on the Seedlings of Cucurbits

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오이類 幼苗에 대한 덩굴마름병균의 病原性

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ABSTRACT

The objective of the study is to determine differences between cucurbits in the pathogenicity of *Didymella bryoniae* isolated from the naturally infected seeds of cucumber and pumpkin. Primary seedling infection of cucumber (*Cucumis sativus* L.), oriental melon (*Cucumis melo* var. *makuwa* Makino), pumpkin (*Cucurbita pepo* L.) and watermelon (*Citrullus vulgaris* Shrad.) occurred on the radicle, hypocotyl and cotyledons and symptoms on each crop were very similar. Infection of the radicle generally caused pre-emergence rot, while infection on the hypocotyl and cotyledons provided further inoculum for infection of the first true leaves and the stem. In cross inoculation tests, all isolates of *D. bryoniae* could infect cucumber, oriental melon, pumpkin and watermelon at different growth stages and there were not much differences in pathogenicity or susceptibility between isolates of the pathogen and crops tested. The susceptibility of cucumber and pumpkin was markedly influenced by prevailing humid conditions.

Key words: *Didymella bryoniae*, cucurbit disease.

要 約

本 研究는 自然罹病된 오이와 호박 種子에서 分離한 덩굴마름병균(莖枯病菌)의 病原性的의 差異를 調査한 것이 다. 오이, 참외, 호박 및 수박의 幼苗에 대한 一次 感染은 幼根·胚軸·子葉에 나타났으며 病징은 作物에 관계 없이 비슷하였다. 幼根이 병들었을 때는 發芽前 腐敗現象으로 나타났고 胚軸과 子葉의 感染은 第一葉과 줄기로 이어지는 接種源이었다. 交互接種試驗 結果 供試된 덩굴마름병균의 모든 分離菌은 오이, 참외, 호박, 수박의 종 자 및 유묘에 대하여 病原性이 있었으나 分離菌이나 供試作物間에는 差異가 별로 나타나지 않았다. 오이와 호 박의 感受性은 多濕條件에 의해서 크게 影響을 받았다.

INTRODUCTION

Gummy stem blight incited by *Didymella bryoniae* (Auersw.) Rhem is one of the most important diseases on cucurbits in many countries(2,3,4,7,8,9). It was

found in many commercial watermelon and oriental melon fields in Korea. Gummy stem blight on cucurbits causes crown blight, extensive defoliation and fruit rotting.

Grossenbacher(6) reported that when vines were

inoculated with *D. bryoniae* through wounds those of watermelon and muskmelon were very susceptible; those of pumpkin, gourd (*Lagenaria vulgaris*) and dishcloth gourd (*Luffa cylindrica*) were only mildly affected, and those of cucumber were not infected. Chiu and Walker(1) also reported that leaves of cucumber and squash were very resistant to *D. bryoniae* when they are in the seedling stage. On the other hand, Weber(10) noted wilting of leaves of young cucumber seedlings incited by *D. bryoniae*.

The objective of this study is to determine differences between cucurbits in pathogenicity of *D. bryoniae* isolated from the seeds of cucumber and pumpkin.

MATERIALS AND METHODS

Experiments with eight isolates of *D. bryoniae* isolated from naturally infected seeds of cucumber and pumpkin were carried out by three inoculation methods i.e., seed inoculation, soil inoculation and leaf inoculation. Crops tested were cucumber (cv. Seoul Madi-oi), oriental melon (cv. Euncheon-Chamoi), pumpkin (cv. Seoul Madi-Hobak) and watermelon (cv. Shindaewha no. 3). Seeds of each crop were used after surface sterilization in a solution containing one per cent available sodium hypochlorite for five minutes. For these seed, soil and leaf inoculations experiments, disease ratings were made at different intervals after incubation.

For seed inoculation experiment, a spore suspension of each isolate was prepared. The forty seeds of each crop were soaked in the spore suspension at room temperature in a covered beaker for 48 hours. Thereafter, they were removed, washed and plated out by standard blotter method. For soil inoculation the inoculum was developed on autoclaved moist wheat, incubated for 3 weeks and then mixed with 50% volume of pathogen-free soil. A 1-2 cm deep layer of inoculated soil was placed over pathogen-free soil in each pot (16 x 20cm). Ten seeds of each crop were separately planted at a depth of 1 cm, and for control in soil without inoculum. Three separate pots for each isolate and each crop were adequately replicated. Leaf inoculation was performed on seedlings about one month old and on plants at the third leaf stage. All

leaves of each plant were rubbed by a sterile cotton mop which was soaked in a heavy spore suspension of the pathogen. Ten seedlings were with sterile-distilled water to serve as control.

Throughout the experimental period for soil inoculation and leaf inoculation, seedlings were placed in a daylight room at 20°C and maintained in a moist

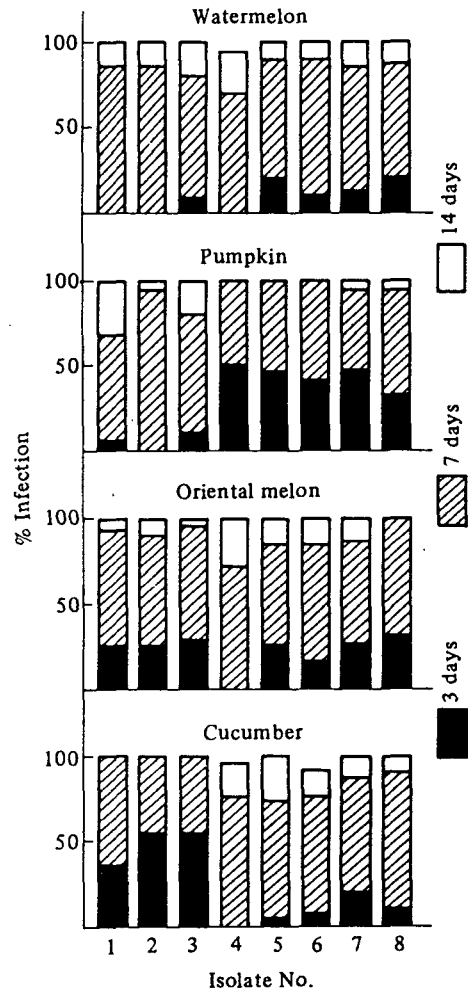


Fig. 1. Disease incidence of seedlings of cucumber, oriental melon, pumpkin and watermelon originating from seed cross-inoculated with eight isolates of *Didymella bryoniae*. Percent infections were estimated at different intervals (3, 7 and 14 days) of incubation. Isolate No. 1-3 isolated from naturally infected seeds of cucumber and No. 4-8 isolated from pumpkin seeds. Isolate No. 1-5 isolated from Korean seeds, isolate No. 6 from Lesotho, Isolate No. 7 from France and Isolate No. 8 from Italy.

conditions with plastic covers.

RESULTS

Seed inoculation. Result obtained from seed inoculation test was in Fig. 1. In general symptoms appeared on the crown and tip of radicle (Fig. 3). These symptoms were very similar to those on lesions from the naturally infected seeds grown on blotters. But in this experiment, many of seedlings showed the symptom on cotyledons. Pycnidial formation were commonly found on the seed coat, radicle and cotyledons where symptoms developed. Perithecia were also observed very often on seed coat of oriental melon, pumpkin and watermelon from three days after inoculation, but not on cucumber seeds. In early stage of infection, original hosts were more severe than others. Later, most of the seedlings showed typical symptom of *D. bryoniae*. It is evident that non of the inoculated seed samples of cucumber, oriental melon, pumpkin and watermelon showed the resistance to isolates of *D. bryoniae* from different hosts and different countries.

Soil inoculation. The result of cross inoculation of eight isolates of *D. bryoniae* by soil inoculation method is presented in Fig. 2. The reduction of seedling emergence was very much high percentage in each isolate and each crop (Fig. 4). On established seedlings, spotting of cotyledons was common (Fig. 6). In general, local spot on cotyledons which extended to hypocotyl and killed the plant or first-formed leaves were induced as a result of contact with infected cotyledons accompanied by excess moisture. When the non-emerged seeds were examined, those had started to germinate and killed by the pathogen.

Leaf inoculation. In leaf inoculations all isolates of *D. bryoniae* produced similar symptoms on cotyledons and first-formed leaves of the different hosts (Fig. 5). The first symptoms were visible two days after inoculation. The initial symptoms on cotyledons were small water soaked lesions which later became blighted and extended to hypocotyl and the first-formed leaf. They were irregularly shaped leaf-spots, appearing gray green at first and later becoming dark brown 7 days after inoculation. Sometimes, blight of the whole leaf followed. Under the excess moisture conditions,

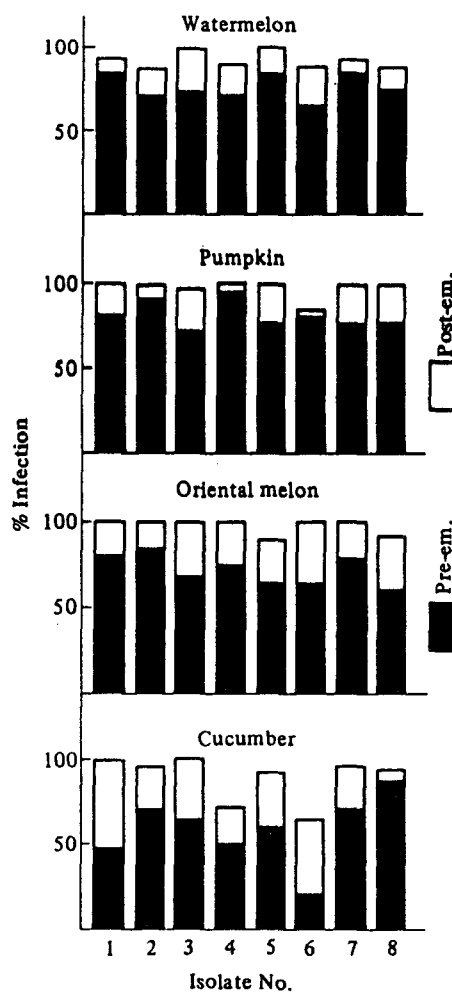


Fig. 2. Disease incidence of seedlings of cucumber, oriental melon, pumpkin and watermelon grown in soil inoculated with eight isolates of *Didymella bryoniae*. Pre-em.: Pre-emergence infection, Post-em.: Post-emergence infection.

symptoms which resembled those produced on growing-on test developed gradually from the initial part of infection to other parts of the plant.

DISCUSSION

In cross-inoculation tests, the eight isolates of *D. bryoniae* could infect cucumber, oriental melon, pumpkin and watermelon at different stages of growth. In early stage of infection, original host showed more severe symptoms than others when seed inoculation methods was used. There was not much differences

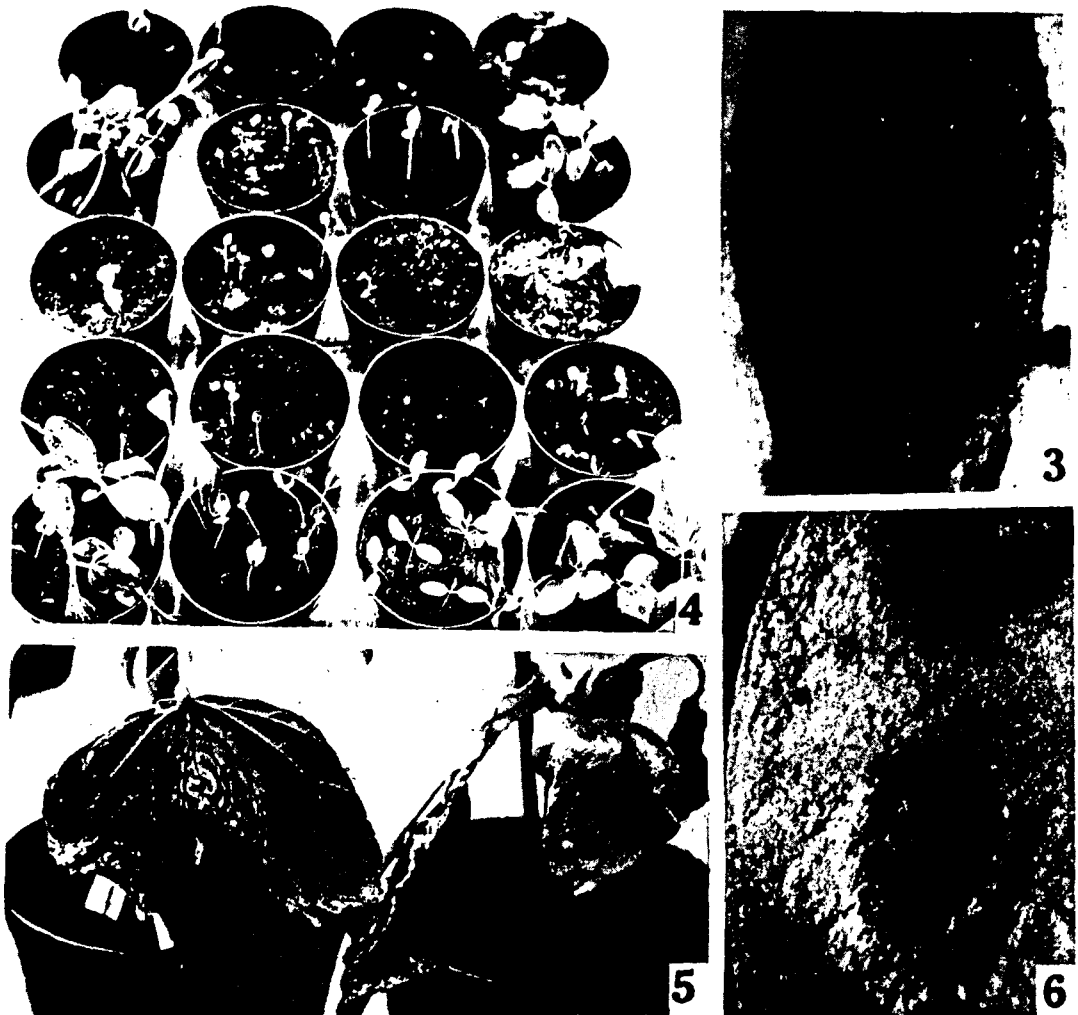


Fig. 3. Symptom of radicle rot with numerous pycnidia of *Didymella bryoniae* from cucumber seed.

Fig. 4. Reactions of cucurbits to isolates of *Didymella bryoniae* when soils were inoculated. From top to bottom: isolate 6, isolate 4, isolate 2, isolate 1 and control. From left to right: pumpkin, watermelon, oriental melon and cucumber.

Fig. 5. Symptoms of gummy stem blight developed on leaves and cotyledons of cucumber plants.

Fig. 6. Tan spots on abaxial surface of cotyledon of pumpkin seedling.

in pathogenicity or susceptibility between isolates of the pathogen and crops tested. Cotyledons, leaves and hypocotyl of cucumber, oriental melon, pumpkin and watermelon were very susceptible under wet conditions, but symptoms on cucumber and pumpkin were checked when the dry conditions maintained. Chiu and Walker(1) pointed out that cucumber and pumpkin were very resistant when they are in the seedling stage. Gindrat and Sijak(5) reported that *D. bryoniae* was highly virulent on cucumber, but weakly virulent on vegetable marrow. It has been found that this disease commonly occurred on cucum-

ber plants in green house (2,5). Fletcher and Preece(4) also noted that severe outbreak of this disease have often been associated with nurseries with poor heating systems where high humidities were maintained throughout the evening and night in greenhouse. With these observations it is considered that susceptibility of cucumber and pumpkin are mainly influenced by prevailing moisture conditions.

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