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Anthracnose of Cyclamen Caused by *Colletotrichum gloeosporioides* Penz.

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Colletotrichum gloeosporioides Penz.에 의한 시클라멘 탄저병

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ABSTRACT: Anthracnose symptoms were observed on the commercially cultivating cyclamen in Chonbuk province in 1995. The symptoms of infected flowers were small, circular and dark brown ring-spots or water-soaked lesions and gradually changed to black blight. Mycelial colony of the isolates was light or whitish gray to dark gray on potato dextrose agar. Conidia were straight cylindrical and obtuse at the apex and measured 8.3~15.0×2.5~6.3 μm in size. Appressoria were brown to dark brown and clavate, but most of them were irregular. Acervuli on lesions were brown, rounded and measured 50~140×32.5~90 μm in size. The optimum temperatures for mycelial growth and conidial sporulation were ranged from 25 to 30°C. Thus, based on mycological characteristics of the fungus, the causal agent of cyclamen anthracnose was identified as *Colletotrichum gloeosporioides* Penz.

***Key words*: Mycological characteristics, *Colletotrichum gloeosporioides*, cyclamen anthracnose**

Cyclamen, perennial bulbs plant belonged to primrose family, blooms from late October through April of the following year. The demand for cyclamen (*Cyclamen* spp.) as an ornamental decoration plant increases in winter season (14).

Lately, symptoms of anthracnose on cyclamen had been occurrred in Seoul and surrounding cities. Generally, the disease occurrs in leaves, stems and flowers resulted in loss of marketable plants. The infection rate of anthracnose on cyclamen was up to 20% in high temperature and humidity of vinyl house during July~August. Anthracnose has been known to be caused by a number of fungal species belonged to genus *Colletotrichum* (8, 11). The genus of *Colletotrichum* contains many species that cause anthracnose or blight on a wide range of ornamental plants. In Korea, anthracnose of ornamental plants was reported in 1991 and 1993 by Kim *et al* (7, 8) and anthracnose of statice was reported by Choi *et al* (3). In Japan, Okayama and Tsujimoto (13) found that *Glomerella cingulata* (*Colletotrichum gloeosporioides*) which isolated from strawberry anthracnose could infect the cyclamen. Kijima and Minegisi (6) reported that buds of cyclamen were infected by many pathogens including *Colletotrichum* spp. In India, the causal agent of cyclamen anthracnose was identified as *C. gloeosporioides* by Madhu-Meeta *et al* (12). Recently, In Japan, Kobayashi (9) was able to select the cyclamen varieties resistant to anthracnose, but the disease has not been reported in Korea.

Thus, this study was conducted to identify the causal agent associated with cyclamen anthracnose and examine the effect of temperature on the mycological characteristics of the fungus.

MATERIALS AND METHODS

Isolation and Preservation. The fungus was isolated from infected flowers of cyclamen which purchased from flower market in Iksan on December, 1995

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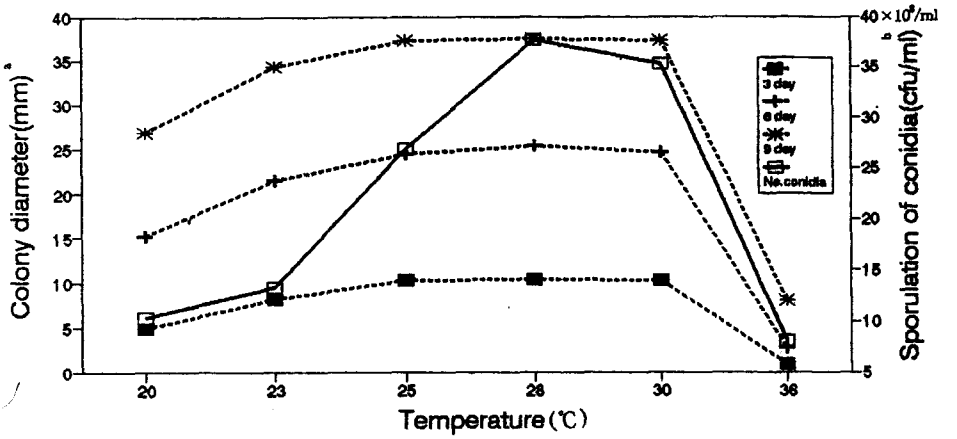

Fig. 1. Effect of temperature on mycelial growth and conidial sporulation of *Colletotrichum gloeosporioides* isolated from diseased cyclamen.

[a] Values are means of 3 replications and stanard deviation after 3, 6 and 9 days of incabation on PDA.

[b] After 10 days of incubation on PDA under different temperatures.

(Fig. 2-1). Cutted samples were surface-sterilized with 0.5% of NaOCl for 1 min., rinsed with sterilized distilled water in three times, placed on potato dextrose agar (PDA) and incubated at 25°C. Single spore was isolated by dilution method in water agar and mono-conidial culture was obtained by cultivating the single spore on PDA for 7 days at 25°C. The isolates were transferred to PDA slants and, kept at 5°C for preservation and further use.

Examination of Cultural and Morphological Characteristics. The characteristics of the isolates were examined after incubating at 25°C for 14 days on PDA and growth rate of mycelia were also investigated. The shape, size and color of conidia, appressoria and acervuli were also examined under the microscope and scanning electron microscope with 10 day old cultures. The size of conidia was measured using a micrometer with twenty conidia which taken from diseased tissues and cultures. Mycelial growth and sporulation of conidia were investigated at 20, 23, 25, 28, 30 and 36°C. A disc of 0.5 cm diameter from 10 day old culture was seeded in the center of petri plates containing PDA. Each experiment was replicated three times. The plates were placed in the pre-set temperature regimes and colony diameter was measured 3, 6 and 9 days after inoculation. Sporulation of conidia was examined with conidial suspension of 30 m*l* placed in incubator. The number of sporulating conidia per m*l* was counted with a hemocytometer.

Pathogenicity test. Isolates were cultured on PDA at 25°C for 14 days. Conidial suspension was prepared from PDA cultures. The concentration of conidia for inoculation was adjusted to 10^6 conidia per m*l*. For the test, 'unwounded' and 'wounded' sets of cyclamen plants (leaf, flower) were inoculated by spraying the spore suspension. Control plants were sprayed with sterilized distilled water. The inoculated flowers and leaves were wrapped with plastic bags and incubated at 25°C under 100% relative humidity. Readings of the symptoms were made about 2~3 days after inoculation.

RESULT AND DISCUSSION

Cultural and Morphological Characteristics. The fungus isolated from diseased cyclamen plant initially formed whitish-gray colony and gradually became dark gray colored and formed rounded ring on PDA. The colony also formed light orange-colored conidial mass in the center about 3 days after incubation and developed into black mass of acervuli. Conidia were aseptate, colorless, cylindrical and rounded at the apex and measured 8.3~15×2.5~6.3 μm in size (Fig. 2, 3). Setae and sclerotia were not found on PDA culture. Appressoria were brown cleavate-cylindrical and measured 10~12.5×5~7.5 μm in size (Fig. 2-5, 6). Acervuli were dark brown and measured 50~140×32.5~90 μm in size on diseased tissues (Table 1, Fig. 2-4).

The morphological characteristics of *Colletotrichum* sp. isolated from the diseased cyclamen were compared with the others (Table 1). In this study, the size

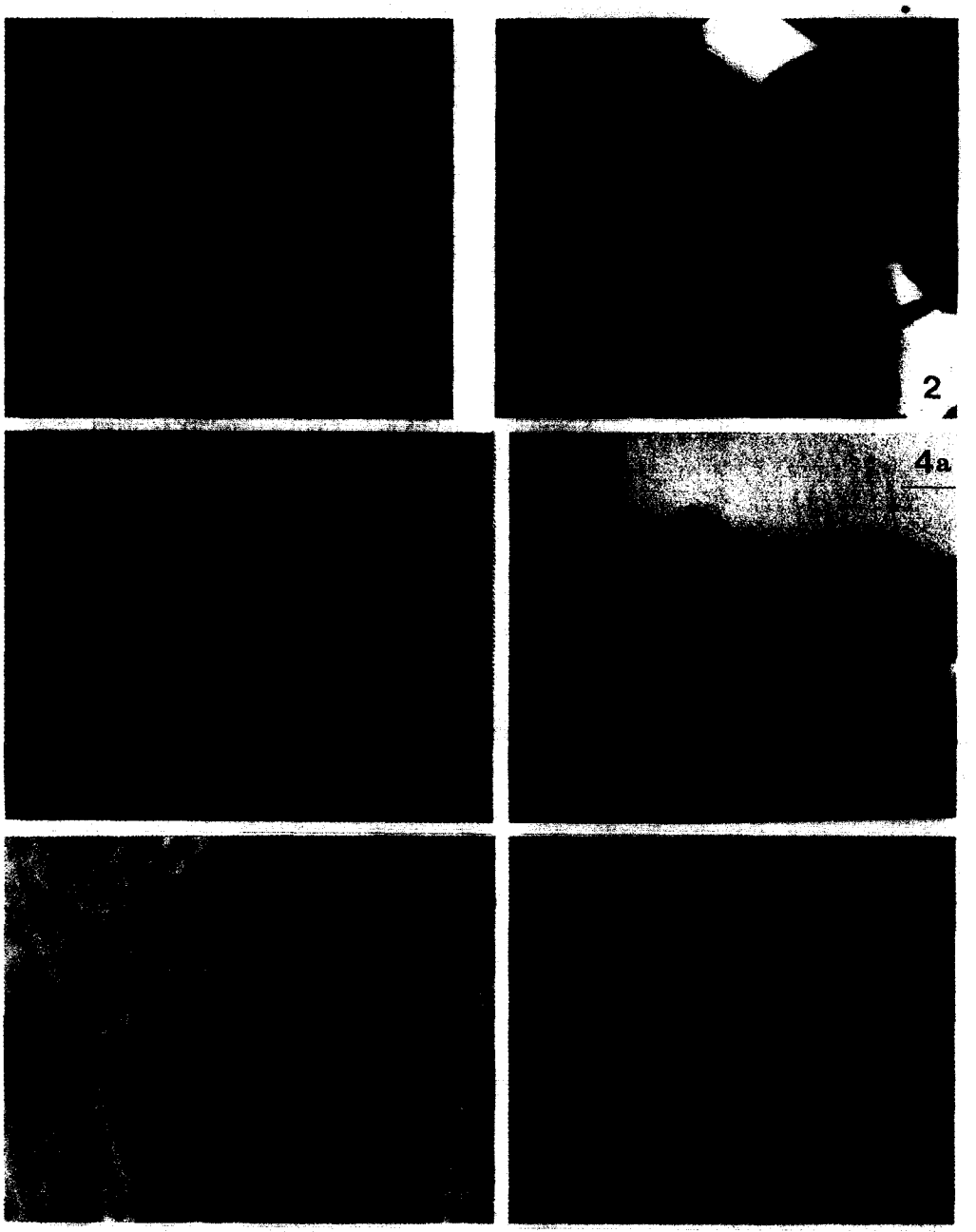

Fig. 2. Symptoms of anthracnose caused by ***Colletotrichum gloeosporioides*** in the flowers (Fig. 2-1), leaves lesion (Fig. 2-2). Photomicrograph (Fig. 2-3a) and scanning electron micrograph (Fig. 2-3b) of *C. gloeosporioides* conidia (Fig. 2-3) on PDA. Photomicrograph of the section (Fig. 2-4a) and surface (Fig. 2-4b) of *C. gloeosporioides* acervulus formed on diseased plants. Scanning electron micrograph (Fig. 2-5) and photomicrograph (Fig 2-6) of *C. gloeosporioides* appressorium (2-6a : irregular shape, 2-6b : clavate shape) formed from hyphae on PDA. The scale bars represent 10 μm (3a, 4a, 4b, 6a, 6b).

and shape of conidia, and presence of sclerotia and setae were consistent with the anthracnose pathogen of citrus and bean reported by by Chung and Koh (4) and Han and Lee (5), respectively. But the isolates did not

Table 1. Comparison of mycological characteristics of *Colletotrichum gloeosprioides* associated with cyclamen anthracnose

	Mycological characteristics[a]		
	This study	Han and Lee (5), Chung and Koh (4)	Sutton(16), Arx(1)
Conidia			
shape	cylindrical	cylindrical	straight obtuse at the apex (16) cylindrical (1)
color	colorless	colorless	–[b]
size	8.25~15×2.5~6.3 μm (12.27×4.25 μm)	7.5~17.8×3.5~5.6 μm (10.5×4.0 μm) (5) 14.4~19×4.6~6.0 μm (4)	9~24×3~4.5 μm (16) 10~21×4~6 μm (1)
Appressoria			
shape	clavate or irregular sometimes becoming complex	lobed, clavate sometimes becoming complex	clavate or irregular sometimes becoming complex
color	brown to dark brown	sepia brown (5)	–
size	10~12.5×5~7.5 μm (10.2×5.4 μm)	5.3~10.5×10.5~17.5 m (8.0×13.3 μm) (5) 6.5~11.3×5.0~8.5 μm (4)	6~20×4~20 μm
Acervulus			
shape	rounded or occasionally elongated, eruption		–
color	brown	pale pink (5), dark brown (4)	–
size	50~140×32.5~90 μm (86.1×61.1 μm)	57.7~86.6×72.2~122.7 μm 80~150×50~120 μm (4)	–
Setae	absent	absent (5), present (4)	absent
Sclerotia	absent	absent	absent

[a] After 10 days of incubation on PDA at 25°C.
[b] Not described.

form setae and sclerotia. Thus, the pathogen was identified as *C. gloeosporioides* because mycological characteristics were similar to those of reported by Sutton (17) and Arx *et al* (12). And the others of the characteristics, such as shape and color of apperssoria and size of acervuli were also similar to some other reports (4, 5, 10, 13).

Incubation Temperature. The temperature for the mycelial growth and conidial sporulation of *C. gloeosporioides* were ranged from 25°C to 30°C on PDA (Fig. 1). But, mycelial growth and conidial sporulation of the fungus were retarded at 20~23°C. The greatest retardment of the mycelial growth and conidia sporulation were observed at the higher temperature such as 36°C about 3, 6 and 9 days after inoculation. The results are consistent with the pathogen of citrus anthracnose and red pepper anthracnose reported by Chung and Koh (4) and Park *et al* (15), respectively.

Pathogenicity. The pathogenicity of *C. gloeosporioides* which isolated from diseased cyclamen plant was confirmed by artificial inoculation. Inoculation both unwounded and wounded flowers developed symptoms about 2~3 days after inoculation (Table 2). Initial symptoms were appeared on the flowers as small, circular, water-soaked lesions (Fig. 2-1). Lesions enlarged and became double ring spot or turned tan. Inoculation on the wounded plants developed small brown round spot which often expanded into ring spot on leaves about 3 days after inoculation (Fig. 2-2). But, No symptom

Table 2. Pathogenecity of *Colletotrichum gloeosporioides* on cyclamen plant

Part inoculated	Inoculation	
	Wounded	Unwounded
Leaf	+[a]	–[c]
Flower	++[b]	++

[a] ++ : severe symptom
[b] + : mild symptom
[c] - : no symptom

was appeared on the leaves of unwounded-inoculated plants. This result showed that *C. gloeosporioides* was not able to penetrate the wax layer of the inoculated plants and similar result was obtained by Okayama and Tsujimoto (13).

Pests have been the problems for commercial production of cyclamen since cyclamen was the perrenial ornamental plant. In the field, infection rate of cyclamen anthracnose was up to 20%, 90% and 30% in overall, leaves and flowers, respectively (8). Thus, to find a successfully methods for controlling cyclamen anthracnose, it is necessary to study the physiological and ecological characteristics of the pathogen. Especially, anthracnose formed symptoms not only on flowers, but also on petioles, surfaces of bulb, young buds, occurrence of anthracnose would be observed in all steps of plant growth. Since no study was done on the the host range of *C. gloeosporioides*, it is also necessary to study the host range of the fungus. Thus, integrated disease management techniques such as sanitation of field should be developed.

요 약

1995년 12월 전라북도 지역 내에 유통되고 있는 시클라멘의 꽃잎 중앙부에 진회색~흑색 원형 반점을 형성하거나, 꽃잎 주변에 흑색 수침상을 나타내는 탄저병의 발생이 관찰되었다. 병반으로 부터 분리된 균은 PDA 배지상에서 회백색을 띄었으며, 8.3~15×2.5~6.3 μm 크기의 양끝이 둥근 무색, 단포의 원통형~타원형의 분생포자를 형성하였고, 갈색 곤봉형의 부착기와 이병조직에서 갈색 둥근형의 분생자층을 형성하였다. 이에 따라 시클라멘 탄저병징에서 분리한 병원균은 국내에서 미보고된 *Colletotrichum gloeosporioides*로 동정되었으며, 균사생장 및 분생포자 형성을 위한 배양적정 온도는 25~30°C였다.

REFERENCES

1. Arx, J. A. von. 1981. *The genera of fungi sporulating in pure culture.* (3rd. ed. J. Cramer. Vaduz.) pp.168-169, 215-220.
2. Arx, J. A. von. 1987. *Plant pathogenic fungi.* Gebruder Borntraeger, Berlin. pp.218-220.
3. Choi, J. S., Cheong S. S., Kim, J. M. and So, I. Y. 1994. Anthracnose of statice caused by *Glomerella cingulata* (Stonem.) Spaulding & Schrenk in Chonbuk Province. *Korean J. Plant Pathol.* 10(2) : 148-150.
4. Chung, H. J. and Koh, Y. J. 1992. Anthracnose of citron caused by *Colletotrichum gloeosporioides* Penz. *Korean J. Plant Pathol.* 8(1) : 70-74.
5. Han, K. S. and Lee, D. H. 1995. Identification and etiological characteristics of anthracnose fungi isolated from soybean, small red bean and green bean. *Korean J. Plant Pathol.* 11(1) : 30-38.
6. Kijima, T. and Minegisi, N. 1985. Occurrence of new pathogens of bud of blight of cyclamen. *Bulletin Tochigi-Agri.-Exp.-Stat.* 31 : 89-101.
7. Kim, W. G., Cho, W. D., Lee, Y. H. and Lee, E. J. 1991. Anthracnose of ornamental plants caused by *Colletotrichum gloeosporioides* Penz.. *RDA J. Agri. Sci.* 33(2) : 20-25.
8. Kim, W. G., Cha, Y. S., Cho, W. D. and Lee, Y. H. 1993. Occurrence of anthracnose on six kinds of ornamental plants caused by *Colletotrichum gloeosporioides. RDA. J. Agri. Sci.* 35(2) : 327-330.
9. Kobayashi, N. 1994. Variety resistance to cyclamen anthracnose, characteristics of the pathogen infection, dispersal and susceptibility to agricultural chemicals. *Proceedings of the association for plant protection of Kyushu.* 40 : 75-81.
10. Lee, D. H. 1994. Etiological characteristics of *Colletotrichum acutatum, C. gloeosporioides* and *Glomerella cingulata* isolated from apples. *Korean J. Plant pathol.* 10(2) : 105-111.
11. Lee, E. J., Lee, K. H., Cho, W. D., Kim, W. G and Ryu, H. Y. 1989. *Compendium of ornamental plant diseases with colour plates.* Agricultural Siences Institute. 210pp.
12. Madhu-M. Madhu-B. Meeta-M and Bhatia-M. 1992. Some new host records on ornamental plants. *Indian Phytopathology* 45(2) : 279-280.
13. Okayama, K. and Tsujimoto, A. 1994. Occurrence of strawberry anthracnose caused by *Glomerella cingulata* (Stoneman) Spaulding et Schrenk and pathogenicity of the fungus. *Ann. Phytopathol. Soc. Japan* 60(5) : 617-623.
14. Park, I. H., Song, K. T., Lee, C. Y. and Lee, M. W. 1979. *The particulars floricultures. Cyclamen* spp. Sunginmunhwasa, Seoul. pp.265-272.
15. Park, K. S. and Kim, C. H. 1994. Effect of temperature, relative humidity, and free water period on lesion development and acervulus formation of *Colletotrichum gloeosporioides* on red pepper. *Korean J. Plant Pathol.* 10(1) : 34-38.
16. Sutton, B. C. 1980. *The Coeolomycetes.* Commonwealth Mycological Institute, Kew, England. pp. 523-537.

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