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Occurrence of Sclerotinia Rot in Cruciferous Crops Caused by *Sclerotinia* spp.

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Cruciferous crops grown in greenhouses and fields in Korea were surveyed from 1995 to 2000. Sclerotinia rot most severely occurred up to 30% in cabbage. Incidence of the disease was as high as 20% at its maximum in Chinese cabbage and rape and 10% in radish, but as low as less than 1 or 2% in broccoli and kale. Symptoms of Sclerotinia rot commonly developed on leaves and stems of the crucifers, but rarely on rachises of broccoli. A total of 112 isolates of *Sclerotinia* species was obtained from the diseased crucifers. Out of the isolates, 103 isolates were identified as *S. sclerotiorum,* and the rest as *S. minor* based on their morphological and cultural characteristics. *S. sclerotiorum* was isolated from all the crucifers, while *S. minor* was isolated from Chinese cabbage, broccoli, and kale. Six isolates of *S. sclerotiorum* and three isolates of *S. minor* were tested for their pathogenicity to the crucifers by artificial inoculation. All the isolates of the two *Sclerotinia* spp. induced rot symptoms on the plants of the crucifers tested, which were similar to those observed in the fields. The pathogenicity tests revealed that there was no significant difference in the susceptibility of the crucifers to the isolates of *S. sclerotiorum.* However, in case of *S. minor,* radish was relatively less susceptible to the pathogen.

Keywords : Crucifer, pathogenicity, Sclerotinia rot, *Sclerotinia sclerotiorum, S. minor.*

Cruciferous crops are cultivated worldwide, and most are considered as important vegetables in many countries. They are mostly grown in cool conditions, which are favorable for the occurrence of Sclerotinia rot on the crops (Purdy, 1979; Willetts and Wong, 1980). In Korea, some cruciferous crops including Chinese cabbage are cultivated all over the country regardless of season. Severe outbreaks of rot symptoms with sclerotial formation were observed during a disease survey of cruciferous crops in several locations in the country. *Sclerotinia* spp. were consistently isolated from the diseased plant parts. The disease of the crops caused by *Sclerotinia* spp. has been generally called as Sclerotinia rot (Dillard and Hunter, 1986; Kim and Cho, 1998; Cho et al., 1997). The disease was also recorded as Sclerotinia stem rot, white blight, watery soft rot, or crown rot depending on the species of the crucifers (Farr et al., 1989).

It has been reported that *Sclerotinia sclerotiorum* (Lib.) de Bary and *S. minor* Jagger cause Sclerotinia rot in some cruciferous crops (Boland and Hall, 1994; Dillard and Hunter, 1986; Farr et al., 1989; Koike et al., 1994; Melzer et al., 1997; Purdy, 1979; Tai, 1979). In Korea, Cho et al. (1997) briefly described symptomatical and developmental characteristics of the disease in five cruciferous vegetable crops with some mycological characteristics of the causal *Sclerotinia* species. Kim and Cho (1998) also reported some mycological and pathological characteristics of *S. sclerotiorum* and *S. minor* causing Sclerotinia rot of vegetable crops. Few studies have been conducted on the detailed characteristics of the disease occurrence and the pathogenicity of the causal *Sclerotinia* species in cruciferous crops. This study was conducted to reveal trends of the disease occurrence in cruciferous crops in Korea, and the pathological aspects of the causal *Sclerotinia* spp.

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Materials and Methods

Field survey and collection of diseased samples. Cruciferous crops grown in greenhouses and fields in Korea were surveyed from 1995 to 2000. Incidence of Sclerotinia rot on the crops was investigated, and diseased plants were collected. The severity of the disease was rated in terms of percentage of infected plants among 50-100 plants observed with three replicates in each infected field.

Isolation. *Sclerotinia* spp. were isolated from the lesions according to the method described previously (Kim et al., 1999). Nine to 25 mm^2 lesion pieces cut from the diseased plant parts were placed on 2% water agar (WA) after surface-sterilizing with 1% sodium hypochlorite solution for 1 minute. The plates were incubated for 1-2 days at 22°C. The fungi grown from the lesion pieces were transferred to potato dextrose agar (PDA) slants and cultured for identification.

Examination of morphological and cultural characteristics. Each isolate was cultured on PDA in 9-cm-diameter petri dishes at 22°C in the dark for 20 days for the production of sclerotia.

Sclerotia produced on the medium were examined for morphological characteristics, and were preserved in a low temperature incubator at 0°C for 1 month. Then, the sclerotia were placed in 250-ml flasks with sterile wet sand and incubated at 15°C for 1-5 months in alternating cycles of 12-hour fluorescent light and 12-hour darkness to induce the formation of apothecia. Apothecia produced from the sclerotia during the incubation were collected and examined for the morphological features. Nuclei in the ascospores were stained with DAPI (Martin, 1987), and were observed under a fluorescent light microscope.

Colony morphology of each isolate on PDA was examined 12 days after incubation at 22°C in the dark. Temperature range for mycelial growth of the isolates was examined in PDA culture. Optimum temperature was determined based on the mycelial growth length of the isolates per 24 hours.

Pathogenicity test. One cultivar each of cruciferous crops such as Chinese cabbage [*Brassica campestris* L. ssp. *pekinensis* (Lour.) Olss.], cabbage (*B. oleracea* L. var. *capitata* L.), radish (*Raphanus sativus* L.), broccoli (*B. oleracea* L. var. *botrytis* L.), kale (*B. oleracea* L. var. *acephala* DC.), and rape (*B. napus* L.) was used for the pathogenicity tests. Seeds of each crucifer were sown in a plastic pot (29 cm in height and 21 cm in diameter) filled with sterile soil in a greenhouse at 18-28°C. Seedlings of each crucifer were transplanted into new plastic pots filled with sterile soil 15 days after sowing and were cultivated in the greenhouse.

One isolate of each *Sclerotinia* species from each host was used for the pathogenicity tests to the hosts. Fresh mycelial mats (6 mm in diameter) of each isolate grown on PDA were placed on the leaves or stems of 5-10 cm above ground of 53-day-old host plants grown in the plastic pots. PDA disks of the same size were placed on the leaves or stems of the plants as control. The pots with inoculated plants were placed in dew chambers with 100% relative humidity at 22°C for 48 hours and then moved into the greenhouse. Virulence of the isolates was rated based on the degree of rot symptoms induced 22 days after inoculation. The inoculation test was performed with three replicates.

Results

Disease incidence and symptoms. Sclerotinia rot commonly occurred in six cruciferous crops grown in several locations in Korea (Table 1). Occurrence of the disease was observed in 5 of 52 Chinese-cabbage-growing locations, 1 of 16 cabbage-growing locations, 2 of 40 radish-growing locations, 3 of 5 broccoli-growing locations, 2 of 3 kale-growing locations, and in 1 rape-growing location surveyed during the growing seasons. The disease severely occurred up to 30% in cabbage. Incidence of the disease was as high as 20% at its maximum in Chinese cabbage and rape and 10% in radish, but as low as less than 1 or 2% in broccoli and kale.

Symptoms of Sclerotinia rot commonly developed on leaves and stems of the crucifers but rarely on rachises of broccoli (Fig. 1). Infected plant parts rotted with white to gray yellow discoloration. Cottony mycelia frequently developed on the infected plant parts. Globose to irregular black sclerotia were produced on the infected plant parts at the late stages of the disease development.

Isolation and identification. A total of 112 isolates of *Sclerotinia* species was obtained from lesions of Sclerotinia rot in six cruciferous crops (Table 2). The *Sclerotinia* species were isolated from leaves of Chinese cabbage, cabbage, radish and broccoli, and stems of radish, broccoli, kale, and rape. The *Sclerotinia* species were also isolated from rachises of broccoli. Out of the 112 isolates, 103 isolates were identified as *S. sclerotiorum,* and the rest as *S. minor* based on their morphological and cultural characteri-

Table 1. Incidence of Sclerotinia rot in cruciferous crops in Korea from 1995 to 2000

Host (No. of locations surveyed)	Location where *disease occurred*	No. of fields *investigated*	No. of fields *infected*	% infected plants[a]
Chinese cabbage (52)	Gwangjugun	18	8	less than 2
	Namwon	21	2	less than 2
	Namyangju	15	2	less than 2
	Wanju	28	4	5-20
	Wonju	18	2	less than 1
Cabbage (16)	Jeju	51	8	5-30
Radish (40)	Suwon	15	1	less than 2
	Yeoju	18	3	1-10
Broccoli (5)	Hoengseong	5	1	less than 1
	Hwaseong	4	1	less than 1
	Namyangju	3	1	less than 1
Kale (3)	Namyangju	22	5	less than 2
	Suwon	1	1	less than 1
Rape (1)	Jeju	12	3	1-20

[a]Fifty to 100 plants in each infected field were investigated with three replicates.

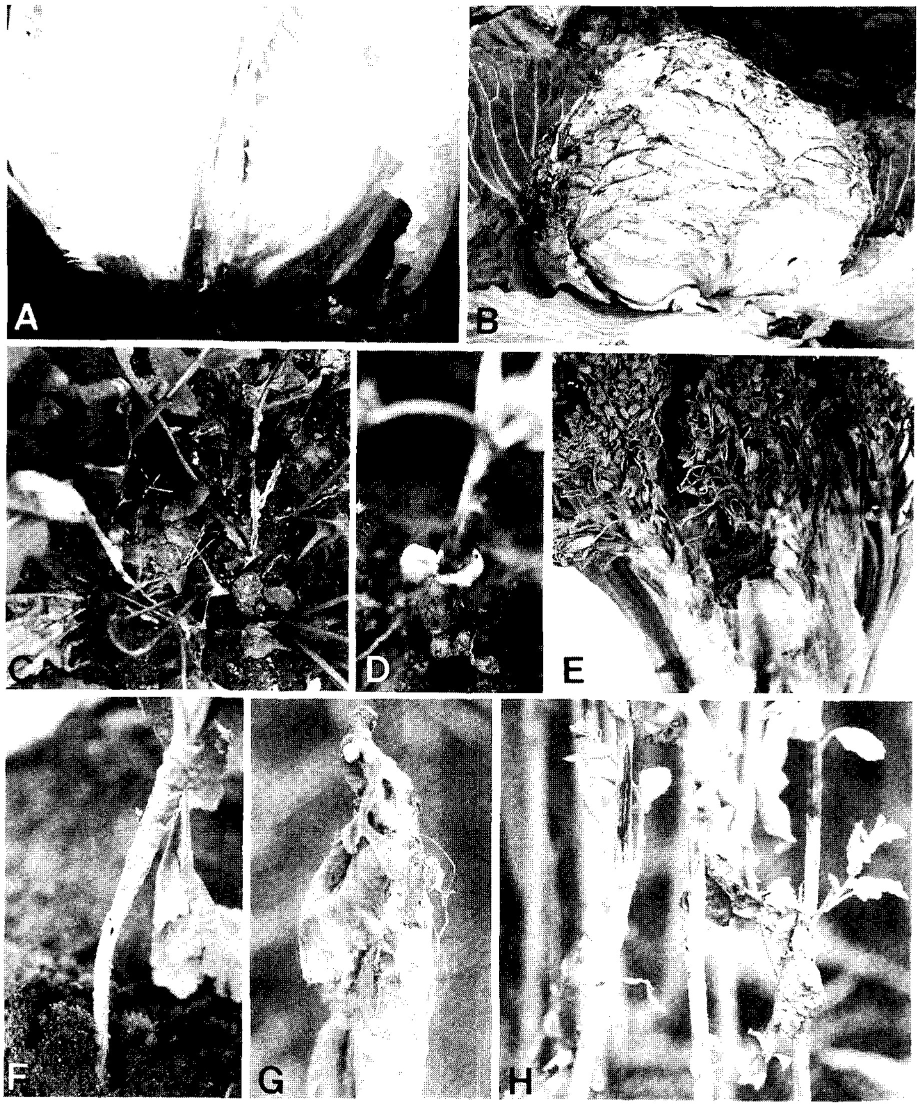

Fig. 1. Symptoms of Sclerotinia rot in six cruciferous crops grown in the fields. **A**, Chinese cabbage; **B**, cabbage; **C**, radish; **D** and **E**, broccoli; **F** and **G**, kale; **H**, rape.

Table 2. Isolation and identification of *Sclerotinia* spp. from diseased plant parts of six cruciferous crops

Host	No. of isolates obtained				No. of isolates identified	
	Leaf	Stem	Rachis	Total	*S. sclerotiorum*	*S. minor*
Chinese cabbage	32	0	0	32	27	5
Cabbage	13	0	0	13	13	0
Radish	8	6	0	14	14	0
Broccoli	1	14	2	17	16	1
Kale	0	18	0	18	15	3
Rape	0	18	0	18	18	0
Total	54	56	2	112	103	9

stics as described by previous workers (Jagger, 1920; Kohn, 1979a; Willetts and Wong, 1980). *S. sclerotiorum* was isolated from all the crucifers, and *S. minor* from Chinese cabbage, broccoli, and kale.

Morphological and cultural characteristics. Colonies of *S. sclerotiorum* on PDA consisted of white to gray mycelia,

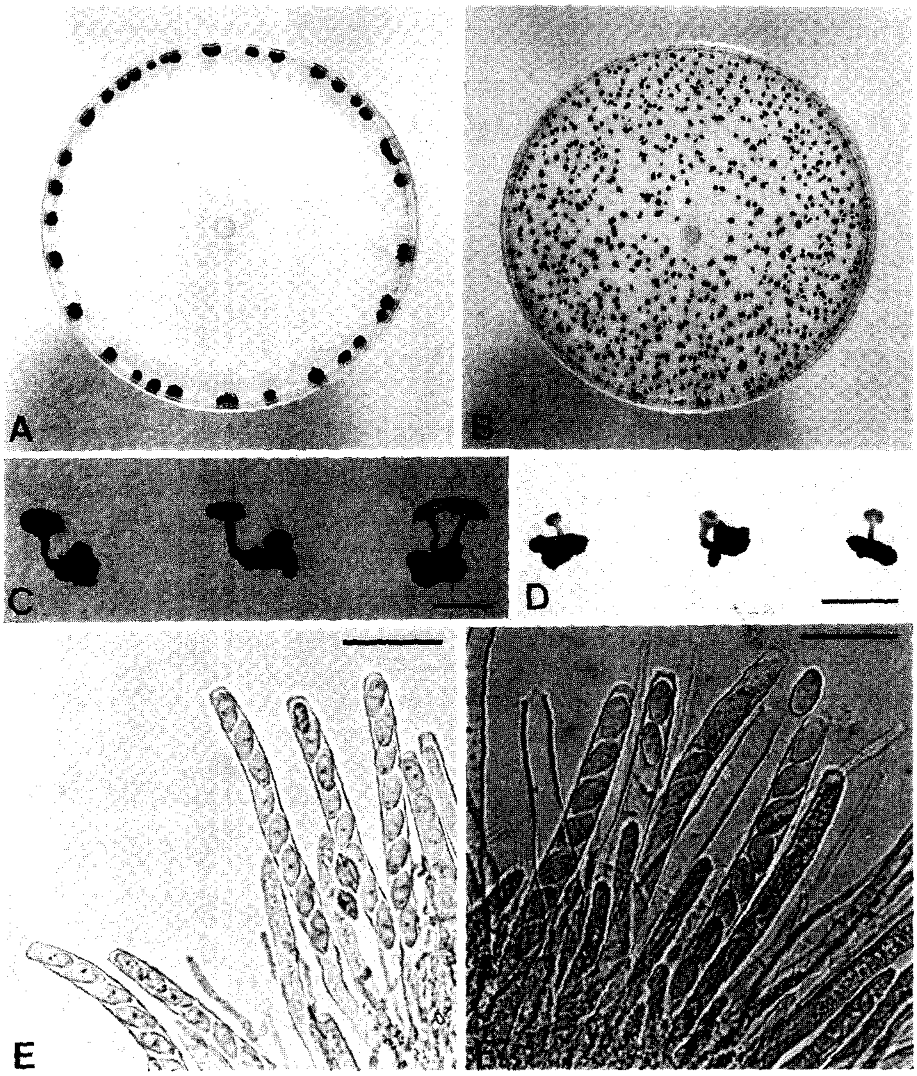

Fig. 2. Cultural and morphological features of *Sclerotinia sclerotiorum* (A, C, and E) and *S. minor* (B, D and F) isolated from cruciferous crops. **A** and **B**, 12-day-old colonies on PDA at 22°C in the dark; **C** (scale bar=5 mm) and **D** (scale bar=4 mm), apothecia produced from sclerotia; **E** and **F**, asci and ascospores (scale bar=30 μm).

and globose to irregular and black sclerotia (Fig. 2A). *S. minor* produced a lot of sclerotia on the medium, which were small, globose to irregular, and black (Fig. 2B). A sclerotium of *S. sclerotiorum* produced one to several apothecia (Fig. 2C), and that of *S. minor* mostly produced only one (Fig. 2D). Asci of *S. sclerotiorum* and *S. minor* were very similar, which were cylindrical and 8-spored (Fig. 2E and F). Ascospores of the two species were also very similar, which were hyaline, ellipsoid to ovoid. The morphological and cultural characteristics of the two species differed in terms of size of sclerotia and apothecia, and number of sclerotia formed on the medium (Table 3).

Pathogenicity. Six isolates of *S. sclerotiorum* and three isolates of *S. minor* induced rot symptoms on the six cruciferous crops inoculated (Table 4). The symptoms were similar to those observed in the fields. *Sclerotinia* spp. were re-isolated from the lesions of the plants inoculated. The pathogenicity tests revealed that there was no significant difference in virulence between the isolates of *S. sclerotiorum* and *S. minor* to the crucifers. Also, there was no significant difference in the susceptibility of the crucifers to the isolates of *S. sclerotiorum*. However, in case of *S. minor*, radish was relatively less susceptible to the pathogen.

Discussion

S. sclerotiorum and *S. minor* are known to cause Sclerotinia rot in a variety of crops, and the latter has somewhat narrower host range than the former (Kohn, 1979a; Kohn, 1979b; Purdy, 1979; Willetts and Wong, 1980). It has been

Table 3. Morphological and cultural characteristics of *Sclerotinia sclerotiorum* and *S. minor* isolated from cruciferous crops

Examination	Division	Description of characteristics	
		S. sclerotiorum	*S. minor*
Sclerotium	Formation	15-40/Petri dish	500-800/Petri dish
	Color	Black	Black
	Shape	Globose to irregular	Globose to irregular
	Size	0.6-10.0×0.6-6.5 mm	0.5-3.6×0.5-2.2 mm
Apothecium	Formation	1-8/sclerotium	1-2/sclerotium
	Shape	Cup-shaped	Cup-shaped
	Diameter of disks	1.5-7.2 mm	0.9-2.9 mm
	Length of stalks	1.6-8.0 mm	1.1-4.0 mm
	Width of stalks	0.5-1.0 mm	0.2-0.5 mm
Ascus	Shape	Cylindrical	Cylindrical
	Size	110-162×8-11 μm	122-178×8-12 μm
Ascospore	Shape	Ellipsoid to ovoid	Ellipsoid to ovoid
	Size	10-16×4-8 μm	10-20×5-9 μm
	No. of nuclei	2	4
Mycelial growth	Minimum temperature	1°C	0°C
	Maximum temperature	30°C	29°C
	Optimum temperature	22-24°C	20-22°C

Table 4. Pathogenicity of isolates of *Sclerotinia sclerotiorum* and *S. minor* in six cruciferous crops by artificial inoculation

Sclerotinia species	Isolate	Isolate source	Virulence of isolates on plants (cultivars[a])[b]					
			Chinese cabbage (Jangmi)	Cabbage (Ohzora)	Radish (Taeyang)	Broccoli (HN)	Kale (DS)	Rape (NW)
S. sclerotiorum	S96-33	Kale	++	++	++	++	++	++
	S97-80	Broccoli	++	++	++	++	+	+
	S97-200	Chinese cabbage	++	+	+	++	+	+
	S97-210	Cabbage	++	++	+	++	++	+
	S97-223	Radish	++	++	+	++	++	++
	S98-17	Rape	++	++	++	++	++	++
S. minor	S96-22	Chinese cabbage	++	++	+	++	+	++
	S96-224	Broccoli	++	++	+	++	+	++
	S97-75	Kale	++	++	+	++	++	+
Control			-	-	-	-	-	-

[a] HN, DS, and NW represent abbreviations for seed companies Heungnong, Daeshin, and Nongwoo, respectively, producing unknown cultivars of the tested plants.

[b] Disease severity was rated 22 days after inoculation. ++ = above 2.0 cm of lesion length or wholly rotted; + = 0.5-2.0 cm of lesion length; – = no symptom.

reported that *S. sclerotiorum* causes Sclerotinia rot on several crucifers (Boland and Hall, 1994; Cho et al., 1997; Dillard and Hunter, 1986; Farr et al., 1989; Purdy, 1979; Tai, 1979). However, there has been little known on the disease occurrence of crucifers caused by *S. minor*. The present study showed that *S. sclerotiorum* and *S. minor* are associated with the occurrence of Sclerotinia rot in six cruciferous crops in Korea. *S. minor* was only isolated from Chinese cabbage, broccoli, and kale inducing typical Sclerotinia rot symptoms in all the plants by artificial inoculation, while *S. sclerotiorum* was isolated from all the crucifers. Based on the pathogenicity test, *S. minor* may have caused the disease in cabbage, radish, and rape in the field although it was not isolated from the three crops.

There was no information on the sclerotial density of *S. sclerotiorum* and *S. minor* in the fields of the cruciferous crops. It was assumed that more *S. minor* sclerotia are distributed in the fields than those of *S. sclerotiorum* because the former produces small and numerous sclerotia on the hosts like that on the medium. However, *S. sclerotiorum* was more frequently isolated from the diseased crucifers than *S. minor*, suggesting that the sclerotial viability of *S.*

minor was lower than that of *S. sclerotiorum* in the field. As suggested previously (Kim and Cho, 2002), further study is needed to clarify sclerotial viability of the two species in the field.

Price and Calhoun (1975) reported that there are differences in virulence of *S. sclerotiorum* isolates to individual hosts and in susceptibility of the host plants to different isolates. Kim et al. (1999) found that there are some differences in virulence of *S. sclerotiorum* isolates to cucurbitaceous vegetable crops and in susceptibility of some of the crops to the isolates. It has also been reported that there are differences in susceptibility of cultivars or lines of some crops to the fungus (Cassells and Walsh, 1995; Grau and Bissonnette, 1974; Orellana, 1975; Porter et al., 1975). However, the present study showed that there was no significant difference in virulence of the isolates of *S. sclerotiorum* to cruciferous crops, as recently reported by Kim and Cho (2002) in composite vegetable crops. On the other hand, *S. minor* was less virulent on radish than on other crucifers. Consequently, it was considered that there might be some differences in susceptibility of the crops to the isolates of *S. minor*. Further study is needed to reveal resistance of radish using more varieties.

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