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Occurrence of Sclerotinia Rot on Composite Vegetable Crops and the Causal *Sclerotinia* spp.

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Composite vegetable crops grown in greenhouses and open fields in Korea were surveyed from 1995 to 1999. Occurrence of Sclerotinia rot was observed in 123 of 277 lettuce fields, in 11 of 18 head lettuce fields, in 12 of 14 endive fields, and in 4 of 38 garland chrysanthemum fields surveyed during the growing seasons. The disease most severely occurred up to 80% on lettuce. Incidence of the disease was as high as 20% at its maximum on endive and garland chrysanthemum but relatively low on head lettuce. Symptoms of the disease developed on leaves of all the composites, crowns of lettuce and head lettuce, and stems of garland chrysanthemum. Out of 240 isolates of *Sclerotinia* species obtained from the diseased composites, 169 isolates were identified as *Sclerotinia sclerotiorum*, and the others as *S. minor* based on their morphological and cultural characteristics. *S. sclerotiorum* was isolated from all the composites, and *S. minor* only from lettuce and endive. Eight isolates of *S. sclerotiorum* and four isolates of *S. minor* were tested for their pathogenicity to the composites by artificial inoculation. All the isolates of the two *Sclerotinia* spp. induced rot symptoms on the plants of the composites by artificial inoculation, which were similar to those observed in the fields. The pathogenicity tests revealed that there is no significant difference in virulence of the isolates to the composites and in susceptibility of the composites to the isolates.

KEYWORDS: Composite, pathogenicity, Sclerotinia rot, *Sclerotinia sclerotiorum*, *S. minor*

Composite vegetable crops including lettuce are very popular as a fresh vegetable in the world. In Korea, they are cultivated in greenhouses during cool and cold seasons and in open fields in alpine areas during the summer. The cool and humid conditions are favorable for occurrence of Sclerotinia rot from the early to the late growing stages of the composites. The disease very commonly occurs on the composites in greenhouses because of the cool and humid conditions in the microclimate. Continuous cropping of the composites also results in a cause of severe outbreaks of the disease due to the increased density of sclerotia in the field. It was reported that the disease mostly occurs on leaves of composite vegetable crops, and the disease name was also recorded as Sclerotinia drop rot, drop, decay, or watery soft rot depending on the hosts (Jagger, 1920; Farr *et al.*, 1989). In general, the disease is often called Sclerotinia rot (Kim and Cho, 1998).

It has been reported that *Sclerotinia sclerotiorum* (Lib.) de Bary and *S. minor* Jagger cause sclerotinia rot on some composite vegetable crops (Boland and Hall, 1994; Farr *et al.*, 1989; Melzer *et al.*, 1997; Purdy, 1979). Recently Li *et al.* (2000) reported that *Sclerotinia nivalis* I. Saito also causes the disease on lettuce. In Korea, Cho *et al.* (1997) briefly described symptomatical and developmental characteristics of the disease on four composite vegetable crops and 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. Little studies have been conducted on detailed characteristics of the disease occurrence and pathogenicity of the causal *Sclerotinia* species on composite vegetable crops. This study was conducted to reveal trends of the disease occurrence on the composite vegetable crops in Korea, and etiological and mycological aspects of the causal *Sclerotinia* spp.

Materials and Methods

Field survey and collection of diseased samples. Composite vegetable crops grown in greenhouses and open fields in Korea were surveyed from 1995 to 1999. Incidence of Sclerotinia rot on the crops was investigated, and diseased plants were collected. Severity of the disease was rated in terms of percentage of infected plants among 100 plants observed with three replicates in each 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 plated on 2% water agar (WA) after surface-sterilizing with 1% sodium hypochlorite solution for one min. The fungus was isolated from the lesion pieces after one to two days of incubation at 22°C. The isolates 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-diame-

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ter petri dishes at 22°C in the dark for 20 days for production of sclerotia. Sclerotia produced on the medium were examined for the morphological characteristics and preserved in a low temperature incubator at 0°C for one month. The sclerotia were put in 250 m*l*-flasks with sterile wet sand and incubated at 15°C for one to five months in alternating cycles of 12 hr fluorescent light and 12 hr darkness. 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 observed under a fluorescent microscope.

Colonies of each isolate on PDA were examined 12 days after incubation at 22°C in the dark. Temperature range for mycelial growth of the isolates was examined in PDA culture. Linear length of mycelial growth of the isolates per 24 hr at 22°C was measured. Microconidia produced in PDA and WA culture at room temperature were examined.

Pathogenicity test. Two cultivars Cheongchima and Jeokchima of lettuce (*Lactuca sativa* L.) and one cultivar of head lettuce (*L. sativa* L. var. *capitata* L.), endive (*Cichorium endivia* L.) and garland chrysanthemum (*Chrysanthemum coronarium* L.) were used for pathogenicity tests. Seeds of each cultivar were sown in a plastic pot (29 cm in height and 21 cm in diameter) filled with sterile soil in a greenhouse at 18~30°C. Seedlings of each cultivar were transplanted into new plastic pots filled with sterile soil 18 days after sowing and cultivated in the greenhouse.

Two isolates of each *Sclerotinia* species from each host were used for inoculation tests to the hosts. Fresh mycelial mats of 6 mm in diameter from PDA cultures of each isolate were placed on the leaves or stems of 5 to 10 cm above ground of 2-month-old host plants grown in the plastic pots. PDA disks of the same size were placed on the leaves or stems of control plants. The pots with inoculated plants were placed in dew chambers with 100% relative humidity at 22°C for 48 hr and then moved into the greenhouse. Virulence of the isolates was rated based on the degree of rot symptoms induced ten days after inoculation. The inoculation test was performed in three replicates.

Results

Disease incidence and symptoms. Sclerotinia rot very commonly occurred on four composite vegetable crops grown in many locations in Korea (Table 1). Occurrence of the disease was observed in 123 of 277 lettuce fields, in 11 of 18 head lettuce fields, in 12 of 14 endive fields, and in 4 of 38 garland chrysanthemum fields surveyed during the growing seasons. The disease most severely occurred up to 80% on lettuce. Incidence of the disease was as high as 20% at its maximum on endive and garland chrysanthemum but relatively low on head lettuce.

Table 1. Incidence of Sclerotinia rot on composite vegetable crops in Korea from 1995 to 1999

Host	Location	No. of fields surveyed	No. of fields infected	% infected plants
Lettuce	Chungju	56	16	5~80
	Daejeon	8	0	0
	Goyang	16	0	0
	Gwangju	49	26	1-80
	Icheon	5	0	0
	Incheon	5	0	0
	Namwon	1	1	less than 1
	Namyangju	13	13	5~40
	Wanju	61	38	1~20
	Yangju	5	0	0
	Yangpyeong	55	29	1~30
	Yeoju	3	0	0
Head lettuce	Gimhae	11	9	1~5
	Hoengseong	1	0	0
	Hongcheon	2	0	0
	Pocheon	1	0	0
	Pyeongchang	3	2	less than 1
Endive	Gwangju	2	0	0
	Namyangju	8	8	1~20
	Yangpyeong	4	4	2~20
Garland chrysanthemum	Cheongwon	9	1	less than 1
	Chungju	6	3	5~20
	Goyang	8	0	0
	Gwangju	3	0	0
	Namyangju	2	0	0
	Yangpyeong	10	0	0

Symptoms of Sclerotinia rot developed on leaves of all the composites, crowns of lettuce and head lettuce, and stems of garland chrysanthemum (Fig. 1). Infected leaves and stems rotted with white to grayish 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 totai of 240 isolates of *Sclerotinia* species was obtained from lesions of Sclerotinia rot on four composites (Table 2). The *Sclerotinia* species were most frequently isolated from leaves of lettuce, head lettuce and endive, and stems of garland chrysanthemum. Out of the 240 isolates, 169 isolates were identified as *S. sclerotiorum*, and the others as *S. minor* according to the morphological and cultural characteristics described by previous workers (Kohn, 1979a; Willetts and Wong, 1980). *S. sclerotiorum* was isolated from all the composites, and *S. minor* only from lettuce

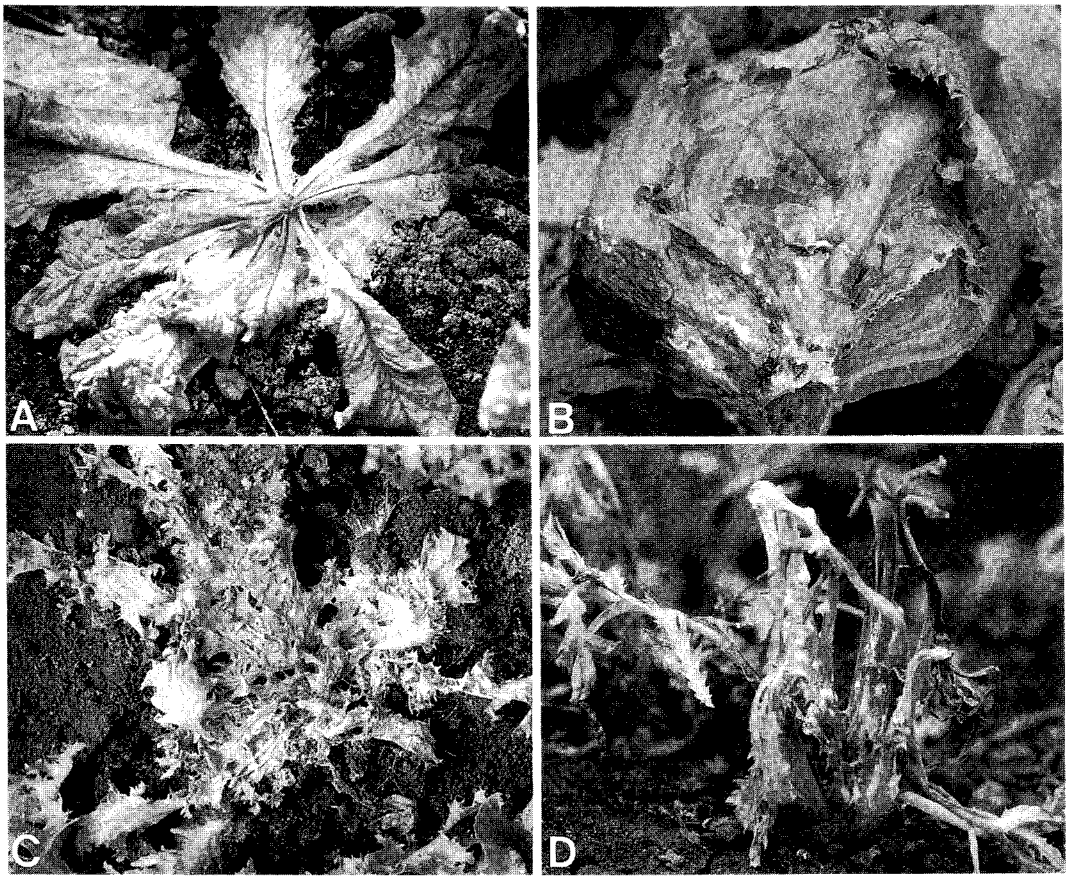

Fig. 1. Symptoms of Sclerotinia rot on four composites grown in the fields. A, lettuce; B, head lettuce; C, endive; D, Garland chrysanthemum.

and endive.

Morphological and cultural characteristics. Colonies of *S. sclerotiorum* on PDA consisted of white to gray mycelia 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. 2E). *S. sclerotiorum* produced one to several apothecia from a sclerotium (Fig. 2B), and *S. minor* mostly one (Fig. 2F). Asci of *S. sclerotiorum* and *S. minor* were very similar, which were cylindrical, 8-spored (Fig. 2C and G). Ascospores of the two species were also very similar, which were hyaline, ellipsoid to ovoid. There were two nuclei in the ascospores of *S. sclerotiorum* (Fig. 2D) and four nuclei in those of *S. minor* (Fig. 2H). The morphological and cultural characteristics of the two species differed in size of sclerotia and apothecia, number of sclerotia formed and mycelial growth rate on the medium (Table 3). The other mycological characteristics of the two species were very similar.

Pathogenicity. Eight isolates of *S. sclerotiorum* and four isolates of *S. minor* induced rot symptoms on the four composites inoculated (Table 4). The symptoms were similar to those observed in the fields. *Sclerotinia* spp. were re-isolated from the lesions on the plants inoculated. The pathogenicity tests revealed that there is no significant difference in virulence of the isolates to the

Table 2. Isolation and identification of *Sclerotinia* spp. from diseased plant parts of four composites

Host	No. of isolates obtained				No. of isolates identified	
	Leaf	Crown	Stem	Total	*S. sclerotiorum*	*S. minor*
Lettuce	128	37	0	165	111	54
Head lettuce	29	1	0	30	30	0
Endive	32	0	0	32	15	17
Garland chrysanthemum	3	0	10	10	10	0
Total	192	38	10	240	169	71

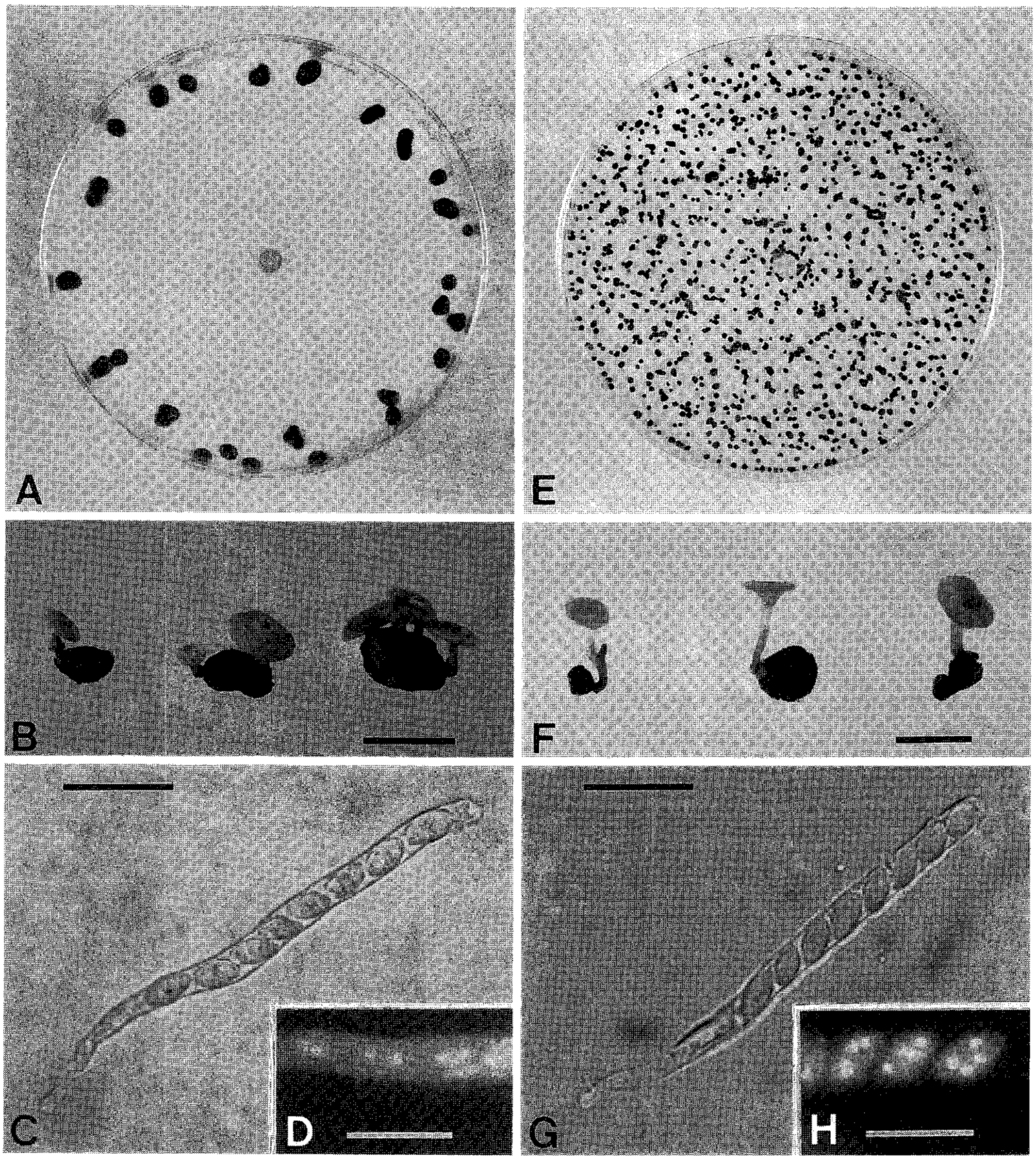

Fig. 2. Cultural and morphological features of *Sclerotinia sclerotiorum* (A-D) and *S. minor* (E-H) isolated from composites. A and E, 12-day-old colonies on PDA at 22°C in the dark; B (scale bar = 5 mm) and F (scale bar = 3 mm), apothecia produced from sclerotia; C and G, asci and ascospores (scale bar = 30 μm); D and H, nuclei in the ascospores stained with DAPI (scale bar = 15 μm).

composites and in susceptibility of the composites to the isolates.

Discussion

S. sclerotiorum and *S. minor* have wide host ranges although the latter has somewhat narrower host range than the former (Kim and Cho, 1998; Kohn, 1979a, 1979b; Purdy, 1979; Willetts and Wong, 1980). The present study reveals that *S. sclerotiorum* and *S. minor* cause Sclerotinia rot on four composite vegetable crops in Korea. *S. minor* was only isolated from lettuce and endive while *S. sclerotiorum* was isolated from all the composites. However, isolates of *S. minor* induced Sclerotinia rot symptoms on the plants of head lettuce and garland chrysanthemum as well as lettuce and endive by artificial inoculation. Accordingly, it is probable that *S. minor* causes the disease on head lettuce and garland chrysanthemum in the fields although it was not isolated from the two crops. Another species, *S. nivalis* which was found in carrot and some other plants in Japan (Saito, 1997) was reported to cause Sclerotinia rot on lettuce in China (Li *et al.*, 2000). The *Sclerotinia* species has not been found yet in Korea.

S. sclerotiorum and *S. minor* differ in size of sclerotia and apothecia, and number of nuclei in the ascospores. Size of sclerotia and apothecia of *S. sclerotiorum* is big-

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

Examination	Division	Description of characteristics	
		S. sclerotiorum	*S. minor*
Sclerotium	Formation	15-42/Petri dish	280-1000/Petri dish
	Color	Black	Black
	Shape	Globose to irregular	Globose to irregular
	Size	0.6~10.0×0.6~6.5 mm	0.5~3.8×0.5~2.2 mm
Microconidium	Shape	Globose	Globose
	Size	2~4 μm	2.6~4.2 μm
Apothecium	Formation	1-8/sclerotium	1-2/sclerotium
	Shape	Cup-shaped	Cup-shaped
	Diameter of disks	0.6~8.2 mm	0.6~2.7 mm
	Length of stalks	2.1~17.0 mm	1.5~6.8 mm
	Width of stalks	0.3~1.4 mm	0.2~0.7 mm
Ascus	Shape	Cylindrical	Cylindrical
	Size	120~200×8~11 μm	124~190×8~11 μm
Ascospore	Shape	Ellipsoid to ovoid	Ellipsoid to ovoid
	Size	10~18×5~8 μm	10~20×5~10 μm
	No. of nuclei	2	4
Mycelial growth	Minimum temperature	1°C	0°C
	Maximum temperature	30°C	28°C
	Optimum temperature	22~24°C	20~22°C
	Linear length/24 hr at 22°C	21.5~28.5 mm	11.6~20.0 mm

Table 4. Pathogenicity of isolates of *Sclerotinia sclerotiorum* and *S. minor* on four composites by artificial inoculation

Sclerotinia species	Isolate	Isolate source	Virulence of isolates on plants			
			Lettuce	Head lettuce	Endive	Garland chrysanthemum
S. sclerotiorum	S96-121	LT[a]	++[b]	++	++	++
	S97-125	LT	++	++	+	++
	S99-057	HL	++	++	+	++
	S99-073	HL	++	++	+	++
	S96-009	ED	++	++	++	++
	S97-064	ED	++	++	++	++
	S97-106	GC	++	++	++	++
	S97-111	GC	++	++	+	++
S. minor	S96-250	LT	++	++	+	++
	S96-244	LT	++	++	+	++
	S96-013	ED	++	++	++	++
	S96-138	ED	++	++	++	++
Control			–	–	–	–

[a]Abbreviation for hosts. LT : lettuce, HL : head lettuce, ED : endive, GC : garland chrysanthemum.
[b]Disease severity was rated ten days after inoculation. ++ : wholly rotted, + : partially rotted, – : no symptom.

ger than that of *S. minor*. There is no significant difference between the two species in shape and size of the asci and ascospores. The number of nuclei in the ascospores of *S. sclerotiorum* is two while that of *S. minor* is four. The morphological characteristics of the two species examined by the present authors are similar to those reported by other workers (Kohn, 1979a; Willetts and Wong, 1980).

Cultural characteristics of the two species somewhat differ in terms of number of sclerotia formed and mycelial growth on the medium. *S. minor* produces many more small sclerotia on the medium than *S. sclerotiorum*. The temperature range for mycelial growth of *S. sclerotiorum* is slightly higher than that of *S. minor*. The mycelial growth of *S. sclerotiorum* is faster than that of *S. minor* at the optimum temperature. Willetts and Wong (1980) reported that temperature range for mycelial growth of *S. sclerotiorum* and *S. minor* is 0 to 35°C with an optimum temperature of 20 to 25°C. The minimum and the optimum temperatures for mycelial growth of the two species are similar to those examined by the present authors. However, the maximum temperature for mycelial growth of the two species is somewhat higher than that examined by the present authors.

Sclerotia of *S. sclerotiorum* and *S. minor* in soil causes infection of host plants by forming mycelia or apothecia after a resting period (Purdy, 1979; Willetts and Wong, 1980). The present study reveals that the isolation frequency of *S. minor* from composite vegetable crops is lower than that of *S. sclerotiorum.* There is no information on the sclerotial density of *S. sclerotiorum* and *S. minor* in the fields of composite vegetable crops. The inoculum density of *S. minor* in fields with a history of losses due to *Sclerotinia* species was found to be 10 to 100 times greater than that of *S. sclerotiorum* (Adams and Ayers, 1979). It is supposed that sclerotia of *S. minor* are distributed in the fields many more than those of *S. sclerotiorum* because the species produces small and numerous sclerotia on the hosts like on the medium. However, *S. sclerotiorum* was more frequently isolated from the diseased composites than *S. minor*, suggesting that the sclerotial viability of *S. minor* is lower than that of *S. sclerotiorum* in the field. 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 been also 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 shows that there is no significant difference in virulence of the isolates to composite vegetable crops and in susceptibility of the crops to the isolates.

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