# Soil-Environmental Factors Involved in the Development of Root Rot/Vine on Cucurbits Caused by *Monosporascus cannonballus*

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A root rot/vine decline disease occurred naturally on bottle gourd-stocked watermelon, melon, oriental melon and squash grown in greenhouses, but not on these plants grown in fields. Self-rooted watermelon, cucumber, pumpkin and luffa were also proven to be hosts of the pathogen by artificial inoculation in this experiment. The pathogen was identified as Monosporascus cannonballus by comparing microscopic characteristics of fungal structures with those of previously identified fungal strains. Our field investigations showed that the temperature and electric conductivity of soil in infected greenhouses were higher and the soil moisture content was lower than in noninfected greenhouses. To investigate soil-environmental factors affecting disease development, greenhouse trials and inoculation experiments were conducted. The host plants inoculated and grown under conditions of high soil temperature and electrical conductivity  $(35 \pm 2^{\circ}C, 3.2-3.5 \text{ mS})$  and with low soil moisture content (pF 3.0-4.5) were most severely damaged by the fungal disease. Since plants growing in greenhouses are usually exposed to such environmental conditions, this may be the reason why the monosporascus root rot/vine decline disease has occurred only on cucurbits cultivated in greenhouses but not in field conditions.

**Keywords:** cucurbits, root rot/vine decline, soil electric conductivity, soil moisture content, soil temperature.

Watermelon is the most popular fruit in the summer time among the cucurbits in Korea, and now produced all the year round by greenhouse cultivation. Since the greenhouse culture of watermelon as well as other cucurbits was widespread, incidences of major diseases in the past such as *Fusarium* wilt, gummy stem rot and anthracnose, have been decreased. On the other hand, a group of new borne diseases have been prevalent, causing serious economic losses, one of which is a root rot/vine decline disease that

has recently occurred on cucurbits worldwide (Martyn and Miller, 1996; Mertely et al., 1991, 1992, 1993; Park et al., 1994; Stanghellini et al., 1996).

The disease was first reported by Pollack and Uecker (1974), caused by Monosporacus cannonballus Pollack and Uecker, ocurring especially in hot and dry areas, Texas and Arizona, of the USA, and in other countries such as Japan, Israel and Taiwan (Martyn and Miller, 1996; Reuveni et al., 1983; Uematsu et al., 1985). Park et al. (1994) first reported the occurrence of the disease on bottle gourd-stocked watermelon cultivated at a region nearby Chochiweon in Korea.

This disease is typically soil-borne and mainly occurs in desert or stress conditions, such as hot and dry regions in other studies (Martyn and Miller, 1996). High salt soil conditions may be an important factor for this disease development. Host range of this pathogen expands not only to cucurbits, but also to other crops such as wheat (Reuveni and Krikun, 1983). The disease has occurred mainly at the later developmental stage of the plant growth and thereby influenced on the fruit quality. Fruits could not be harvested if plants are severely damaged. There is no information about economic losses in Korea, but economic losses in USA alone estimate about 10-25% losses of the crops annually (Martyn and Miller, 1996).

Control strategies for this disease have not been established well because of no information about epidermiology of the disease, and only soil fumigation with methyl bromide has been studied (Martyn and Miller, 1996). The objectives of this study are to identify fungal isolates from infected hosts, and to investigate natural hosts and environmental factors of the disease. Possible environmental factors involved in disease development, such as temperature, water content, and electric conductivity were examined.

## Materials and Methods

**Isolation and culture of the pathogen.** Five fungal isolates were isolated from infected cucurbits and used in this experiment. Two fungal isolates, MW97-1 and MS98-1, were isolated from watermelon and squash, respectively, cultivated at Kwangsan area of

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Chonnam province. MM97-1 was isolated from melon cultivated at Kwangyang area of Chonnam province, and MW98-1 and MO98-1 were isolated from watermelon and oriental melon, respectively, cultivated at Jeongup area of Chonbuk province.

The fungi were isolated from the infected roots bearing perithecia or showing brown spots. The infected roots were washed with sterilized water and maintained at 26°C for 48 hr in a humid chamber to produce perithecia. The pathogens were confirmed by the observation of perithecia, asci and ascospores under a light microscope. The isolates were incubated on potato dextrose agar (PDA) at 28°C for more than 40 days to produce perithecia, and the perithecia were used as a inoculum for further study. The mycelial cultures were maintained at 4°C.

Pathogenicity tests. Seeds of watermelon (cv. Keumcheon), oriental melon (cv. Keumssaragi), muskmelon, cucumber (cv. Marketer), luffa and pumpkin were surface-sterilized by 0.2% sodium hypochlorite for 2 minutes, rinsed three times with sterile water, and pregerminated under sterile conditions on water-moistened Whatman 3-mm paper. The pregerminated seeds were planted in a plastic pot  $(10.5 \times 11.5 \times 6.5 \text{ cm})$  containing medium [sandy soil 3: peat moss (PRE-MIX) 1, v/v] sterilized by basamid (OCI Co.) and the plants were maintained in a growth chamber (20°C, 30,000 Lux, 16-h light/8- h dark, RH 50-60%). When the plants fully developed the fourth-leaves, they were inoculated with the fungal isolates. For preparation of the fungal inoculum, all fungal isolates were grown at 28°C on PDA for more than 40 days. Inoculum was prepared by suspension of the fungal isolates in sterile water and adjusted to  $4.7 \times 10^7$  spores/ml. About 5 ml of the prepared fungal suspension was poured into 5 cm in depth of soil around the main root area of a plant with a sterile 10-ml glass pippette. The inoculated plants were maintained in the growth chamber, and the development of symptoms such as brown spots on the root and wilting of leaves, was observed 40 days after inoculation. In vitro tests on fungal growth. Mycelial growth and maturity of ascospores were measured at different growth temperatures, 18°C, 22°C, 26°C, 30°C, 34°C, and 38°C. The fungal pathogens grown for 4 days on PDA were cut in the same size (1 cm × 1 cm), reinoculated on PDA plates, and incubated in growth chambers that were set at different temperatures. After 3 and 7 days, diameters of mycelial growth were measured. After 10 days of incubation, blackish perithecia were harvested with sterile water and blended to release ascospores. Maturity of ascospores was examined under the light microscope, and measured by their coloration. Each experiment was performed at least 3 times with 5 different plates. Greenhouse and field experiments. All field surveys were conducted in infected and noninfected greenhouses in the areas of the pathogen isolation mentioned above. Soil temperatures of 5-10 cm in depth in greenhouses were measured at 2-4 pm in late July through mid August. Soil samples (200 g each) were collected from greenhouses, suspended with sterile water to 1:5 (sample: water, v/v), measured electric conductivity (EC) by an EC meter. Soil water contents in greenhouses were observed with the naked eye and differentiated to dry, moderate, and humid statuses.

To examine environmental factors involved in the disease development, temperatures, EC, and water contents of soil in greenhouses were differently adjusted as follows. Greenhouses were shaded with polyethylene net to prevent temperature inside the greenhouses from increasing. Heating lines for the high temperature block and tap water lines using plastic pipe (10 mm in diameter) for the low temperature block were installed 10 cm deep in soil. One pregerminated seed was planted into a plastic pot and grew in the growth chamber until the 3rd or 4th leaf was fully developed. The plant was transplanted into different temperature blocks at intervals of 30 cm, and spaces were filled with soil. The pathogen inoculation was carried out by pouring 30 ml of spore suspension around the stem base of the plant. The high temperature block was maintained at  $37 \pm 2^{\circ}$ C and the low temperature area block maintained at  $23 \pm 2^{\circ}$ C constantly. Temperature of each block was checked at 9 a.m., 2 p.m., and 6 p.m., and the disease development was examined everyday.

Potted plants were supplied with 1/1,000 of Hysporex (II-Soo Chemical Co.) as nutrients until the 3rd or 4th leaf was fully developed. To adjust soil EC differently, EC 4.5 mS and 2.0 mS Hysporex solutions were supplied to the potted plant to make the high and low EC blocks, respectively. Soil EC values were maintained to 3.2-3.5 mS in the high EC block and 1.2-1.6 mS in the low EC block. Measurement of EC was performed using 3-cm-deep soil in each pot.

Different water contents of soils were adjusted by frequency of watering. After inoculation of the pathogen, the high water content block was maintained pF 0-2 and the low one was maintained pF 3.0-4.7. Temperature in the greenhouse during water content and EC experiments was kept at  $23 \pm 2^{\circ}$ C.

Five plants per each experiment was used, and the same experiment was replicated at least three times and the results were analyzed by T-test and Duncan's multiple range test.

## Results

Morphological characteristics of the pathogens. Size and shape of ascospores and perithecia of the fungal isolates from different hosts were compared with cucurbit isolates previously identified as M. cannonballus. There were statistically no differences (P < 0.05) in size and shape between the fungal isolates by Duncan's multiple range tests (Table 1). The fungal isolates were identified as M. cannonballus because the morphological characteristics of the fungal structures including perithecia, asci, and ascospores were very similar to those of the previously identified isolates (Table 2). Perithecia of M. cannonballus isolates were viewed as small black bulges or rarely round shape in root cortex of infected tissues. Color of perithecia was darkbrown and outer-surface of perithecia formed a dark-brown periapical ring. Sizes of perithecia were between 240- $420 \times 230-530 \,\mu\text{m}$  (average  $320 \times 454 \,\mu\text{m}$ ). Asci from naturally collected perithecia were not observed because of breakdown of the structure. Asci in perithecia of the fungal isolates grown for ten days on PDA plates were clearly observed under the light microscope, but ascus structure was disappeared when the fungal isolates were getting

Table 1. Mophological characteristics of Monosporascus isolates collected from various cucurbit plants

Fungal isolate		Size (µm)						
	Host -	Perithecium	Ascus	Ascospore				
MW98-1	Watermelon (Geum-chun)	280-400 × 380-500	55-100 × 35-50	32.5-45				
	,	$(320 \times 460) \text{ x}^{\text{b}}$	$(80 \times 45) \text{ x}$	(39) x				
MO98-1	Oriental melon (Geumsaragi)	260-390 × 360-530	$50-110 \times 35-50$	37.5-45				
11000	( (	$(340 \times 480) x$	$(94 \times 48) \text{ x}$	(38) x				
MM97-1	Melon	$260-380 \times 320-420$	$53-100 \times 35-50$	35-47.5				
		$(300 \times 460) \text{ x}$	$(79 \times 43) \text{ x}$	(40) x				
MW97-3	Watermelon (Geum-chun)	$280-420 \times 360-520$	$50-100 \times 36-52$	30-48				
	, , ,	$(320 \times 480) \text{ x}$	$(78 \times 45) \text{ x}$	(42) x				
MS98-1	Squash	$240-400 \times 230-480$	$55-110 \times 36-53$	35-47.5				
	<del>1</del>	$(320 \times 390) x$	$(83 \times 46) x$	(44) x				

<sup>&</sup>lt;sup>a</sup>Size of ascus formed on potato-dextrose arar 30 days after incubation

Table 2. Comparison of the descriptions for Monosporascus cannonballus between previously reported and reported in this study

Characteristics	Park et al. (1994)	Pollack & Uecker (1974)	This study (MW98-1)	
Perithecium size (µm)	220-570 (diameter)	almost 500 (diameter)	280-400 × 380-500 (diameter) (320 × 460)	
shape color	globose, periapical ring present dark brown	globose, periapical ring present light brown to dark brown	globose, periapical ring present dark brown	
Ascus size (μm)	50-110 × 35-50 (diameter)	56-90 × 30-55 (diameter)	55-100 × 35-50 (diameter) (80 × 45)	
shape	clavate to pyriform	clavate to pyriform	clavate to pyriform	
color	transparent	transparent	transparent	
septum	none	none	none	
Ascospore				
size (μm)	$30-45 \times 35-55$ (diameter)	$25-50 \times 30-55$ (diameter)	32.5-45 (diameter) (39 cm)	
shape	globose	globose	globose	
color	light brown to dark brown	light brown to dark brown	light brown to dark brown	

older. The ascus of the pathogen consisted of bi-layer with thick wall, clavate to pyriform in shape, and  $82.8 \times 45.5~\mu m$  in size (Table 2 and Fig. 1). Each ascus contained a single, dark, and spherical ascospore. When the ascospore matured, its color was changed from dark brown to black and became opaque. The surface of the ascospore was smooth and shiny after its release from the ascus, but no germ pore was observed (Fig. 1).

Optimal temperatures for mycelial growth and perithecial formation were 30-34°C, but optimal temperature required for ascospore maturing was 30°C. Ascospores were matured 92% 10 days after inoculation at 30°C, but only 35% and 17% of ascospores matured at 26°C and 34°C, respectively. At 18°C and 38°C, mycelial growth was slow and the perithecia were not observed even 60 days after

inoculation.

# Soil environmental factors in infected greenhouses.

Temperature and EC in infected greenhouses were higher than in noninfected greenhouses (Table 3). In addition, soil of the infected greenhouses was very dry. These observations may indicate that high temperature, high EC, and dry soil may be important environmental factors involved in the severe development of the disease.

To examine the effect of soil temperature on the development of the root rot/vine decline disease, different soil temperatures (high temperature,  $35 \pm 2^{\circ}$ C, and low temperature,  $25 \pm 2^{\circ}$ C) were set in the same greenhouse. In the high temperature conditions, all plants including watermelon, melon, gourd, oriental melon, and cucumber wilted within 45 days after inoculation of the pathogen and even-

<sup>&</sup>lt;sup>b</sup>Means followed by the same letter within a column are not different at P=0.05 by Duncan's multiple range test.

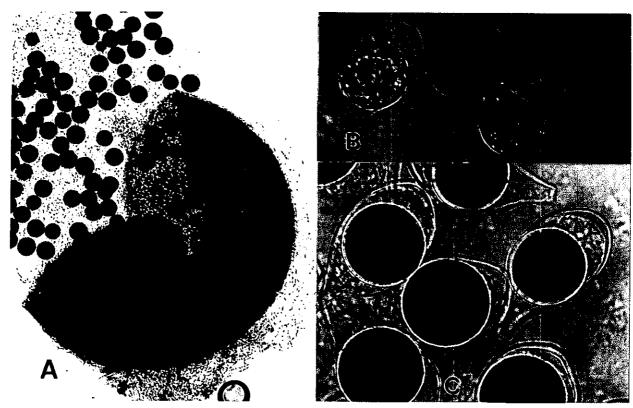


Fig. 1. Monosporascus cannonballus. (A) Ascospores released from a ruptured perithecium. (B) Young (pale) and matured (dark) ascospores. (C) Asci and ascospores.

**Table 3.** Field survey of soil temperature and electric conductivity in greenhouses in which monosporascus root rot/vine decline has occurred severely

Location		Soil ter	np. (°C)ª	Elec conductiv	% infec-	
		no infection	severe infection	no infection	severe infection	tion
Jeung-eup	l	$30 \pm 3$	39 ± 3	$2.0 \pm 2$	$2.8 \pm 2$	40
	2	$30 \pm 3$	$40 \pm 3$	$2.5 \pm 5$	$2.9 \pm 2$	70
	3	$27 \pm 3$	$40 \pm 3$	$1.8 \pm 2$	$2.8 \pm 1$	20
Kwang-san	1	$30 \pm 3$	$35 \pm 3$	$3.2 \pm 6$	$5.1 \pm 3$	80
	2	$30 \pm 3$	$35 \pm 3$	$3.7 \pm 3$	$3.5 \pm 5$	40
	3	$31 \pm 3$	$36 \pm 3$	$3.4 \pm 3$	$4.9 \pm 2$	50
	4	$30 \pm 3$	$38 \pm 3$	$2.5 \pm 2$	$2.7 \pm 2$	15
Average		29.72 <sup>x</sup>	37.58 <sup>y</sup>	2.73 <sup>x</sup>	3.54 <sup>y</sup>	_

<sup>&</sup>lt;sup>a</sup>Mean value of 5 replications (standard error) measured in depth of 5-10 cm underground at 2-4 p.m. during mid July throgh mid August. Different letters indicate significant differences among treatments at P=0.05, according to Duncan's multiple range test.

tually died within 55 days after inoculation. In the low temperature conditions, only watermelon wilted at 60 days

**Table 4.** Effect of incubation temperature on *in vitro* mycelial growth, perithecial production and ascospore maturity of *Monosporascus cannonballus* MW97-3 on potato-dextrose agar (PDA)

Temp.		al growt PDA at		Duration for perithecial	Maturity rate of ascospores 10 days	
(°C)	3 days	5 days 7 day		formation (days)	after perithecial pro- duction (%)	
18	_a	28	28	_		
22	8	45	85	30-33	31	
26	16	55	85	20-23	35	
30	45	85	85	17-20	92	
34	47	85	85	17-20	17	
38	14	60	60	_	_	

a-: no growth, no perithecial formation, or no ascospore maturing

after the pathogen inoculation, and growth of the plant decreased in some degrees, but the aboveground part of the plant was not dead.

Root infection, perithecial formation, population of M. cannonballus were also higher in the high temperature block than in the low temperature block (Table 5). Root infection index of all plants, except pumpkin and luffa, was 3 (infection rate 41-70%) in the high temperature block, but

<sup>&</sup>lt;sup>b</sup>Mean value of EC at the position where the soil temperatures was measured. Different letters indicate significant differences among treatments at P=0.05, according to Duncan's multiple range test.

Table 5. Effect of soil temperature (ST) on the development of monsporascus root rot/vine decline in cucurbit plants

	Root infe	ction rate <sup>a</sup>	Perithecial	formation <sup>b</sup>	Isolation frequency of fungi <sup>c</sup> (%)	
Plant tested	High ST (35 ± 2°C)	Low ST (23 ± °C2)	High ST (35 ± 2°C)	Low ST (23 ± 2°C)	High ST (35 ± 2°C)	Low ST (23 ± 2°C)
Watermelon (Geum-Chun)	3 a <sup>d</sup>	2 b	+++	++	70	50
Melon	3 a	2 b	+++	+	60	30
Gourd	3 a	1 b	++	+	30	14
Oriental melon (Geumsaragi)	3 a	2 b	+++	++	80	20
Cucumber (Marketer)	3 a	1 b	+++	_	76	24
Pumpkin	2 a	1 ab	_	_	15	5
Luffa	1 a	1 b		_	10	3

<sup>&</sup>lt;sup>a</sup>Root infection rate: 0, no infection; 1, 0-20%; 2, 21-40%; 3, 41-70%; 4, 71-100%.

that of the low temperature was 2 (infection rate 21-40%). Even though pumpkin and luffa were infected by *M. can-nonballus*, disease incidence and isolation frequency of the fungi were very low, and perithecia of the fungus were not formed.

Different temperatures and EC combinations were introduced in the greenhouse to investigate the effects of temperature and EC on the disease development. The root infection index in high temperature and high EC condition was 4 (infection rate, 71-100%) and plants died eventually, but the root infection index in low temperature and low EC was 1 (infection rate 1-20 %), not affecting on the plant growth (Table 6). The infection indices, perithecial formations, and isolation frequencies of the fungi were moderately high in the combinations of high temperature/low EC or low temperature/high EC, no plants died. These results indicate that high temperature and high EC conditions in the greenhouse may be important environmental factors for the severe disease development.

The severe root rot/vine decline disease was observed in

dry greenhouses during field surveys. To confirm the effect of soil water contents on the disease development, dry soil (pF 3-4.5) and humid soil conditions (pF 0-2) were artificially introduced. The root infection, perithecial formation, and isolation frequency of the fungi in dry soil were higher than in humid soil. Most plants in the dry conditions died 70 days after inoculation of the pathogen (Table 7).

### Discussion

In 1997 and 1998, five *Monosporascus* isolates were collected from gourd-stocked watermelon, melon, muskmelon, and squash which were grown in greenhouses at several areas of Chonnam and Chonbuk provinces. All the plant species showed wilt symptoms in their aboveground parts in the late developmental stage. All fungal isolates from the plants were identified as *M. cannonballus* in comparison with their morphological characteristics of previous descriptions reported by Pollack and Uecker (1974) (Table 1, 2). The conspicuous features of *M. cannonballus* are

Table 6. Effect of soil temperature (ST) and electric conductivity (EC) on the development of monosporascus root rot and vine decline in cucurbit plants

		Root infection rate <sup>a</sup>		Formation of perithecium <sup>b</sup>		Isolation frequency of the fungi <sup>e</sup> (%)	
Plant tested	EC	High ST (35 ± 2°C)	Low ST (23 ± 2°C)	High ST (35 ± 2°C)	Low ST (23 ± 2°C)	High ST (35 ± 2°C)	Low ST (23 ± 2°C)
Watermelon	high (3.2-3.5 mS)	4 x <sup>d</sup>	2 y	+++	. ++	80	64
	low (1.2-1.6 mS)	3 y	1 z	++	+	60	22
Gourd	high (3.2-3.5 mS)	4 x	2 z	+++	_	80	42
	low (1.2-1.6 mS)	3 y	1 w	++	_	62	10

<sup>&</sup>lt;sup>a</sup> Root infection rate: 0, no infection: 1, 0-20%; 2, 21-40%; 3, 41-70%; 4, 71-100%.

b, no perithecia formed; +, some perithecia; ++, perithecia in many places; +++, perithecia in all places.

emeasured by the number of root segments containing 15 brown spots from which the pathogenic fungus was isolated.

<sup>&</sup>lt;sup>d</sup>Means followed by the same letter within a column are not different at P=0.05 by by Duncan's multiple range test. Each plot consists of 5 plants.

b, no perithecia formed; +, some perithecia; ++, perithecia in many places; +++, perithecia in all places.

e measured by the number of root segments containing 15 brown spots from which the pathogenic fungus was isolated.

<sup>&</sup>lt;sup>d</sup> Means followed by the same letter within a column are not different at P=0.05 byDuncan's multiple range test. Each plot consists of 5 plants.

Table 7. Effect of soil water content (SWC) on the development of monsporascus root rot/vine decline in cucurbit plants

	Root infection rate <sup>a</sup>		Formation of	perithecium <sup>b</sup>	Isolation frequency of the fungi <sup>e</sup> (%)	
Plant tested	High SWC (pF0-2)	Low SWC (pF3-4.5)	High SWC (pF0-2)	Low SWC (pF3-4.5)	High SWC (pF0-2)	Low SWC (pF3-4.5)
Watermelon	2 y <sup>d</sup>	3 x	_	+	20	80
Gourd	1 y	2 x	_	+	15	46
Melon	2 xy	3 x	+	++	30	63
Cucumber	2 y	3 x	_	+	15	

<sup>&</sup>lt;sup>a</sup>Root infection rate: 0, no infection; 1, 0-20%; 2, 21-40%; 3, 41-70%; 4, 71-100%.

presence of one ascospore per asci and absence of anamorph (Kirkun, 1985; Pollack and Uecker, 1974). This disease has been reported in hot and dry areas worldwide, such as Texas and Arizona in the USA, Israel, India, Taiwan, and Japan (Martyn and Miller, 1996; Mertely et al., 1991, Mertely et al., 1993; Reuveni and Krikun, 1983; Stanghellini et al., 1996; Watanabe, 1979). Park et al. (1994) first isolated M. cannonballus from the rotted roots of bottle gourd stocks in damaged watermelon fields near Chochiwon in Korea. Economically significant losses by Monosporascus root rot/vine decline disease on melon have been reported in USA, Japan, and Israel (Martyn and Miller, 1996; Mertely et al., 1992; Morita and Furutani, 1996; Reuvenie et al., 1983; Stanghellini et al., 1996; Uematsu et al., 1985, 1992). M. cannonballus infects most of cucurbit plants (Uematsu and Sekiyama, 1990). In Korea, only bottle gourd stocks of watermelon have been reported as host of M. cannonballus (Park et al., 1994). According to our investigations, roots of watermelon, melon, oriental melon, and squash were newly added as natural hosts for the pathogen in the field. In addition, roots of self-stocked watermelon, cucumber, pumpkin, and luffa were also infected by the inoculation tests in the greenhouse (Table 5).

Optimal temperature for mycelial growth and perithecial formation of *M. cannonballus* isolates was 30-34°C, but the optimal temperature required for ascospore maturing was 30°C (Table 4). Previously reported optimal temperatures for growth of regional isolates of the pathogen were varied as 30-35°C (Martyn and Miller, 1996), 30°C (Park et al., 1994), 28-32°C (Uematsu and Akayama, 1990), and 45°C (Reuveni an Krikun, 1983). These result may indicate the different adaptation abilities of the *M. cannonballus* isolates in different inhabitants. In general, optimal temperature for the growth of *M. cannonballus* is above 30°C, which may explain the sevece occurrence of this disease in hot, dry, and high EC soil areas such as Texas, Arizona, Israel, and Libya. In Korea and Japan, the Monosporascus root rot/

vine decline disease mainly occurrs in greenhouses during the hot summer season, but not in the open field. These cultivating conditions may raise soil temperatures to the favorable conditions for growth and infection of *M. cannonballus*, thus creating an artificial niche for the fungus in an otherwise temperate zone.

The root rot/vine decline is a typical soil-borne disease, thus soil environmental factors may be important for the disease development. In our field surveys, high temperature, high EC, dry conditions of soil were common in severely infected greenhouses. Our artificial environmental studies in the greenhouse supported the field observations and confirmed that those environments are the most important factors related to the disease incidence in greenhouses. The infection rate, perithecial formation, isolation frequency of the fungi in high temperature  $(35 \pm 2^{\circ}C)$  was higher than those in low temperature  $(23 \pm 2^{\circ}C)$ . In addition, the three factors were much higher in the combination of high temperature and high EC than other combinations such as low temperature/high EC or low EC, and high temperature/low EC. However, it is not clear which of temperature, EC, and soil dryness is the most important factor in severe disease occurrence.

The severity of the disease on cucurbits was increased in alkaline soil that accumulated high concentrations of salts. *M. cannonballus* grew optimally pH 6 to 7 on plates, but also grew at pH 9.0 (Martyn and Miller, 1996; Park et al., 1994). The alkalization of soil may be mainly due to accumulation of salts in soil. Therefore, the occurrence of severe disease in high EC soil may be mainly due to change of soil pH to alkaline in which the fungal pathogen can grow very well. Martyn and Miller (1996) suggested that alkaline and high salts in soil may be additional important environmental factors in the disease development.

One of the important soil factors in the disease development is dryness of soil. The infection rate, perithecial formation, and ascospore formation were higher in dry soil (pF 3-4.5) than humid soil (pF 0-2). It is well known that dry-

b, no perithecia formed; +, some perithecia; ++, perithecia in many place; +++, perithecia in all place.

emeasured by the number of root segments containing 15 brown spots from which the pathogenic fungus was isolated.

<sup>&</sup>lt;sup>d</sup>Means followed by the same letter within a column are not different at P=0.05 by Duncan's multiple range test. Each plot consists of 5 plants.

ness of soil increase accumulation of salts in surface soil. Our results confirmed the Monosporascus root rot/vine decline disease on cucurbits occurs severely in high temperature, high EC, dry, and alkaline soil conditions. To control this disease in the greenhouse, those environmental conditions should be avoided especially during hot summer days.

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