Plant Pathol. J. 20(3) : 165-171 (2004)

Potential Biotypes in Korean Isolates of *Bipolaris cactivora* Associated with Stem Rot of Cactus

Jeong Ho Kim, Myoung-Il Jeoung[1], Ick-Hwa Hyun[2] and Young Ho Kim*

School of Agricultural Biotechnology and Center for Plant Molecular Genetics & Breeding Research, Seoul National University, Seoul 151-742, Korea

[1]National Horticultural Research Institute, Rural Development Administration, Suwon 440-310, Korea

[2]National Plant Quarantine Office, Anyang, Korea

(Received on June 20, 2004; Accepted on August 11, 2004)

A total of 62 isolates of *Bipolaris cactivora* causing cactus stem rots were isolated from major cactus-growing areas in Korea. Colony morphology of the isolates on potato-dextrose agar was differentiated into aerial (CA) and non-aerial mycelial types (CB). CA had profound aerial mycelium with grayish brown (CA-1), light brownish (CA-2), and brownish (CA-3) pigmentations; respectively, while CB had dark brownish pigmentations. CA had conidia of less dark pigmentation and acute terminal end. CB had darker and more round-end conidia. Twenty-eight amplified fragments were produced by polymerase chain reaction (PCR) with a set of 2 random primers. The sizes of amplified DNA fragments ranged approximately from 0.1 to 2.3 kb. The isolates were classified into 2 major genomic DNA random amplified polymorphic DNA (RAPD) groups at the genomic similarity of 97.7% and 95.1%, respectively. Cluster analysis of genetic similarity among the isolates generated a dendrogram that clearly separated all isolates into SA or SB. This result suggests that there may be two morphotypes of *B. cactivora* in Korea that may differ in their genetic constitutes.

Keywords **:** *Bipolaris cactivora*, cactus stem diseases, colony morphology, polymerase chain reaction (PCR), random amplified polymorphic DNA (RAPD), conidial type.

A grafted cactus, comprising two different cactus species as a stock and as a scion (like a flower bud), is a major commercial product. It is one of the most important ornamental plants for export in Korea due to its high quality in the world market. The most widely cultivated stock is a three-angled (*Hylocereus trigonus*) cactus, with a scion that is a plain (*Gymnocalycium mihanovichii*) cactus.

One of the limiting factors for cultivating grafted cacti in greenhouses is the presence of cactus diseases, especially the stem rot caused by fungi. Various fungal diseases causing stem rot are known worldwide. However, only four fungal diseases have been known to occur in grafted cactus in Korea.

The major causal fungi are *Fusarium oxysporum* (Hyun et al., 1996), *Bipolaris cactivora* (old names: *Helminsosporium cactivorum* and *Drechslera cactivora*) (Chang et al., 1998), *Rhizoctonia solani* (Lee et al., 1989) and *Glomerella cingulata* (anamorph; *Colletotrichum gloeosporioides*) (Kim et al., 2000b). *G. cingulata* (*C. gloeosporioides*) occurs naturally in Jeju island (Kim et al., 2000a).

The stem rot caused by *B. cactivora* is one of the most destructive diseases in cactus. The pathogen is widely distributed in cactus-growing areas. In the past, it caused a severe epidemic in some greenhouses in Korea (Chang et al., 1998). This disease mostly occurs in young cylindrical cactus plants (three-angled cactus), especially basal and middle parts of the stem. Rots also occur at the upper end of stock cactus and spread to scion cactus, or sometimes at scion cactus alone (Chang et al., 1998).

Many of the cactus plants (Cactaceae) are not native to Korea, but have been introduced from various tropical and subtropical regions such as America. This suggests that pathogens of cactus may have different origins and diverse genetic backgrounds.

In this study, *B. cactivora* isolates in Korea were examined for biological and genetic diversity to give basic information in managing the stem rot of cactus caused by the pathogen.

Materials and Methods

Fungal isolates. In 1999 and 2000, occurrence of cactus stem rot was surveyed in cactus greenhouses of Suwon and its vicinity, Anseong, Eumseong, Cheonan, Daegu, and Goyang.

A variety of stem rot symptoms have been frequently found in cactus greenhouses. For fungal isolation, cactus stem tissues with rot symptoms were excised and surface-sterilized with 75% eth-

*Corresponding author.
Phone) +82-2-880-4675, FAX) +82-2-873-2317
E-mail) yhokim@snu.ac.kr

anol for 30 sec, followed by a 60 sec rinse with 1% sodium hypochlorite, and placed on water agar (WA). Fungal mycelia grown out of the tissues were transferred to fresh potato-dextrose agar (PDA), cultured at 25°C, unless otherwise noticed. Identification of isolates for *B. cactivora* was based on mycological characteristics proposed by Alcorn (1983, 1988).

Cultural characters of *B. cactivora*. Agar discs (5 mm in diameter) from the growing edge of cultures on PDA were placed in the center of fresh PDA, and grown for 7 days. Colony morphology was visually examined. Classification of colony morphology was based on the formation of aerial mycelium and colony pigmentation. Conidia were collected from each isolate and examined under a light microscope (Axiophot, Zeiss, Germany). Conidial morphology and pigmentation were examined visually under the microscope.

Virulence and host range of *B. cactivora* isolates. Twelve isolates of *B. cactivora* with different colony morphology were cultured on PDA at 25°C in an incubator for 7days. The fungal conidia were collected in sterilized water (adjusted to 10^6 conidia/ml). A drop (6-7 μl) of the conidial suspension was placed on the center of stem discs of various cacti such as *Eriocereus jusbertii, Cereus peruvianus, C. teragonus, Myrtillocactus geomertrizans, Eriocereus tortuosus, Gymocalycium mihanovichii, Echinocactus grusonii, Mammillaria elongata, Lobivia nealeana, Eriocactus leninghusii,* and *Notocactus scopa.*

After inoculation, they were placed in glass Petri dishes containing moistened paper towel at 25°C and 100% RH. Rot symptom development was examined 7 days after inoculation.

DNA amplification and genetic analysis

DNA extraction. Fungi were grown in potato-dextrose broth (PDB) in still culture for 4 days at 25°C in the incubator. Mycelium was harvested, filtered through cheesecloth, washed with sterile distilled water, and transferred to filter paper for removal of excess water. After being frozen at -20°C, mycelia were dried in a freeze drier. Freeze-dried mycelia (1 g) were ground to fine powder in liquid nitrogen. The mycelial powder was transferred to a 1.5-ml Efendorf tube. Lysis buffer (50 mM Tris-HCl, 50 mM EDTA, 3% SDS, and 1% β-mercaptoethanol) was added. DNA was extracted twice with phenol:chloroform (1:1) with RNase treatment to remove RNA.

Finally, total DNA was precipitated in ethanol. The DNA was dissolved in TE buffer (10 mM Tris-HCl, pH 7.5; 0.1 mM EDTA) and quantified by TKO 100 Fluorometer (Hoefer, LA, USA).

Polymerase chain reaction (PCR) for DNA amplification. Amplification reactions were performed in 50 volumes containing 10 mM Tris-HCl (pH 8.3), 50 mM KCl, 1.5 mM $MgCl_2$, 125 μM each of dATP, dCTP, dGTP, and dTTP (Perkin-Elmer Cetus, Norwalk, CT), 100 pmole of 10-mer random primer (Operon Technologies, Alameda, CA, USA), 20 ng of genomic DNA, and 2 U of *Taq* polymerase (Perkin-Elmer Cetus, Norwalk, CT, USA).

Two primers (5'-GGGTAACGCC-3', 5'-AGATGCAGCC-3') were selected for DNA amplification. Amplifications were performed in Perkin-Elmer GenAmo PCR System 9600, and programmed for 40 cycles of 1 min 30 sec at 94°C, 2 min at 40°C, and 2 min at 72°C by using the fastest available transitions between each temperature.

Random amplified polymorphic DNA (RAPD) and data analysis. PCR products were electrophoresed in 1.5% (W/V) agarose gel and detected by staining with ethidium bromide. Lambda DNA digested with *HindIII* and 100 bp ladder-size markers were used as molecular weight markers. RAPD patterns were compared for the isolates of *B. cactivora*. The genetic

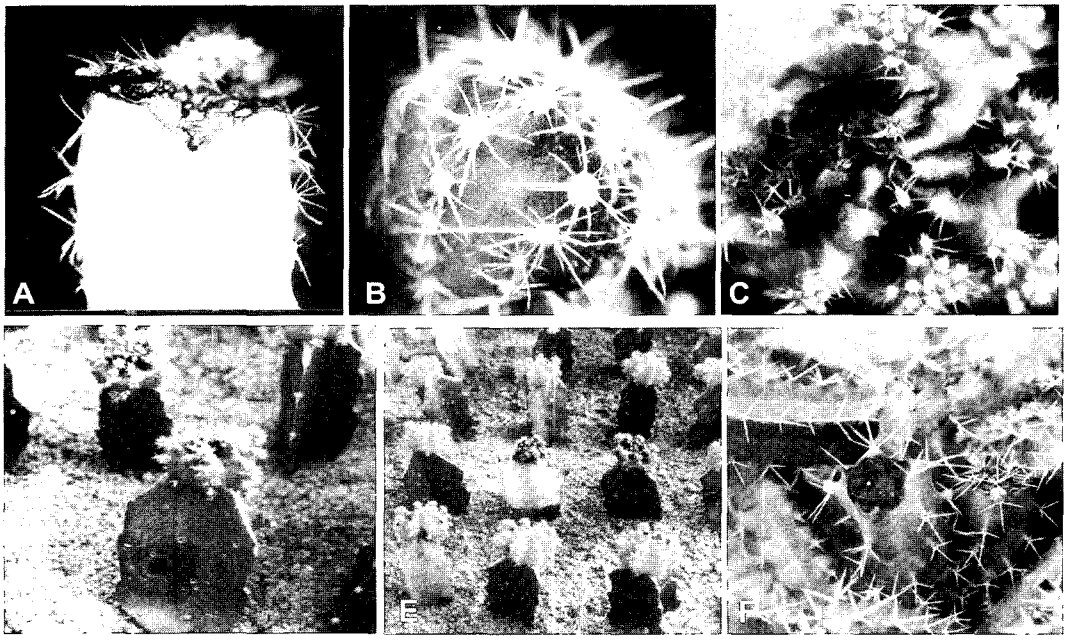

Fig. 1. Symptoms of stem rots on various cactus species caused by *Bipolaris cactivora*. **A**: a rot on the upper stem of *Cereus tetragonus*. **B, C**: small and large stem rots formed on *Gymocalycium mihanovichii*. **D, E**: symptoms on graft-cactus, showing various rots on stock (*Hylocereus trigonus*) and scion (*G. mihanovichii*) cacti. **F**: rot lesion on *Echinocereus pentalophus*.

Table 1. *Bipolaris cactivora* isolates with their colony types, and conidial types used in this study

Isolate No.	Isolate name	Colony type[b]	Conidial type[c]	Area isolated	Host plant
1	GG-1	CA-1	SA	Goyang	*Gymocalycium mihanovichii*
2	CH-1	CA-1	SA	Cheonan	*Hylocereus trigonus*
3	GG-2	CA-1	SA	Goyang	*G. mihanovichii*
4	GG-3	CA-1	SA	Goyang	*G. mihanovichii*
5	CG-1	CA-1	SA	Cheonan	*G. mihanovichii*
6	GG-4	CA-1	SA	Goyang	*G. mihanovichii*
7	CG-2	CA-1	SA	Cheonan	*G. mihanovichii*
8	YC-1	CA-1	SA	Yeoju	*Cereus peruvianus*
9	CH-2	CA-1	SA	Cheonan	*Hylocerus trigonus*
10	GG-5	CA-1	SA	Goyang	*G. mihanovichii*
11	GS-1	CA-1	SA	Goyang	*Chamaecereus silvestrii*
12	CH-3	CA-1	SA	Cheonan	*H. trigonus*
13	CS-1 (712-11)[a]	CA-1	SA	Cheonan	*Ch. silvestrii*
14	YU-1 (803-46)	CA-1	SA	Yeoju	unknown
15	UG-1 (1159)	CA-1	SA	unknown	*G. mihanovichii*
16	KG-1 (803-27)	CA-1	SA	Kimcheon	*G. mihanovichii*
17	CG-3 (803-38)	CA-1	SA	Cheonan	*G. mihanovichii*
18	UG-2 (1158)	CA-1	SA	unknown	*G. mihanovichii*
19	YC-2	CA-2	SA	Yeoju	*C. peruvianus*
20	YC-3	CA-2	SA	Yeoju	*C. peruvianus*
21	YE-1	CA-2	SA	Yeoju	*Echinocereus pentalophus*
22	YM-1	CA-2	SA	Yeoju	*Mammillaria elongata*
23	YC-4	CA-3	SA	Yeoju	*C. peruvianus*
24	CH-4	CA-3	SA	Cheonan	*H. trigonus*
25	CH-5	CA-3	SA	Cheonan	*H. trigonus*
26	CG-4	CA-3	SA	Cheonan	*G. mihanovichii*
27	GU-1	CB	SB	Goyang	unknown
28	GL-1	CB	SB	Goyang	*Lobivia nealeana*
29	GL-2	CB	SB	Goyang	*L. nealeana*
30	GE-1	CB	SB	Goyang	*Echinocereus pentalophus*
31	GE-2	CB	SB	Goyang	*Echinocereus dasyacanthus*
32	GC-1	CB	SB	Goyang	*C. peruvianus* .
33	GG-6	CB	SB	Goyang	*G. mihanovichii*
34	GC-2	CB	SB	Goyang	*C. peruvianus*
35	YC-5	CB	SB	Yeoju	*C. peruvianus*
36	GC-3	CB	SB	Goyang	*C. peruvianus.*
37	GG-7	CB	SB	Goyang	*G. mihanovichii*
38	CT-1	CB	SB	Cheonan	*Trichocereus macroonus*
39	GC-4	CB	SB	Goyang	*C. peruvianus*
40	GC-5	CB	SB	Goyang	*C. peruvianus*
41	CC-1 (612-4)	CB	SB	Cheonan	*C. peruvianus*
42	CU-1 (517-12)	CB	SB	Cheonan	unknown
43	CS-2 (1157)	CB	SB	Cheonan	*Ch. silvestrii*
44	CC-2 (1155)	CB	SB	Cheonan	*Cereus tetragonus*
45	KI-2 (803-31)	CB	SB	Kimcheon	*Isolatocereus dumortier*
46	GS-2 (1150)	CB	SB	Goyang	*Ch. silvestrii*
47	CS-3 (612-1)	CB	SB	Cheonan	*Ch. silvestrii*
48	CS-4 (517-17)	CB	SB	Cheonan	*Ch. silvestrii*
49	CC-3 (517-9)	CB	SB	Cheonan	*C. peruvianus*
50	CC-4 (517-6)	CB	SB	Cheonan	*Cereus neopitahaja*
51	AG-5 (614)	CB	SB	Anyang	*G. mihanovichii*
52	CU-2 (803-22)	CB	SB	Cheonan	unknown
53	CC-5 (1156)	CB	SB	Cheonan	*C. tetragonus*
54	CS-5 (803-20)	CB	SB	Cheonan	*Ch. silvestrii*
55	GS-3 (1152)	CB	SB	Goyang	*Ch. silvestrii*
56	GC-6 (1149)	CB	SB	Goyang	*C. peruvianus*
57	GC-7 (712-7)	CB	SB	Goyang	*C. peruvianus*
58	CC-6 (1151)	CB	SB	Cheonan	*C. peruvianus*
59	GG-8 (712-8)	CB	SB	Goyang	*G. mihanovichii*
60	CS-6 (612-2)	CB	SB	Cheonan	*Ch. silvestrii*
61	GG-9 (712-10)	CB	SB	Goyang	*G. mihanovichii*
62	CS-7 (1153)	CB	SB	Cheonan	*Ch. silvestrii*

[a] Figures in parentheses are original isolate numbers.
[b] colony type: aerial mycelium with grayish brown (CA-1), light brownish (CA-2), and brownish (CA-3) pigmentations, and poor aerial mycelium with dark brownish pigmentation (CB)
[c] Conidial type: SA: brown with more acute terminal end; SB: dark brown with more round terminal end.

similarity among isolates was estimated by using the simple matching coefficient based on the presence (coded 1) or absence (coded 0) of data of each band. A dendrogram was constructed based on the binomial data matrix of RAPD analysis. The non-weighed pair group method with an arithmetic average (UPGMA) option in the Numerical Taxonomy System for Personal Computer (NTSYS-pc), version 1.80 (Rohlf, 1983) was used.

Results

Stem rot symptoms and mycological characters of *B. cactivora*. Symptoms caused by *B. cactivora* were variable (Fig. 1). Symptoms on stock cactus were pale yellow, with water-soaked lesions at the initial stage, followed by a change in the color from pale yellow into light brown and rotting later (Fig. 1A-C, F). Sometimes the whole stem was destroyed (Fig. 1. E).

On scion cactus, small spots with an irregular margin initially appeared and enlarged with time. Cactus scions turned light brown and dry leading to death (Fig. 1D, E). Sometimes these became rotten with terminal end turning black (Fig. 1E).

In this study, 62 isolates of *B. cactivora* were isolated from these stem rots (Table 1). Colony morphology of the isolates could be differentiated into aerial mycelial (CA) and non-aerial mycelial types (CB) (Fig. 2). The aerial mycelial type contained three colony types: CA-1, CA-2, and CA-3 -- with colony pigmentations of grayish brown, dark brown, and light brown; respectively. The non-aerial mycelial type (CB) had mostly dark brown colonies (Fig. 2).

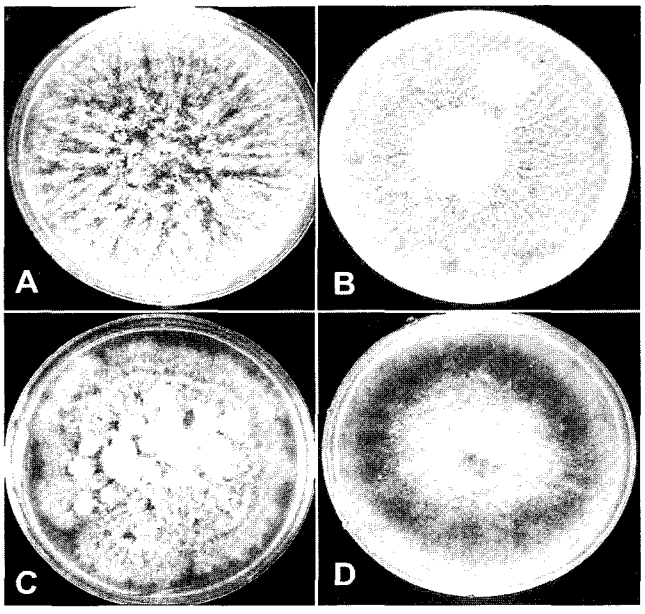

Fig. 2. Colony morphology of *Bipolaris cactivora*. A: Aerial mycelia with grayish brownish pigmentation (CA-1). B: Aerial mycelia with light brownish pigmentation (CA-2). C: Aerial mycelia with brownish pigmentation (CA-3). D: Non-aerial mycelial type with dark brownish pigmentation (CB).

In continuous sub-culturing, most isolates retained the same colony morphology; however, some variations in colony color were noted in CA-2 and CA-3. The most abundant colony type was CB comprising 36 isolates, followed by CA-1 which had 18 isolates (Table 1). Four isolates were isolated for each of CA-2 and CA-3. The colony types were neither related to areas nor to cactus plants from which the isolates were obtained (Table 1).

Mycological characteristics of *B. cactivora* isolates in the present study matched well with that of the previous description (Durbin et al., 1955). Conidiophores were erect, elongated, brown, rarely branched, irregularly nodulose, and light colored at the tip. Conidia are borne singly, hyaline at first, later becoming thick-walled and dark olive to chocolate brown, 1- to 5- septate (usually 3). For the most part, these are cylindrical and straight with rounded ends, but occasionally bifurcate types. Their size ranged from and 25-50 × 7-11 um (data not shown).

Conidial morphology varied somewhat among the isolates, which could be differentiated altogether into two types. Conidial type A (SA) was less dark and somewhat conical at the terminal end, while conidial type B (SB) had more dark conidia and/or more round at the terminal end (Fig. 3).

Generally SA included isolates of aerial colony types CA-1, CA-2, and CA-3; but SB comprised SB isolates (Table 1). However, there was no relationship between conidial types and isolated areas and hosts (Table 1).

Virulence of *B. cactivora* isolates. Symptoms developed 4 days after inoculation with brown to black spots at inoculated areas. Brown and black rot symptoms appeared around 7 days after inoculation in all cactus species inoculated (except *E. grusonii, E. grusonii,* and *M. elongata*

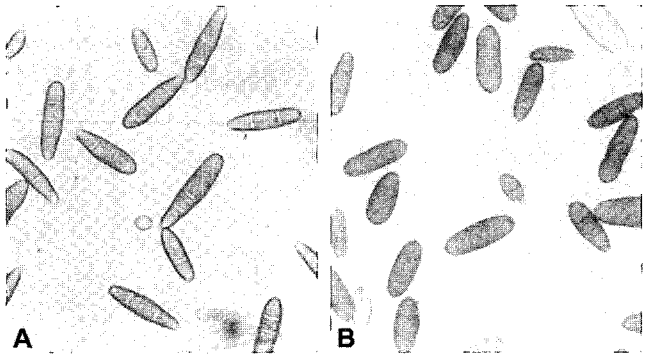

Fig. 3. Typical morphological characters of *Bipolaris cactivora* conidia, showing lighter pigmentation and more acute terminal end for A than for B. Conidial type B involves non-aerial mycelial type of colony (CB).

Table 2. Pathogenicity of *Bipolaris cactivora* isolates in various cactus species

Cactus species[a]	Virulence of isolates											
	CA-1[c]			CA-2			CA-3			CB		
	GG-1	CH-1	CG-1	YC-2	YC-3	YE-1	YC-4	CH-4	CH-5	GC-3	YC-5	CS-4
EJ	+++	+++	+	++	++	++	+++	+++	+++	++	++	++
CP	+++	++	++	++	++	++	++	+++	+++	+++	++	++
MG	++	++	++	+++	++	+++	+++	+++	++	++	++	++
CT	+++	+++	+++	+++	+++	+++	+++	+++	+++	+++	+++	+++
ET	+++	++	+++	++	++	++	++	+++	+++	++	++	++
GM	+++	+++	+++	+	+	+++	+++	+++	+++	++	+++	+++
EcG	++	+++	+++	++	−	+++	+++	+	+++	−	+++	+++
MmE	+++	+	−	nt	nt	nt	nt	nt	nt	++	+	+
LN	+++	+	+++	+++	++	+	+	+++	+	+++	+++	+
EcL	+++	+++	+++	+++	+++	+++	+++	+++	+++	+++	+++	+++
NS	++	+++	+++	+++	+++	+++	+++	++	+++	+	++	++

[a] EJ: *Eriocereus jusbertii*, CP: *Cereus peruvianus*, MG: *Myrtillocactus geometrizans*, CT: *Cereus tetragonus*, ET: *Eriocereus tortuosus*, GM: *Gymocalycium mihanovichii*, EcG: *Echinocactus grusonii*, MmE: *Mammillaria elongate*, LN: *Lovia nealeana*, ErL: *Eriocactus leninghausii*, and NS: *Notocactus scopa*

[b] +: weak rot, ++: moderate rot, +++: severe rot, −: no symptom, nt: not tested

[c] colony type: aerial mycelium with grayish brown (CA-1), light brownish (CA-2), and brownish (CA-3) pigmentations, and poor aerial mycelium with dark brownish pigmentation (CB)

by *B. cactivora* isolates YC-3, GC-3, and CG-1, respectively) (Table 2).

There was no significant difference in virulence between the two conidial types and among the 12 isolates used in this study. However, less severe rot symptoms were developed in some combinations of cacti and *B. cactivora* isolates.

DNA amplification and RAPD analysis. Two primers, which gave clear reproducible bands with genomic DNAs of the isolates of *B. cactivora*, were analyzed for RAPD patterns. Concentrations of DNA templates, primers, and dNTP were determined in preliminary trials to get unambiguous amplification patterns. The profiles were reproducible. The two primers used revealed polymorphisms useful for differentiating isolates of *B. cactivora*.

Twenty-eight amplified fragments were produced by polymerase chain reaction (PCR), and the sizes of amplified DNA fragments ranged approximately from 0.1 kb to 2.3 kb (Figs. 4, 5). The two primers tested distinguished CB isolates of *B. cactivora* from those of CA-1, CA-2, and CA-3 due to the absence of 4 amplification products except the RAPD patterns of isolate BMD4 that were similar to those of CA isolates.

Cluster analysis. By combining the RAPD results with the

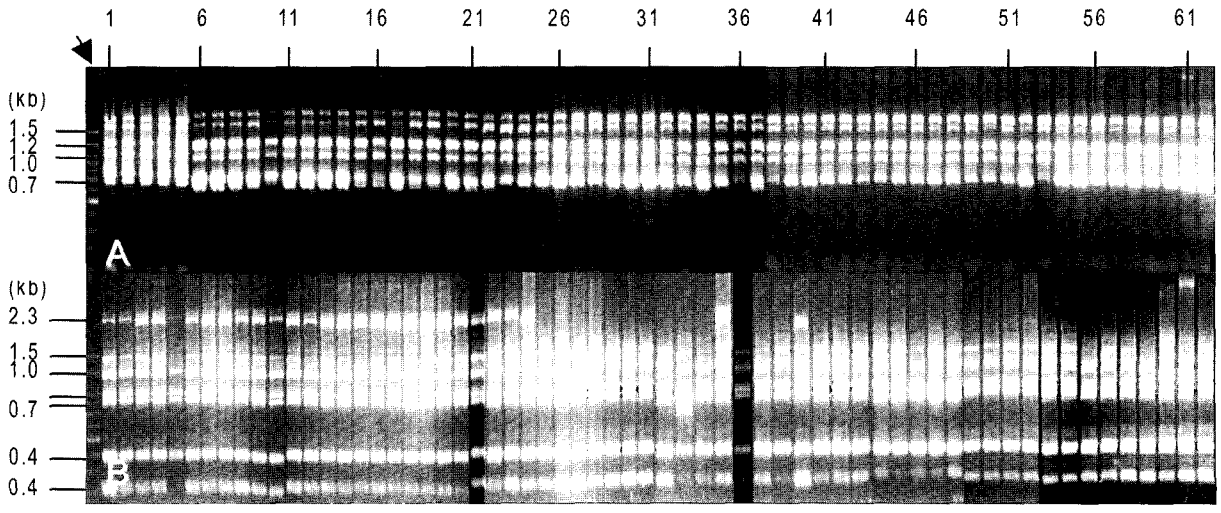

Fig. 4. Gel electrophoresis of polymerase chain reaction products with two primers, A09 primer (**A**) and F03 primer (**B**), of DNA from isolates of *Bipolaris cactivora*. Numbers are isolate numbers designated in Table 1. Arrow: molecular weight markers.

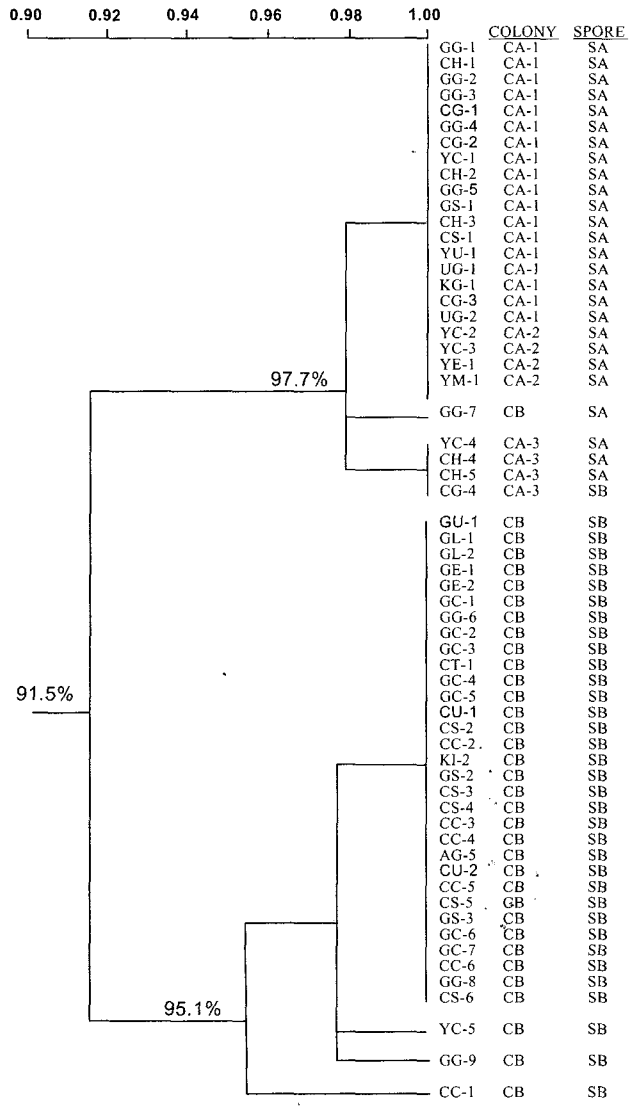

Fig. 5. Cluster analysis of 62 *Bipolaris cactivora* isolates isolated from stem rots of cactus plants based on RAPD using two primer sets (5'-GGGTAACGCC-3'; 5'-AGATGCAGCC-3').

two primer sets, 28 band positions were scored for presence versus absence (1/0) for all the isolates tested. The combined data from 28 RAPD bands were analyzed by clustering using UPGMA in NTSYS-pc to produce a dendrogram (Fig. 5). At the similarity level of 91.4%, genomic DNA RAPD groups of the 62 isolates of *B. cactivora* were classified into two major groups at the genomic similarity of 97.7% and 95.1%, respectively. These two RAPD groups matched well with the two conidial types except isolate GG-7 (Fig. 5).

Discussion

Genus *Bipolaris* is an anamorph of *Cochliobolus*. It belongs to Dematiaceae in Hyphomycetes, Fungi Imperfecti (Hawksworth et al., 1995). Formerly, it had the name of *Helminthosporium* that was originally established by Link in 1809, and has been changed to various genera.

At present, original genus *Helminthosporium* is separated into 4 genera such as *Helminthosporium*, *Bipolaris*, *Exserohilum*, and *Drechslera* (Sivanesan, 1987). Their teleomorphs also support the separation into *Bipolaris*, *Drechslera*, and *Exserohilum, which are Cochliobolus*, *Pyrenophora*, and *Setophaeria*; respectively. *Cochliobolus* is a common teleomorph of both *Bipolaris* and *Curvularia* anamorphs.

Symptoms. *Bipolaris cactivora* in general causes top or basal rot on various cactus species (Chang et al., 1998; Chase, 1982; Drubin et al., 1955). Initial symptoms are yellow lesions that are well defined. These yellow lesions turn to dark brown at later stages. Entire plants may rot 2-4 days after infection. The fungus penetrates into the plant stems through the stomata or wounds. Sometimes it directly penetrates, especially in young stems. The mycelium grows intercellularly and produces conidiophores that break through the epidermis or stomata to form abundant conidia under moist conditions (Durbin et al., 1955).

In this study, *B. cactivora* isolates were obtained from various rot symptoms of cactus plants. They were all identified as *B. cactivora*, but had some variations in mycological characters. In particular, colony morphology of the isolates on PDA was differentiated to 4 types. One of these was very distinct from other colony types which showed poor aerial mycelium with dark brown colony color (colony type B).

Also, isolates of this colony type mostly had dark brown conidia and more round terminal end (conidial types B, SB). The other conidial type had rather conical terminal end of conidia (conidial type A, SA). By image analysis, the roundness of conidial terminal end was measured, in which SB (average roundness index of 0.67) had generally more round terminal end than that of the SA (average roundness index of 0.57) (unpublished data). This confirmed the morphological difference quantitatively. In this study, therefore, the colony type of the fungus may be closely related to the conidial type.

RAPD patterns of genomic DNAs using two sets of random primers were compared to genetically differentiate isolates of *B. cactivora*. The isolates were clustered into two different subgroups depending on colony and conidial types except one isolate. This suggests that the isolates with same type of colony and conidial morphology have similar genetic constituents. RAPD has been widely used to detect genetic polymorphisms in fungi (Assigbetse et al., 1994; Crowhurt et al., 1991; GraJai-Martin, et al., 1993; Guthrie et al., 1992; Huff et al., 1994; Hyun et al., 1996; Manulis et

al., 1994; Nicholson et al., 1993). It is not known by this study whether these genetic and morphological differences can classify the fungal isolates into different groups at species level or infraspecies level.

Future studies. More studies such as examination of toxin production and other physiological or molecular characters of the isolates should be conducted to give information on the diversity of the fungus. Virulence of the isolates on various cactus plants varied, but seems not to be specifically related to either colony types or conidial types.

Also with a couple of exceptions, conidial types (colony types) could not differentiate according to isolation areas and host plant species. These results suggest that the isolates with different conidial shapes cannot be physiologically separated to different groups. It may be not possible to trace the origin of each fungus, because the same cactus species or location may have different origins of plant introduction.

In another *Bipolaris* species such as *B. zeicola* (Telimorph *Cochliobolus carbonum*), three races have different virulence on corn, but are not morphologically distinguishable (Cassini and Smith, 1988). No difference was found among the morphologically different *B. cactivora* isolates in our study (Table 2), suggesting that the two potential biotypes may not be related to pathotype. Their genetic and morphological differences simply suggest the differences in originality. It is important to know the potential biotype diversification of pathogens in controlling diseases, especially in terms of pathogenicity and host specificity.

However, in *B. cactivora*, the potentially two biotypes were not related to these parameters, but may be differentiated upon other physiological and ecological characteristics. More studies are needed to verify the relatedness between the morphological differences and disease-causing natures for efficiently controlling the cactus stem rot.

References

Alcorn, J. L. 1983. Generic concepts in *Drechslera, Bipolaris* and *Exserohilum*. *Mycotaxon* 17:1-86.

Alcorn, J. L. 1988. The taxonomy of "*Helminthosporium*" species. *Ann. Rev. Phytopathol.* 26:37-56.

Assigbetse, K. B., Fernadez, M. P. Dubois and Geiger, J.P. 1994. Differentiation of *Fusarium oxysporum* f. sp. *vasinfectum* races on cotton by random amplified polymorphic DNA (RAPD) analysis. *Phytopathology* 84:622-626.

Cassini, R. and Smith, I. M. 1988. *Cochliobolus carbonum*. In: *European Handbook of Plant Diseases*, ed. by I.M. Smith, J. Dunez, D.H. Phillips, and S.A. Archer. Blackwell Scientific Publications, Oxford, UK. 382 pp.

Chang, M., Hyun, I. H. and Lee, Y. H. 1998. Bipolaris stem rot of cactus by *Bipolaris cactivora* (Petrak) Alcorn. *Korean J. Plant Pathol.* 14:661-663.

Chase, A. R. 1982. Stem rot and shattering of Easter cactus caused by *Drechslera cactivora*. *Plant Dis.* 66:602-603.

Crowhurt, R. N., Hawthorne, B. T., Rikkerink, E. H. A. and Templeton, M. D. 1991. Differentiation of *Fusarium solani* f. sp. *cucurbitate* races 1 and 2 by random amplification of polymorphic DNA. *Curr. Gent.* 20:391-396.

Durbin, D. R., Davis, H. L. and Baker, F. K. 1955. A Helminthosporium stem rot of cacti. *Phytopathoogy* 45:509-512.

GraJai-Martin, M. J., Simon, C. J. and Muehlbauer, F. J. 1993. Use of random amplified polymorphic DNA (RAPD) to characterize race 2 of *Fusarium oxysporum* f. sp. *pisi*. *Phytopathology* 83:612-614.

Guthrie, P. A. I., Magill, C. W., Frederiksen, R. A. and Odvody, G. N. 1992. Random amplified polymorphic DNA markers: A system for identifying and differentiating isolates of *Colletotrichum graminicola*. *Phytopathology* 82:832-835.

Hawksworth, D. L., Kirk, O. M., Sutton, B. C. and Pegler, D. N. 1995. *Anisworth & Bisbys Dictionary of the Fungi*. 8th ed. CAB International, Surrey. 616 pp.

Huff, D. R., Bunting, T. E. and Plumley, K. A. 1994. Use of random amplified polymorphic DNA markers for the detection of genetic variation on *Magnaporthe poae*. *Phytopathology* 84:1312-1316.

Hyun, I. H., Lee, S. D., Lee, Y. H. and Heo, N. Y. 1998. Mycological characteristics and pathogenicity of *Fusarium oxysporum* Schlecht. Emend. Snyd. & Hans. causing stem rot of cactus. *Korean J. Plant Pathol.* 14:463-466.

Hyun, J. W. and Park, W. M. 1996. Differentiation of *Fusarium oxysporum* f. sp. *fragariae* isolates by random amplified polymorphic DNA (RAPD) analysis. *Korean J. Plant Pathol.* 12:41-46.

Kim, W. G., Cho, W. D. and Jee, H. 2000. Occurrence of anthracnose on Indian fig cactus caused by *Glomerella cingulata* and *Colletotrichum gloeosporioides*. *Plant Pathol. J.* 16:294-296.

Kim, Y. H., Jun, O. K., Sung, M. J., Shin, J. S., Kim, J. H. and Jeong, M. 2000. Occurrence of Colletotrichum stem rot caused by *Glomerella cingulata* on Graft-cactus in Korea. *Plant Pathol. J.* 16:242-245.

Manulis, S., Kogan, N., Reuven, M. and Ben-Yephet, Y. 1994. Use of the RAPD technique for identification of *Fusarium oxysporum* f. sp. *dianthi* from carnation. *Phytopathology* 84:98-101.

Nicholson, P., Rezanoor, H. N. and Su, H. 1993. Use of random amplified polymorphic DNA (RAPD) analysis and genetic fingerprinting to differentiate isolates of race O, C and T of *Bipolaris maydis*. *J. Phytopathol.* 139:261-267.

Rohlf, F. J. 1993. *NTSYS-pc, Numerical Taxonomy and Multivariate Analysis System*. State Univ. of New York, Stony Brook.

Sivanesan, A. 1987. Graninicolous species of *Bipolaris, Curvularia, Drechslera, Exserohilum* and their teleomorphs. CAB International Mycol. Papers 158:1-261.