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Diversity and Physiological Properties of Root Endophytic Actinobacteria in Native Herbaceous Plants of Korea

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Endophytic actinobacterial diversity in the native herbaceous plant species of Korea was analyzed using a culture-based approach. Sixty one actinobacterial strains were isolated, and assigned to 15 genera based on 16S rRNA gene analysis. The members of the genus *Streptomyces* comprised 45.9% of the total isolates, followed by *Micromonospora* (18.8%), *Rhodococcus* (6.6%), *Microbispora* (4.9%), and *Micrococcus* (4.9%). Other minor constituents included members of *Microbacterium*, *Streptacidiphilus*, *Arthrobacter*, *Dietzia*, *Kitasatospora*, *Herbiconiux*, *Mycobacterium*, *Nocardia*, *Rathayibacter*, and *Tsukamurella*. Among the isolates, 65.6% exhibited at least one hydrolytic enzyme activity out of four, and 45.9% exhibited antagonistic activity against at least one fungal pathogen out of five, thus demonstrating that endophytic actinobacteria can be an important source of bioactive compounds. Notably, most strains of *Streptomyces* proved active for both enzymatic and antagonistic activities.

Keywords: endophytic actinobacteria, native herbaceous plant, *Streptomyces*, *Rhodococcus*

Introduction

Endophytic bacteria are bacteria that reside within the internal tissue of plants, and for the whole or part of their life history live within plant tissues via symbiotic, parasitic, or mutualistic means without causing immediately overt negative effects. The beneficial interactions between the endophytic bacteria and plant hosts have been well studied (Stone *et al.*, 2000; Rosenblueth and Martínez-Romero, 2006; Ryan *et al.*, 2008).

Several mechanisms for plant growth promotion by microorganisms are suggested, including the facilitation of uptake of nutrients such as phosphorus, nitrogen fixation for plant use, sequestration of iron for plants by siderophores, production of plant hormones such as auxins, cytokinins, and gibberellins, lowering of plant ethylene levels, antagonization of plant-pathogenic microbes by reducing the iron available to phytopathogens in the rhizosphere, synthesis of fungal cell-wall-lysing enzymes, and competition with detrimental microorganisms (Coombs and Franco, 2003; Selosse *et al.*, 2004; Rosenblueth and Martínez-Romero, 2006; Shin *et al.*, 2007).

Various members of actinobacteria have been identified as the main constituents of the endophytic bacterial community (Park *et al.*, 2005; Rosenblueth and Martinez-Romero, 2006; Tian *et al.*, 2007; Lee *et al.*, 2008; Yuan *et al.*, 2008; Verma *et al.*, 2009; Wu *et al.*, 2009; Marquez-Santacruz *et al.*, 2010; Zhao *et al.*, 2011). Endophytic actinobacteria are also an important source of various natural products (Strobel *et al.*, 2004; Hasegawa *et al.*, 2006; Ryan *et al.*, 2008; Qin *et al.*, 2011). Actinobacteria encompass bacterial groups that are rich in guanine plus cytosine in genomic DNA, and are also well known for the decomposition of organic matter and for the production of a diverse range of secondary metabolites, including various antibiotics, antitumor and immunosuppressive agents and plant growth hormones (Locci, 1989; Bérdy, 2005). Endophytic actinobacteria can, therefore, be a promising source of biocontrol agents.

In this study, the diversity of the endophytic actinobacterial communities in the root of representative native herbaceous plants in Korea was assessed, and the physiological properties of the actinobacterial isolates were evaluated.

Materials and Methods

Plant sampling

Root samples of Korean native herbaceous plants were collected from Daejeon and Gongju in Chungnam Province, and Goesan and Youngdong in Chungbuk Province between April 2009 and August 2010. The samples were maintained at 4°C and transported to the laboratory for immediate analysis.

Isolation of endophytic actinobacteria

The root samples were washed, sterilized and homogenized in accordance with the previously described procedures (Park *et al.*, 2005). Five gram portions of homogenized root were resuspended in sterile Ringer's solution, then incubated at 30°C in a reciprocal shaker for 30 min. The suspension was subsequently diluted and inoculated on starch

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Table 1. List of endophytic actinobacterial isolates

Plant species (Common name)	Strain	16S rRNA accession number	Closest species	16S rRNA Similarity (%)[a]
Artemisia princeps var.*orientalis* (Mugwort)	SS04-01	JN120913	*Herbiconiux ginsengi* wged11^T	99.9
	SS04-02	JN120914	*Micrococcus yunnanensis* YIM 65004^T	99.9
	SS04-03	JN120915	*Rhodococcus globerulus* DSM 4954^T	99.7
	SS04-04	JN120916	*Rhodococcus cercidiphylli* YIM 65003^T	98.6
	SS04-05	JN120917	*Mycobacterium fluoranthenivorans* FA-4^T	100.0
	SS05-01	JN120918	*Streptomyces griseoflavus* LMG 19344^T	99.2
Capsella bursa-pastoris (Shepherd's purse)	SH05-01	JN120966	*Streptomyces bobili* JCM 4624^T	99.1
	SH05-02	JN120967	*Microbispora amethystogenes* JCM 3021^T	99.2
	SH05-03	JN120968	*Microbispora amethystogenes* JCM 3021^T	99.3
	SH05-04	JN120969	*Streptacidiphilus anmyonensis* AM-11^T	100.0
	SH05-05	JN120970	*Nocardia asteroides* ATCC 19247^T	99.8
	SH05-06	JN120971	*Rhodococcus qingshengii* djl-6^T	99.9
	SH05-07	JN120972	*Rhodococcus qingshengii* djl-6^T	99.8
Chelidonium majus var. *asiaticum* (Greater celandine)	CM05-01	JN120973	*Streptacidiphilus anmyonensis* AM-11^T	99.9
Conyza canadensis (Horse-weed)	HW04-01	JN120919	*Rathayibacter festucae* DSM 15932^T	99.9
	HW04-02	JN120920	*Microbacterium hydrocarbonoxydans* DSM 16089^T	99.8
	HW04-03	JN120921	*Microbacterium maritypicum* DSM 12512^T	99.9
	HW04-06	JN120922	*Streptomyces cacaoi* subsp. *asoensis* NRRL B-16592^T	100.0
	HW04-07	JN120923	*Streptomyces griseorubiginosus* LMG 19941^T	99.8
	HW04-08	JN120924	*Streptomyces cacaoi* subsp. *asoensis* NRRL B-16592^T	100.0
	HW04-09	JN120925	*Streptomyces griseorubiginosus* LMG 19941^T	100.0
	HW04-10	JN120926	*Streptomyces cacaoi* subsp. *asoensis* NRRL B-16592^T	100.0
	HW04-11	JN120927	*Streptomyces cacaoi* subsp. *asoensis* NRRL B-16592^T	100.0
	HW04-12	JN120928	*Streptomyces griseorubiginosus* LMG 19941^T	100.0
	HW04-13	JN120929	*Streptomyces griseorubiginosus* LMG 19941^T	100.0
	HW05-01	JN120930	*Micromonospora carbonacea* DSM 43815^T	98.6
	HW05-02	JN120931	*Micromonospora tulbaghiae* TVU1^T	100.0
	HW05-03	JN120932	*Micromonospora aurantiaca* DSM 43813^T	100.0
	HW05-04	JN120933	*Micromonospora aurantiaca* DSM 43813^T	100.0
	HW05-05	JN120934	*Micromonospora aurantiaca* DSM 43813^T	100.0
	HW05-06	JN120935	*Micromonospora aurantiaca* DSM 43813^T	100.0
	HW05-07	JN120936	*Micromonospora echinospora* 1ATCC 15837^T	99.6
	HW05-08	JN120937	*Micrococcus antarcticus* T2^T	99.4
	HW05-09	JN120938	*Micromonospora echinospora* 1ATCC 15837^T	99.6
	HW05-10	JN120939	*Micrococcus antarcticus* T2^T	99.6
	HW05-11	JN120940	*Micromonospora marina* JSM1-1^T	99.7
Erigeron annuus(Daisy fleabane)	DF09-01	JN120952	*Streptomyces lanatus* NBRC 12787^T	99.0
	DF09-02	JN120953	*Streptomyces griseorubiginosus* LMG 19941^T	99.6
	DF09-03	JN120954	*Streptomyces capoamus* JCM 4734^T	98.9
	DF09-04	JN120955	*Streptomyces griseorubiginosus* LMG 19941^T	99.6
	DF09-05	JN120956	*Streptomyces caeruleatus* GIMN4.002^T	99.1
Iris rossii var.*rossii* (Caudate-bracted iris)	IR04-01	JN120942	*Tsukamurella suncheonensis* SCNU5^T	99.9
	IR04-02	JN120943	*Streptomyces scabrisporus* NBRC 100760^T	99.2
	IR04-03	JN120944	*Streptomyces variegatus* LMG 20315^T	99.4
	IR04-04	JN120945	*Streptomyces variegatus* LMG 20315^T	99.4
	IR04-05	JN120946	*Streptomyces variegatus* LMG 20315^T	99.4
Lamium purpureum (Purple henbit)	PH04-01	JN120947	*Arthrobacter humicola* KV-653^T	99.9
	PH04-02	JN120948	*Kitasatospora viridis* 52108a^T	99.6
	PH04-03	JN120949	*Streptomyces murinus* NBRC 12799^T	98.4
	PH04-04	JN120950	*Streptomyces olivochromogenes* NBRC 3178^T	99.9
	PH04-05	JN120951	*Streptomyces olivochromogenes* NBRC 3178^T	99.6
Physostegia virginiana (Obedient plant)	PY09-02	JN120961	*Micromonospora matsumotoense* IMSNU 22003^T	98.7
	PY09-03	JN120962	*Micromonospora matsumotoense* IMSNU 22003^T	98.7
Rudbeckia bicolor (Pinewoods coneflower)	PW09-01	JN120957	*Streptomyces prunicolor* NRRL B-12281^T	99.7
	PW09-02	JN120958	*Streptomyces mirabilis* NBRC 13450^T	99.4
	PW09-03	JN120959	*Streptomyces prunicolor* NRRL B-12281^T	99.6
	PW09-05	JN120960	*Streptomyces prunicolor* NRRL B-12281^T	99.8
Setaria viridis (Green bristlegrass)	GB09-1	JN120963	*Microbispora rosea* subsp. *rosea* IFO 14044^T	99.7
	GB09-2	JN120964	*Streptomyces lanatus* NBRC 12787^T	99.0
	GB09-3	JN120965	*Streptomyces lanatus* NBRC 12787^T	99.0
Viola mandshurica (Manchurian violet)	MV04-01	JN120941	*Dietzia maris* DSM 43672^T	99.9

[a] Similarity from the identification results using EzTaxon (Chun *et al*., 2007).

casein agar (SCA; Küster and Williams, 1964) supplemented with cycloheximide and nystatin (final concentration of 50 μl/ml each), and the plates were incubated at 30°C for up to 10 days. Based on the morphological characteristics, actinobacteria were selected and subcultured using the same medium.

16S rRNA gene sequencing and phylogenetic analysis

The extraction of total genomic DNA from the isolated strains and PCR amplification were conducted via the previously described procedures (Park *et al*., 2005). The sequence determination of the amplified PCR products was conducted using the services of Macrogen (Korea) and Solgent (Korea). Taxonomic identification of the obtained sequences was carried out using the EzTaxon server 2.1 (Chun *et al*., 2007). The sequences were aligned together with the related reference species using the PHYDIT program version 3.0 (http://plaza.sun.ac.kr/~jchun/phydit). Phylogenetic trees were inferred by the neighbor-joining algorithm using the PHYLIP 3.5c package (http://evolution.genetics.washington.edu/phylip.html). Evolutionary distances for the tree were generated on the basis of the Jukes-Cantor model (Jukes and Cantor, 1969). The bootstrap analysis based on 1,000 resampled datasets was also conducted using the PHYLIP package. The sequences obtained in this study were deposited into the GenBank database under the accession numbers JN120913–JN120973.

Antagonistic activities against plant pathogenic fungi

To assess antagonistic effects against fungal pathogens, the bacterial isolates were streaked onto one side of a Petri dish (1 cm from the edge) containing potato dextrose agar (PDA, Difco). Five fungal pathogens, namely *Alternaria alternata* KACC 42131, *Colletotrichum gloeosporioides* KACC 40003, *Fusarium oxysporum* KACC 41083, *Fusarium solani* KACC 41093, and *Rhizoctonia solani* KACC 40113, were obtained from the Korean Agricultural Culture Collection (KACC), cultured on PDA medium for 10 days, transferred to one side of the Petri dish perpendicular to the bacterial streak, and incubated for 4 days at 25°C. The inhibition zone was recorded by measuring the distance (mm) between the edge of the fungal mycelium and the bacterial streak. All strains were tested in three independent replicates.

Production of hydrolytic enzymes and secondary metabolites

Enzyme activities of bacteria that can promote plant growth or inhibit pathogens were tested. Chitinase activity was tested using the minimal medium of Chernin *et al.* (1995), cellulase activity using the CMC medium of Criquet (2002), protease activity indicated by casein degradation using the skim milk agar (50 ml sterilized skimmed milk mixed with 50 ml of 1/5 tryptic soy agar, the final agar concentration adjusted to 2%), and phosphate-solubilizing activity using the chemically defined medium (NBRIP) of Sulbarán *et al.* (2008). Clearing zones were detected after incubation at 30°C for 7 days for all tests except for protease activity (5 days). The production of indole-3-acetic acid (IAA), a plant growth hormone, was determined using the microplate method developed by Sawar and Kremer (1995).

Results

Identification of endophytic actinobacteria

Sixty-one actinobacterial strains were isolated from the roots of 11 native herbaceous plant species (Table 1). The isolates could be assigned to 15 different genera on the basis of 16S rRNA gene sequence analyses (Figs. 1A and 1B). *Streptomyces* was the most common group, accounting for 45.9% of the total endophytic isolates (28 strains), followed by *Micromonospora* (11 strains), *Rhodococcus* (4 strains), *Microbispora* (3 strains), *Micrococcus* (3 strains), *Microbacterium* (2 strains), and *Streptacidiphilus* (2 strains). Single strains were isolated for each of the genera *Arthrobacter*, *Dietzia*, *Herbiconiux*, *Kitasatospora*, *Mycobacterium*, *Nocardia*, *Rathayibacter*, and *Tsukamurella*. Diversity could be seen at the species level among the isolates belonging to the same genus.

Strains affiliated with *Streptomyces griseorubiginosus* were the most common (6 strains), followed by strains affiliated with *Micromonospora aurantiaca* (4 strains), *Streptomyces cacaoi* subsp. *asoensis* (4 strains), *Streptomyces lanatus* (3 strains), *Streptomyces prunicolor* (3 strains), and *Streptomyces variegatus* (3 strains). Strains affiliated with *S. griseorubiginosus*, *S. lanatus*, and *Streptacidiphilus anmyonensis*, respectively, were isolated from two different plant species.

Notably, a large number of strains belonging to *Micromonospora* were isolated from *Conyza canadensis*, and strains of *Rhodococcus* were from *Artemisia princeps* var. *orientalis* and *Capsella bursa-pastoris*.

Antagonistic activity against fungal pathogens

Among the 61 endophytic actinobacterial isolates, 28 strains exhibited antifungal activity towards at least one fungal pathogen (Table 2). Twenty one strains exhibited antagonistic activity against *Phytophthora capsici*, 11 strains against *Rhizoctonia solani*, 19 strains against *Colletotrichum gloeosporioides*, 14 strains against *Fusarium solani*, and 13 strains against *Alternaria alternata*, respectively. Notably, four strains affiliated with *S. griseorubiginosus*, HW04-12, HW04-13, DF09-02, and DF09-04, strain HW04-08 affiliated with *S. cacaoi* subsp. *asoensis*, and strain PH04-03 affiliated with *S. murinus* exhibited antagonistic activities against all 5 fungal pathogens tested.

Production of hydrolytic enzymes and indole acetic acid

Twenty strains exhibited protease activity, 1 strain phosphatase activity, 18 strains chitinase activity, and 31 strains cellulase activity, respectively (Table 2). As a whole, 40 strains exhibited at least one enzyme activity. Notably, four strains of *Micromonospora* (HW05-01, HW05-02, HW05-05, and HW05-11) and three strains of *Streptomyces* (PH04-03, SH05-01, and SS05-01) exhibited cellulase, chitinase, and protease activities, although not phosphatase activity. Most strains were poor producers of phosphatase. Four strains were found to be prominent indole acetic acid producers, namely *Streptomyces* sp. DF09-05, *Streptomyces* sp. GB09-03, *Streptomyces* sp. DF09-04, and *Micrococcus* sp. HW05-10.

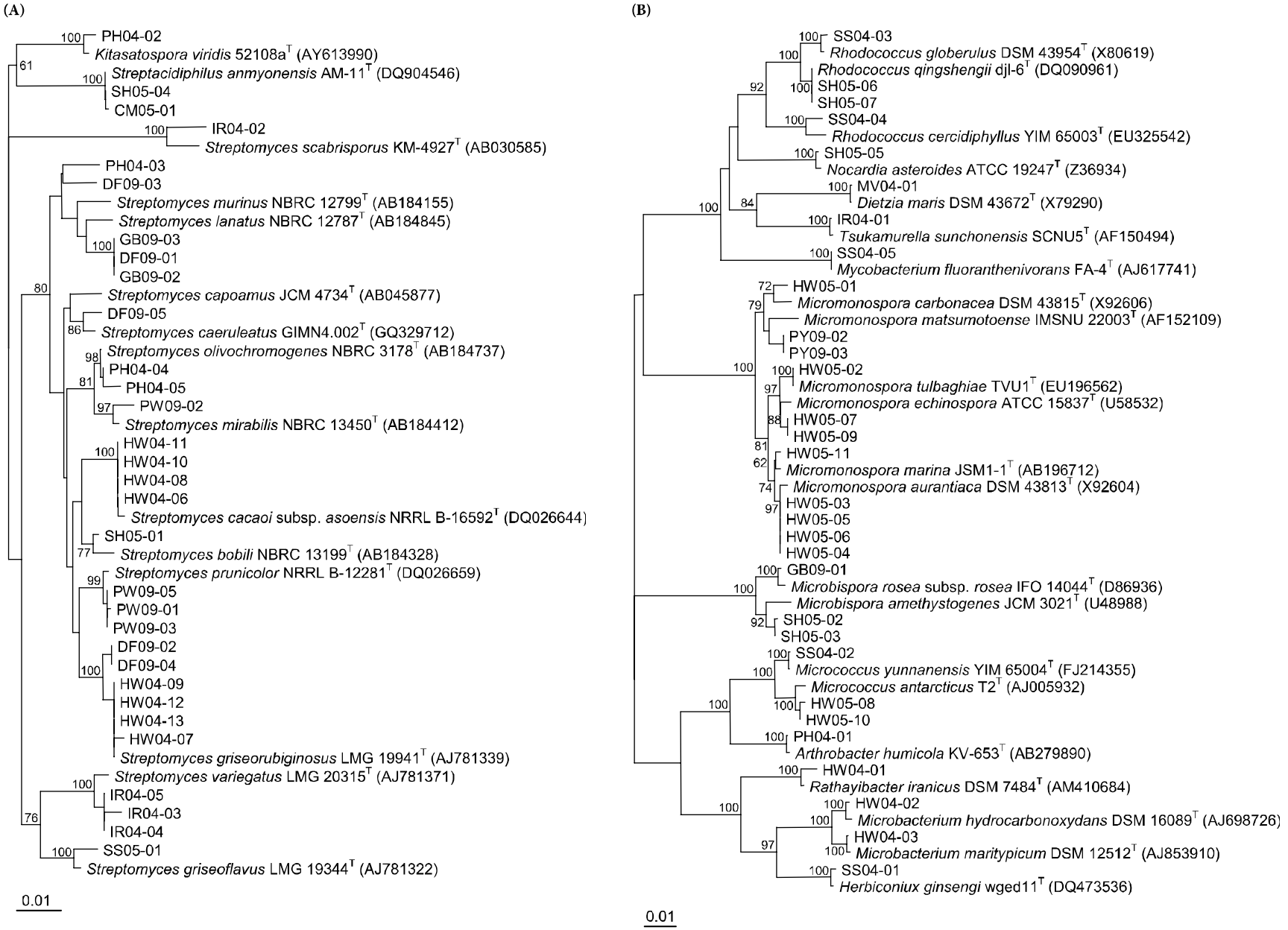

Fig. 1. Phylogenetic trees of root endophytic actinobacterial isolates based on 16S rRNA gene sequences. (A) members of the family *Streptomycetaceae*. (B) members of non-*Streptomycetaceae*. The numbers at nodes are the bootstrap support (%) between the isolates and reference strains. Only the values over 60% are given. Scale bar corresponds to 0.01 substitutions per nucleotide position.

Table 2. Plant growth promoting potential of endophytic actinobacteria

Strain	Production of:					Antagonistic against:				
	Chitinase	Cellulase	Protease	Phosphatase	Indole acetic acid	*P. capsici*	*R. solani*	*C. gloeosporioides*	*F. solani*	*A. alternata*
Kitasatospora sp.										
PH04-02	-	+	-	-	-	+	-	-	-	-
Microbispora sp.										
SH05-02	-	-	+	-	-	-	-	-	-	-
SH05-03	-	-	+	-	-	-	-	-	-	-
Micrococcus sp.										
HW05-10	-	+	+++	-	+	-	-	+	-	-
Micromonospora sp.										
HW05-01	++	+++	+	-	-	-	-	-	-	-
HW05-02	++	+	++	-	-	-	-	-	-	-
HW05-03	-	-	+	-	-	+	-	+	-	-
HW05-04	-	+	+	-	-	-	-	-	-	-
HW05-05	++	+	+	-	-	-	-	-	-	-
HW05-06	-	+	-	-	-	-	-	-	-	-
HW05-07	-	-	-	-	-	+	-	+	-	-
HW05-09	-	++	++	-	-	+	-	-	-	-
HW05-11	++	++	++	-	-	-	-	-	-	-
PY09-02	-	+	+++	-	-	-	-	-	-	-
Rhodococcus sp.										
SH05-06	-	-	-	-	-	-	-	+	-	-
SH05-07	-	-	-	-	-	-	-	+	-	-
Streptacidiphilus sp.										
CM05-01	++	+++	-	-	-	+	-	+	+	-
SH05-04	-	+++	-	-	-	-	-	-	-	-
Streptomyces sp.										
DF09-01	-	++	-	-	-	+	-	+	-	-
DF09-02	++	++	-	-	-	++	++	++	++	++
DF09-03	+	++	-	-	-	++	-	+	-	+
DF09-04	-	++	-	-	+	+	++	++	++	++
DF09-05	-	+++	-	-	+	-	-	-	-	-
GB09-02	-	+	-	-	-	++	++	+	-	-
GB09-03	-	++	-	-	+	+	-	++	+	-
HW04-06	+++	+++	-	-	-	+*	++	-	+	++
HW04-07	-	-	+++	-	-	-	-	-	+	-
HW04-08	+++	+	-	-	-	+	++	++	++	++
HW04-09	++	-	-	+	-	-	-	-	-	+
HW04-10	++	-	-	-	-	-	-	-	-	-
HW04-12	+++	+	-	-	-	++	++	++	++	++
HW04-13	-	++	++	-	-	++	++	++	++	++
IR04-02					-	+	+	++	+	-
IR04-03	-	+	-	-	-	-	-	-	-	-
IR04-05	-	++	+	-	-	-	-	-	-	-
PH04-03	++	+++	++	-	-	++	++	++	++	++
PH04-04	++	-	+	-	-	+	-	-	-	-
PH04-05	-	+++	+	-	-	-	-	-	-	-
PW09-01	++	-	-	-	-	-	-	+	+	+
PW09-02	++	-	+	-	-	-	-	-	-	-
PW09-03	-	+	-	-	-	-	-	-	-	+
PW09-05	-	+++	-	-	-	-	-	-	-	-
SH05-01	+	+++	+	-	-	-	-	-	-	-
SS05-01	++	+	+++	-	-	-	-	-	-	-

-, Negative; +, the size of clear zone between 3 and 5 mm; ++, between 6 and 10 mm; +++, greater than 10 mm.

Discussion

The dominance of *Streptomyces* spp. in the culturable diversity of root endophytic actinobacteria is consistent with most of the previous observations from various plant species (Coombs and Franco, 2003; Tian *et al.*, 2007; Yuan *et al.*, 2008; Verma *et al.*, 2009; Wu *et al.*, 2009; Zhao *et al.*, 2011). In particular, the isolation of a higher number of strains

close to *S. griseorubiginosus* is consistent with the report of Cao *et al.* (2004). The presence of *Micromonospora* (Coombs and Franco, 2003; Lee *et al.*, 2008; Yuan *et al.*, 2008; El-Tarabily *et al.*, 2009; Zhao *et al.*, 2011), *Rhodococcus* (Idris *et al.*, 2004; Sturz and Kimpinski, 2004; Zhao *et al.*, 2011), *Microbispora* (Coombs and Franco, 2003; Lee *et al.*, 2008; Verma *et al.*, 2009), *Micrococcus* (Sturz and Kimpinski, 2004; Aravind *et al.*, 2009) and *Microbacterium* (Zinniel *et al.*, 2002; Sturz and Kimpinski, 2004; Park *et al.*, 2005; Burch and Saranthchandra, 2006; Zakhia *et al.*, 2006; Marquez-Santacruz *et al.*, 2010) was also confirmed in previous studies. However, there has been no report thus far on the presence of endophytic *Streptacidiphilus*. For the genera for which single strains were isolated, the presence of *Arthrobacter* (Sturz and Kimpinski, 2004; Aravind *et al.*, 2009), *Dietzia* (Qin *et al.*, 2009), *Herbiconiux* (Qiu *et al.*, 2007), *Mycobacterium* (Deng *et al.*, 2011), *Nocardia* (Coombs and Franco, 2003; Sturz and Kimpinski, 2004; Kaewkla and Franco, 2010; Deng *et al.*, 2011), *Rathayibacter* (Evtushenko and Takeuchi, 2003), and *Tsukamurella* (Sturz and Kimpinski, 2004) have also been confirmed. *Kitasatospora* has not been reported as a cultured endophyte, but its presence was previously reported through a terminal restriction fragment length polymorphism analysis (Conn and Franco, 2004).

Strains of the genus *Streptomyces* were largely active against fungal pathogens, as 60.1% of the total *Streptomyces* isolates exhibited antagonistic activity against at least one fungal pathogen (Table 2). Most strains of *Streptomyces* were also positive for at least one hydrolytic enzyme activity, notably cellulase (67.9% of the strains). Most *Micromonospora* strains were positive for cellulase and protease activities (72.7% each), but only 27.3% of the strains exhibited antagonism, against either *P. capsici* or *C. gloeosporioides*.

No noticeable correlation was observed between the enzyme production and antagonistic activity. When the phi correlation coefficients were calculated from the method of Shin *et al.* (2007), the value of 0.37 between chitinase production and antagonism against *A. alternata* was the highest, but was still indicative of a low level of correlation between the two properties. It was also worth noting that protease displayed negative phi coefficients with all five antagonistic activities (-0.24~-0.39), thereby indicating that the possession of protease activity is correlated negatively with the antagonism against fungi. In contrast to a previous observation (Shin *et al.*, 2007), little correlation between the production of individual enzymes was observed.

Streptacidiphilus is known to inhabit acidic environments, including forest soil and rhizosphere (Kim *et al.*, 2003; Huang *et al.*, 2004; Wang *et al.*, 2006; Cho *et al.*, 2006, 2008); thus it should not be surprising to find an interaction between these organisms and plants. This is supported, in part, by the fact that both of the two streptacidiphili isolates in this study exhibited strong cellulase activity, and one strain exhibited antagonistic activity against three fungal species (Table 2). Moreover, *Streptomyces* and *Kitasatospora*, the taxonomically closest neighbors, are already known as the principal constituents of the endophytic bacterial community, as explained above.

To the best of our knowledge, this is the first report regarding the diversity of endophytic actinobacteria and their physiological properties in various Korean native plant species, although there have been some studies conducted on selected native and crop plants (Park *et al.*, 2005; Cho *et al.*, 2007; Shin *et al.*, 2007; Lee *et al.*, 2008; Vendan *et al.*, 2010). Using a culture-based approach, the members of the genus *Rhodococcus* and the family *Streptomycetaceae* could be recognized as the main constituents of endophytic actinobacterial community, which could also be confirmed by cultivation-independent methods (data not shown). Members of *Rhodococcus* were also found via culture-independent methods, though not as main constituents, in previous studies (Conn and Franco, 2004; Tian *et al.*, 2007). Most isolates belonging to *Streptomycetaceae* exhibited hydrolytic enzyme activities as well as antagonism against fungal pathogens, suggesting their roles in association with plant hosts. Although the strains of *Rhodococcus* were not active in the production of enzymes and antagonistic activity, unlike streptomycetes, previous studies suggest their positive roles in the relationship with plants (Sheng *et al.*, 2011; Zhao *et al.*, 2011).

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