

Report of 21 unrecorded bacterial species in Korea belonging to the phylum Actinobacteria, discovered during the survey in 2020

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The phylum Actinobacteria includes many groups of aerobic, Gram-stain-positive, rod, or filamentous shaped bacteria. Actinobacteria are known for multicellular differentiation in some groups, and also for production of various secondary metabolites such as antibiotics. During a series of extensive surveys of indigenous prokaryotic species diversity in Korea, bacterial strains belonging to Actinobacteria were isolated from various sources of terrestrial environments. A total of 21 bacterial strains, belonging to 10 genera in 8 families, were isolated as unrecorded species in Korea. Among them, 11 were assigned to the family *Streptomycetaceae*, two species assigned to each of the families *Microbacteriaceae*, *Mycobacteriaceae* and *Nocardioideaceae*, and one species assigned to each of the families *Euzebyaceae*, *Corynebacteriaceae*, *Micrococcaceae* and *Intrasporangiaceae*. At the genus level, *Streptomyces* (10 species) was the most abundant, followed by *Microbacterium* and *Mycolicibacterium* (2 species each), and one species in each of the genera *Corynebacterium*, *Euzebya*, *Arthrobacter*, *Terracoccus*, *Kribbella*, *Nocardioides* and *Yinghuangia*. The detailed descriptions of each unrecorded species are provided.

Keywords: Actinobacteria, *Streptomyces*, unrecorded species

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INTRODUCTION

The phylum Actinobacteria is one of the ubiquitous bacterial groups (Goodfellow *et al.*, 2012; Kim *et al.*, 2021). Actinobacteria includes a large number of Gram-stain-positive, rod or filamentous species with diverse morphological features, and there are currently 6 classes, 32 orders, 55 families and more than 400 genera (List of Prokaryotic Names with Standing in Nomenclature, as of July 2021).

Members of Actinobacteria are widely distributed in diverse terrestrial and aquatic environments, and also within animals, plants and human. Actinobacteria are also well known for their secondary metabolites, notably antibiotics, and about two-thirds of currently used antibiotics come from Actinobacteria (Naikpatil and Rathod, 2011). *Streptomyces* is the representative group of Actinobacteria,

as members of *Streptomyces* are most abundant, particularly in soil, and major producers of antibiotics among Actinobacteria, while genera of Actinobacteria that do not belong to *Streptomyces* are referred to as 'rare' Actinobacteria (Amin *et al.*, 2020).

In a series of extensive surveys on the prokaryotic diversity in Korea in the year 2020, novel strains belonging to the phylum Actinobacteria were isolated from various environmental sources. As a result, a total of 21 unrecorded species of Actinobacteria were identified, and their taxonomic properties are presented.

MATERIALS AND METHODS

A total of 21 bacterial strains assigned to the phylum Actinobacteria were isolated from diverse environmen-

tal samples such as tidal flat, soil, sea sediment, healthy human urine and wetland (Table 1). The samples were processed or treated separately, diluted and spread onto diverse culture media, namely R2A, marine agar (MA), brain heart infusion (BHI) agar and nutrient agar (NA), and incubated at 25–37°C for 2–5 days. The designated strain codes, sources of isolation, culture media and incubation conditions are provided in the descriptions and Table 1. All strains were purified as single colonies and stored in 10–20% glycerol suspension at –80°C as well as lyophilized ampoules.

We observed the colony characteristics of isolates on the same agar media for cultivation. Cellular morphology and cell size were examined using either a transmission electron microscope or a scanning electron microscope. Gram staining was performed using standard procedures. Biochemical characteristics were tested by using API 20NE galleries (bioMérieux) according to the manufacturer's instructions.

The extraction of genomic DNA, PCR amplification of 16S rRNA gene and sequencing were performed using the procedures as described previously (Cho *et al.*, 2018; Lee *et al.*, 2018; Maeng *et al.*, 2018). The 16S rRNA gene sequences of the strains were compared to reference strains using the EzBioCloud (Kim *et al.*, 2012), and their 16S rRNA sequences were aligned using EzEditor2 (Jeon *et al.*, 2014). Phylogenetic trees were generated using the Jukes–Cantor distance model (Jukes and Cantor, 1969) and the neighbor-joining method (Saitou and Nei, 1987) in MEGA 7.0 (Kumar *et al.*, 2016). The support for clades was evaluated using 1,000 bootstrap replicates (Felsenstein, 1985).

RESULTS AND DISCUSSION

A total of 21 bacterial strains belonging to Actinobacteria were isolated (Table 1). The strains represented 21 unrecorded species in Korea, belonging to 11 genera in 8 families. Eleven species were assigned to *Streptomycetaceae*, while two species each were assigned to *Microbacteriaceae*, *Mycobacteriaceae* and *Nocardioideaceae*, and one species each assigned to the families *Euzebyaceae*, *Corynebacteriaceae*, *Micrococcaceae* and *Intrasporangiaceae*. At the genus level, *Streptomyces* (10 species) was the most abundant, and two species each were obtained for the genera *Microbacterium* and *Mycolicibacterium*. Single species were obtained for the remaining genera (Table 1). The electron microscopic images of the isolates are provided in Fig. 1, and the phylogenetic relationship between the isolates and closely related species are presented in Figs. 2 and 3. The detailed descriptions of each unrecorded species are provided as below.

Description of *Corynebacterium amycolatum* CAU 1619

Cells are Gram-stain-positive and rod-shaped. Colonies are irregular, flat, wavy and white colored after incubation for 2–3 days on BHI at 37°C. Positive for nitrate reduction and glucose fermentation; weakly positive for esculin hydrolysis; but negative for indole production, arginine dihydrolase, urease, gelatin hydrolysis, β -galactosidase and cytochrome oxidase. D-Glucose, D-mannose, D-mannitol, *N*-acetyl-glucosamine, D-maltose, potassium gluconate, adipic acid, malic acid, trisodium citrate and phenylacetic acid are utilized as sole carbon sources; but not L-arabinose and capric acid. Strain CAU 1619 (=NIBRBAC 000506277) was isolated from a healthy human urine sample from Seoul. The GenBank accession number for the 16S rRNA gene sequence of strain CAU1619 is MW020537.

Description of *Euzebya rosea* CAU 1611

Cells are Gram-stain-positive and rod-shaped. Colonies are circular, convex, smooth and light coral colored after incubation for 3–5 days on MA at 30°C. Positive for esculin hydrolysis, gelatin hydrolysis, β -galactosidase and cytochrome oxidase; but negative for nitrate reduction, indole production, glucose fermentation, arginine dihydrolase and urease. D-Glucose, L-arabinose, D-mannose, D-mannitol, *N*-acetyl-glucosamine, D-maltose, potassium gluconate, capric acid, adipic acid, malic acid, trisodium citrate and phenylacetic acid are not utilized as sole carbon sources. Strain CAU 1611 (=NIBRBAC000506272) was isolated from a sea sediment sample from Goseong-gun, Gangwon-do. The GenBank accession number for the 16S rRNA gene sequence of strain CAU 1611 is MW012619.

Description of *Terracoccus luteus* BT654

Cells are Gram-stain-positive and rod-shaped. Colonies are circular, convex and yellow colored after incubation for 3 days on R2A at 25°C. Positive for β -galactosidase; but negative for nitrate reduction, indole production, glucose fermentation, arginine dihydrolase, urease, esculin hydrolysis, gelatin hydrolysis and cytochrome oxidase. D-Glucose is utilized as a sole carbon source; but not L-arabinose, D-mannose, D-mannitol, *N*-acetyl-glucosamine, D-maltose, potassium gluconate, capric acid, adipic acid, malic acid, trisodium citrate and phenylacetic acid. Strain BT654 (=NIBRBAC000506198) was isolated from a soil sample from Wonju-si, Gangwon-do. The GenBank accession number for the 16S rRNA gene sequence of strain BT654 is MT993459.

Table 1. Summary of isolated strains belonging to the phylum Actinobacteria and their taxonomic affiliations.

Affiliation	Strain code	NIBR ^a ID	Identification	Similarity (%)	Isolation source	Medium ^b	Culture conditions
<i>Corynebacteriaceae</i>	CAU 1619	NIBRBAC000506277	<i>Corynebacterium amycolatum</i>	98.84	Human urine	BHI	37°C, 2–3 days
<i>Euzeyhaceae</i>	CAU 1611	NIBRBAC000506272	<i>Euzeyha rosea</i>	99.93	Sea sediment	MA	30°C, 3–5 days
<i>Intrasporangiaceae</i>	BT654	NIBRBAC000506198	<i>Terracoccus luteus</i>	99.79	Soil	R2A	25°C, 3 days
<i>Microbacteriaceae</i>	JHSTF-R7	NIBRBAC000506394	<i>Microbacterium saccharophilum</i>	99.38	Tidal flat	R2A	25°C, 5 days
	JHSTF-M27	NIBRBAC000506398	<i>Microbacterium halimionae</i>	99.86	Tidal flat	MA	25°C, 5 days
<i>Micrococcaceae</i>	MMS20-SJTN15	NIBRBAC000506164	<i>Arthrobacter alkaliphilus</i>	99.02	Soil	NA	30°C, 3 days
<i>Mycobacteriaceae</i>	CAU 1609	NIBRBAC000506270	<i>Mycolicibacterium duvalii</i>	99.65	Sea sediment	MA	30°C, 3–5 days
	CAU 1610	NIBRBAC000506271	<i>Mycolicibacterium hippocampi</i>	99.79	Sea sediment	MA	30°C, 3–5 days
<i>Nocardioideaceae</i>	XY6	NIBRBAC000506149	<i>Kribbella karoensis</i>	99.93	Wetland	R2A	30°C, 3 days
	BT677	NIBRBAC000506195	<i>Nocardioides plantarum</i>	98.97	Soil	R2A	25°C, 3 days
<i>Streptomycesetaceae</i>	MMS20-SJTR5	NIBRBAC000506162	<i>Streptomyces dangxiangensis</i>	99.28	Soil	R2A	30°C, 3 days
	MMS20-SJTR12	NIBRBAC000506163	<i>Streptomyces longisporus</i>	99.50	Soil	R2A	30°C, 3 days
	MMS20-HV2-26	NIBRBAC000506170	<i>Streptomyces spinoverrucosus</i>	99.24	Soil	ISP 2	30°C, 3 days
	MMS20-HV4-22	NIBRBAC000506172	<i>Streptomyces avermitilis</i>	98.96	Soil	ISP 2	30°C, 3 days
	MMS20-AI2-20	NIBRBAC000506174	<i>Streptomyces pseudo-griseolus</i>	99.86	Soil	ISP 2	30°C, 3 days
	DM17	NIBRBAC000506140	<i>Streptomyces scabiei</i>	99.72	Wetland	R2A	30°C, 3 days
	MA2	NIBRBAC000506141	<i>Streptomyces cinereospinus</i>	99.45	Wetland	R2A	30°C, 3 days
	MA30	NIBRBAC000506144	<i>Streptomyces albidoflavus</i>	100	Wetland	R2A	30°C, 3 days
	I2-3	NIBRBAC000506146	<i>Streptomyces puniceus</i>	99.93	Wetland	R2A	30°C, 3 days
	SM6	NIBRBAC000506150	<i>Streptomyces phaeofaciens</i>	98.76	Wetland	R2A	30°C, 3 days
	SO314	NIBRBAC000506232	<i>Yinghuangia aomienensis</i>	99.93	Soil	R2A	25°C, 5 days

^aNational Institute of Biological Resources.^bBHI, brain heart infusion agar; MA, marine agar; R2A, Reasoner's 2A agar; NA, nutrient agar; ISP 2, International Streptomyces Project 2 agar.

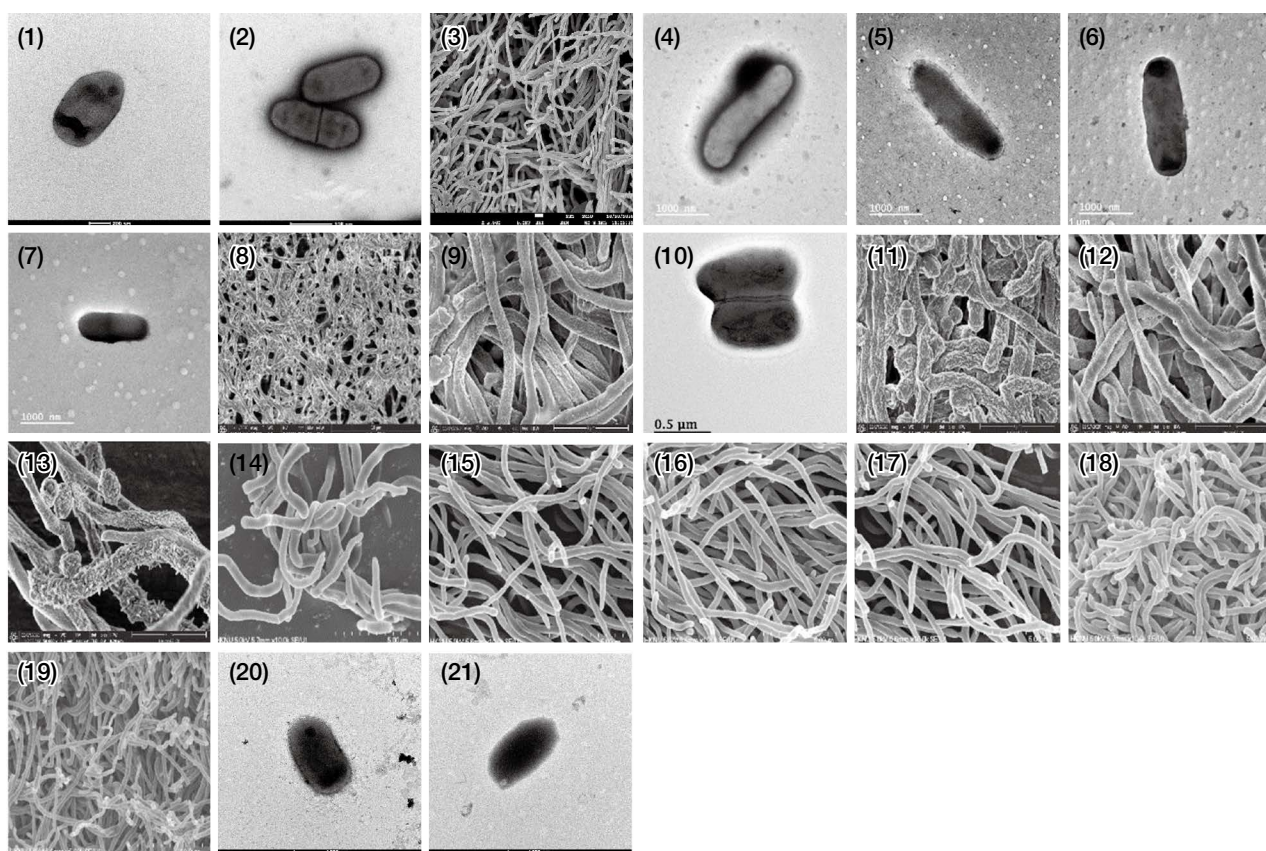


Fig. 1. Transmission electron or scanning electron micrographs of cells of the strains isolated in the study. Strains: 1, JHSTF-R7; 2, JHSTF-M27; 3, SO314; 4, CAU 1609; 5, CAU 1610; 6, CAU 1611; 7, CAU 1619; 8, MMS20-SJTR5; 9, MMS20-SJTR12; 10, MMS20-SJTN15; 11, MMS20-HV2-26; 12, MMS20-HV4-22; 13, MMS20-AI2-20; 14, DM17; 15, MA2; 16, MA30; 17, I2-3; 18, XY6; 19, SM6; 20, BT677; 21, BT654.

Description of *Microbacterium halimionae* JHSTF-M27

Cells are Gram-stain-negative and rod-shaped. Colonies are circular, convex, glistening and light yellow colored after incubation for 5 days on MA at 25°C. Positive for glucose fermentation, esculin hydrolysis and β -galactosidase; but negative for nitrate reduction, indole production, arginine dihydrolase, urease, gelatin hydrolysis and cytochrome oxidase. D-Glucose, L-arabinose, D-mannose, D-mannitol, *N*-acetyl-glucosamine, D-maltose, potassium gluconate, capric acid, adipic acid, malic acid, trisodium citrate and phenylacetic acid are not utilized as sole carbon sources. Strain JHSTF-M27 (= NIBRBAC000506398) was isolated from a tidal flat sample from Seocheon-gun, Chungcheongnam-do.

Description of *Microbacterium saccharophilum* JHSTF-R7

Cells are Gram-stain-negative, ovoid and rod-shaped. Colonies are circular, slightly convex, glistening and light

yellow colored after incubation for 5 days on at R2A media at 25°C. Positive for arginine dihydrolase, esculin hydrolysis and β -galactosidase; but negative for nitrate reduction, indole production, glucose fermentation, urease, gelatin hydrolysis and cytochrome oxidase. D-Maltose is utilized as a sole carbon source; but not D-glucose, L-arabinose, D-mannose, D-mannitol, *N*-acetyl-glucosamine, potassium gluconate, capric acid, adipic acid, malic acid, trisodium citrate and phenylacetic acid. Strain JHSTF-R7 (= NIBRBAC000506394) was isolated from a tidal flat sample from Seocheon-gun, Chungcheongnam-do.

Description of *Arthrobacter alkaliphilus* MMS20-SJTN15

Cells are Gram-stain-positive and V-shaped. Colonies are circular, convex, entire and yellow colored after incubation for 3 days on NA at 30°C. Positive for arginine dihydrolase, urease, β -galactosidase and cytochrome oxidase; weakly positive for glucose fermentation and esculin hydrolysis; but negative for nitrate reduction, indole pro-

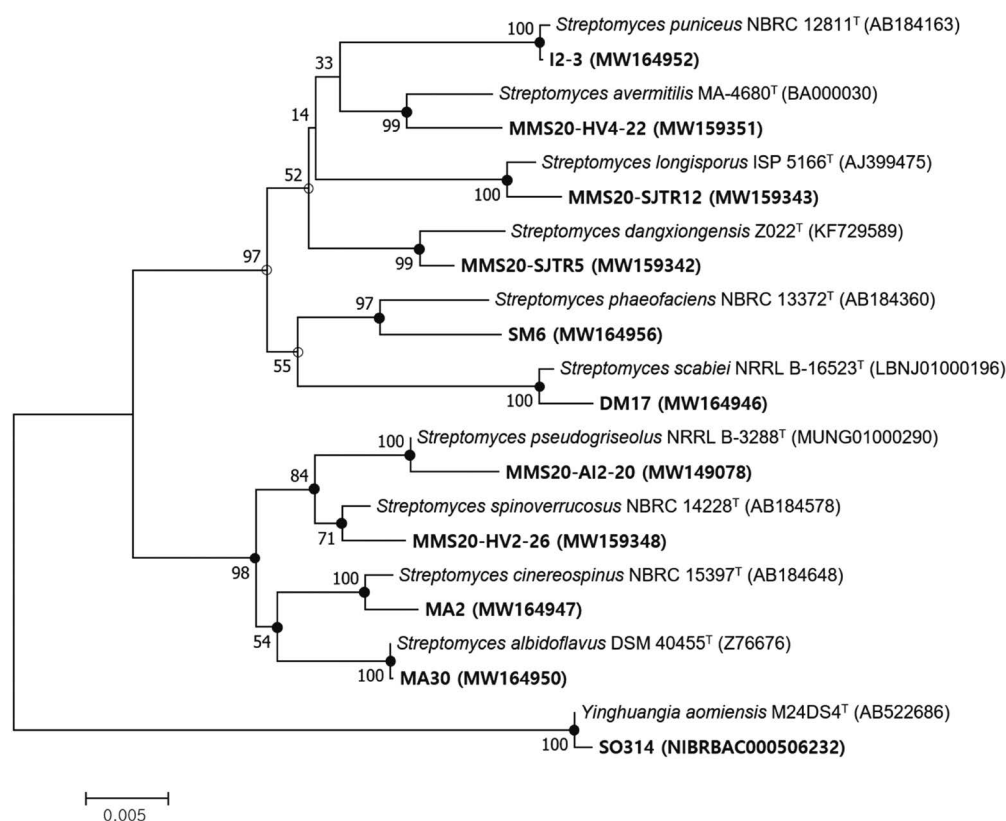


Fig. 2. Neighbor-joining phylogenetic tree based on 16S rRNA gene sequences showing the relationships between the strains isolated in this study and their relatives of the family *Streptomycetaceae* in the phylum Actinobacteria. Bootstrap values (>50%) are shown at branching points. Filled circles indicate the nodes also recovered in the maximum-likelihood and maximum-parsimony trees, and open circles indicate the nodes also recovered in only one of the trees. Bar, 0.005 substitutions per nucleotide position.

duction and gelatin hydrolysis. D-Glucose, D-mannose, D-mannitol, *N*-acetyl-glucosamine, D-maltose, potassium gluconate, malic acid, trisodium citrate, phenylacetic acid are utilized as sole carbon sources; but not L-arabinose, capric acid and adipic acid. Strain MMS20-SJTN15 (=NIBRBAC000506164) was isolated from a soil sample from Sejong-si. The GenBank accession number for the 16S rRNA gene sequence of strain MMS20-SJTN15 is MW159344.

Description of *Mycolicibacterium duvalii* CAU 1609

Cells are Gram-stain-positive and rod-shaped. Colonies are punctiform, convex, smooth and light yellow colored after incubation for 3–5 days on MA at 30°C. Positive for urease; weakly positive for esculin hydrolysis; but negative for nitrate reduction, indole production, glucose fermentation, arginine dihydrolase, gelatin hydrolysis, β -galactosidase and cytochrome oxidase. Malic acid is utilized as a sole carbon source; but not D-glucose, L-arabinose, D-mannose, D-mannitol, *N*-acetyl-glucosamine, D-maltose, potassium gluconate, capric acid, adipic acid, trisodium citrate and phenylacetic acid. Strain CAU 1609

(=NIBRBAC000506270) was isolated from a sea sediment sample from Goseong-gun, Gangwon-do. The GenBank accession number for the 16S rRNA gene sequence of strain CAU 1609 is MW012852.

Description of *Mycolicibacterium hippocampi* CAU 1610

Cells are Gram-stain-positive and rod-shaped. Colonies are punctiform, convex, smooth and yellow colored after incubation for 3–5 days on MA at 30°C. Weakly positive for esculin hydrolysis; but negative for nitrate reduction, indole production, glucose fermentation, arginine dihydrolase, urease, gelatin hydrolysis, β -galactosidase and cytochrome oxidase. D-Glucose, L-arabinose, D-mannose, D-mannitol, potassium gluconate, adipic acid and malic acid are utilized as sole carbon sources; but not *N*-acetyl-glucosamine, D-maltose, capric acid, trisodium citrate and phenylacetic acid. Strain CAU 1610 (=NIBRBAC000506271) was isolated from a sea sediment sample from Goseong-gun, Gangwon-do. The GenBank accession number for the 16S rRNA gene sequence of strain CAU 1610 is MW012620.

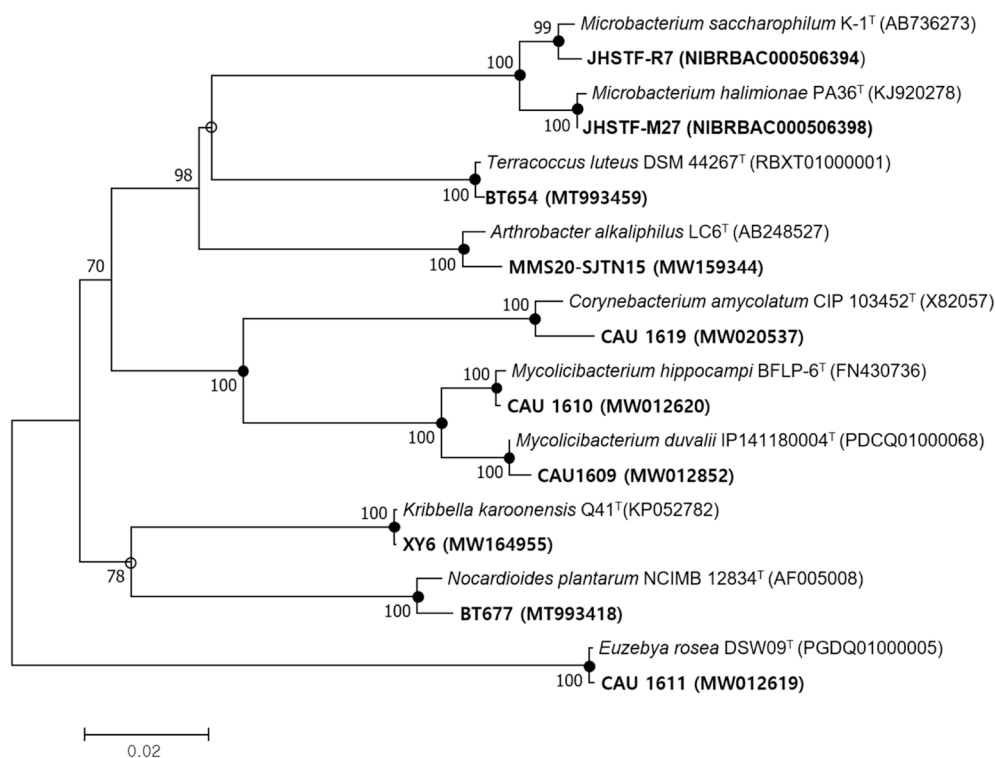


Fig. 3. Neighbor-joining phylogenetic tree based on 16S rRNA gene sequences showing the relationships between the strains isolated in this study and their relatives of the families *Nocardioideaceae*, *Mycobacteriaceae*, *Micrococcaceae*, *Microbacteriaceae*, *Intrasporangiaceae*, *Euzebyaceae* and *Corynebacteriaceae* in the phylum Actinobacteria. Bootstrap values (> 50%) are shown at branching points. Filled circles indicate the nodes also recovered in the maximum-likelihood and maximum-parsimony trees, and open circles indicate the nodes also recovered in only one of the trees. Bar, 0.02 substitutions per nucleotide position.

Description of *Kribbella karoonensis* XY6

Cells are Gram-stain-positive and filamentous. Colonies are with irregular edges and white colored after incubation for 3 days at 30°C. Positive for nitrate reduction, esculin hydrolysis and β -galactosidase; but negative for indole production, glucose fermentation, arginine dihydrolase, urease, gelatin hydrolysis and cytochrome oxidase. D-Glucose, D-mannose, D-mannitol, *N*-acetyl-glucosamine, D-maltose, potassium gluconate, adipic acid and malic acid are utilized as sole carbon sources; but not L-arabinose, capric acid, trisodium citrate and phenylacetic acid. Strain XY6 (= NIBRBAC000506149) was isolated from a wetland sample from Seoul. The GenBank accession number for the 16S rRNA gene sequence of strain XY6 is MW164955.

Description of *Nocardioides plantarum* BT677

Cells are Gram-stain-positive and rod-shaped. Colonies are circular, convex and light yellow colored after incubation for 3 days on R2A at 25°C. Positive for cytochrome oxidase, and weakly positive for urease, esculin hydrolysis and gelatin hydrolysis; but negative for nitrate reduction, indole production, glucose fermentation, arginine dihydro-

lase and β -galactosidase. D-Glucose and potassium gluconate are utilized as sole carbon sources; but not L-arabinose, D-mannose, D-mannitol, *N*-acetyl-glucosamine, D-maltose, capric acid, adipic acid, malic acid, trisodium citrate and phenylacetic acid. Strain BT677 (= NIBRBAC000506195) was isolated from a soil sample from Jeongseon-gun, Gangwon-do. The GenBank accession number for the 16S rRNA gene sequence of strain BT677 is MT993418.

Description of *Streptomyces albidoflavus* MA30

Cells are Gram-stain-positive and filamentous. Colonies are filamentous, umbonate and pale yellow colored after incubation for 3 days at 30°C. Positive for nitrate reduction and esculin hydrolysis; but negative for indole production, glucose fermentation, arginine dihydrolase, urease, gelatin hydrolysis, β -galactosidase and cytochrome oxidase. D-Glucose, L-arabinose, D-mannose, D-mannitol, potassium gluconate, adipic acid, malic acid, trisodium citrate and phenylacetic acid are utilized as sole carbon sources; but not *N*-acetyl-glucosamine, D-maltose and capric acid. Strain MA30 (= NIBRBAC000506144) was isolated from a wetland sample from Seoul. The Gen

Bank accession number for the 16S rRNA gene sequence of strain MA30 is MW164950.

Description of *Streptomyces avermitilis* MMS20-HV4-22

Cells are Gram-stain-positive and filamentous. Colonies are filamentous, raised, undulate and orange or brown colored after incubation for 3 days on ISP2 at 30°C. Positive for nitrate reduction, arginine dihydrolase, urease, esculin hydrolysis, gelatin hydrolysis and β -galactosidase; but negative for indole production, glucose fermentation and cytochrome oxidase. D-Glucose and *N*-acetyl-glucosamine are utilized as sole carbon sources; but not L-arabinose, D-mannose, D-mannitol, D-maltose, potassium gluconate, capric acid, adipic acid, malic acid, trisodium citrate and phenylacetic acid. Strain MMS20-HV4-22 (= NIBRBAC000506172) was isolated from a soil sample from Okcheon-gun, Chungcheongbuk-do. The GenBank accession number for the 16S rRNA gene sequence of strain MMS20-HV4-22 is MW159351.

Description of *Streptomyces cinereospinus* MA2

Cells are Gram-stain-positive and filamentous. Colonies are filamentous, umbonate and white colored after incubation for 3 days at 30°C. Positive for nitrate reduction and esculin hydrolysis; but negative for indole production, glucose fermentation, arginine dihydrolase, urease, gelatin hydrolysis, β -galactosidase and cytochrome oxidase. D-Glucose, D-mannose, *N*-acetyl-glucosamine, potassium gluconate and adipic acid are utilized as sole carbon sources; but not L-arabinose, D-mannitol, D-maltose, capric acid, malic acid, trisodium citrate and phenylacetic acid. Strain MA2 (= NIBRBAC000506141) was isolated from a wetland sample from Seoul. The GenBank accession number for the 16S rRNA gene sequence of strain MA2 is MW164947.

Description of *Streptomyces dangxiongensis* MMS20-SJTR5

Cells are Gram-stain-positive and filamentous. Colonies are circular, umbonate, erose and white colored after incubation for 3 days on R2A at 30°C. Positive for arginine dihydrolase, urease, gelatin hydrolysis, β -galactosidase and cytochrome oxidase; weakly positive for esculin hydrolysis; but negative for nitrate reduction, indole production and glucose fermentation. D-Glucose, L-arabinose, D-mannose, D-mannitol, *N*-acetyl-glucosamine, D-maltose, potassium gluconate, adipic acid, malic acid, trisodium citrate, phenylacetic acid are utilized as sole carbon sources; but not capric acid. Strain MMS20-SJTR5 (= NIBRBAC000506162) was isolated from a soil sample from Sejong-si. The GenBank accession number for

the 16S rRNA gene sequence of strain MMS20-SJTR5 is MW159342.

Description of *Streptomyces longisporus* MMS20-SJTR12

Cells are Gram-stain-positive and filamentous. Colonies are circular, umbonate, erose and white colored after incubation for 3 days on R2A at 30°C. Positive for arginine dihydrolase, urease, esculin hydrolysis, gelatin hydrolysis, β -galactosidase and cytochrome oxidase; but negative for nitrate reduction, indole production and glucose fermentation. D-Glucose, L-arabinose, D-mannose, D-mannitol, *N*-acetyl-glucosamine, D-maltose, potassium gluconate, adipic acid and malic acid are utilized as sole carbon sources; but not capric acid, trisodium citrate and phenylacetic acid. Strain MMS20-SJTR12 (= NIBRBAC000506163) was isolated from a soil sample from Sejong-si. The GenBank accession number for the 16S rRNA gene sequence of strain MMS20-SJTR12 is MW159343.

Description of *Streptomyces phaeofaciens* SM6

Cells are Gram-stain-positive and filamentous. Colonies are filamentous, umbonate and cream colored after incubation for 3 days at 30°C. Positive for nitrate reduction; but negative for indole production, glucose fermentation, arginine dihydrolase, urease, esculin hydrolysis, gelatin hydrolysis β -galactosidase and cytochrome oxidase. D-Glucose, L-arabinose, D-mannose, D-mannitol, *N*-acetyl-glucosamine, D-maltose, potassium gluconate, adipic acid and malic acid are utilized as sole carbon sources; but not capric acid, trisodium citrate and phenylacetic acid. Strain SM6 (= NIBRBAC000506150) was isolated from a wetland sample from Seoul. The GenBank accession number for the 16S rRNA gene sequence of strain SM6 is MW164956.

Description of *Streptomyces pseudogriseolus* MMS20-AI2-20

Cells are Gram-stain-positive and filamentous. Colonies are filamentous, raised, undulate and brown colored after incubation for 3 days on ISP2 at 30°C. Positive for nitrate reduction, arginine dihydrolase, urease, esculin hydrolysis and cytochrome oxidase; but negative for indole production, glucose fermentation, gelatin hydrolysis and β -galactosidase. D-Glucose, *N*-acetyl-glucosamine, D-maltose, potassium gluconate, capric acid and malic acid are utilized as sole carbon sources; but not L-arabinose, D-mannose, D-mannitol, adipic acid, trisodium citrate and phenylacetic acid. Strain MMS20-AI2-20 (= NIBRBAC000506174) was isolated from a soil sample from Okcheon-gun, Chungcheongbuk-do. The GenBank acces-

sion number for the 16S rRNA gene sequence of strain MMS20-AI2-20 is MW149078.

Description of *Streptomyces puniceus* I2-3

Cells are Gram-stain-positive and filamentous. Colonies are filamentous, umbonate and purple to white colored after incubation for 3 days at 30°C. Positive for nitrate reduction, urease, esculin hydrolysis and β -galactosidase; but negative for indole production, glucose fermentation, arginine dihydrolase and gelatin hydrolysis. D-Glucose, D-mannose, D-mannitol, *N*-acetyl-glucosamine, potassium gluconate and malic acid are utilized as sole carbon sources; but not L-arabinose, D-maltose, capric acid, adipic acid, trisodium citrate and phenylacetic acid. Strain I2-3 (=NIBRBAC000506146) was isolated from a wetland sample from Seoul. The GenBank accession number for the 16S rRNA gene sequence of strain I2-3 is MW164952.

Description of *Streptomyces scabiei* DM17

Cells are Gram-stain-positive and filamentous. Colonies are filamentous, umbonate and white colored after incubation for 3 days at 30°C. Positive for nitrate reduction, esculin hydrolysis and β -galactosidase; but negative for indole production, glucose fermentation, arginine dihydrolase, urease, gelatin hydrolysis and cytochrome oxidase. D-Glucose, L-arabinose, D-mannose, *N*-acetyl-glucosamine, D-maltose, potassium gluconate, adipic acid and malic acid are utilized as sole carbon sources; but not D-mannitol, capric acid, trisodium citrate and phenylacetic acid. Strain DM17 (=NIBRBAC000506140) was isolated from a wetland sample from Seoul. The GenBank accession number for the 16S rRNA gene sequence of strain DM17 is MW164946.

Description of *Streptomyces spinoverrucosus* MMS20-HV2-26

Cells are Gram-stain-positive and filamentous. Colonies are filamentous, raised, undulate and white colored after incubation for 3 days on ISP2 at 30°C. Positive for nitrate reduction, glucose fermentation, urease, esculin hydrolysis, gelatin hydrolysis, β -galactosidase and cytochrome oxidase; but negative for indole production and arginine dihydrolase. D-Glucose, L-arabinose, D-mannose, D-mannitol, *N*-acetyl-glucosamine, D-maltose, potassium gluconate, malic acid and trisodium citrate are utilized as sole carbon sources; but not capric acid, adipic acid and phenylacetic acid. Strain MMS20-HV2-26 (=NIBRBAC000506170) was isolated from a soil sample from Okcheon-gun, Chungcheongbuk-do. The GenBank accession number for the 16S rRNA gene sequence of strain MMS20-HV2-26 is MW159348.

Description of *Yinghuangia aomiensis* SO314

Cells are Gram-stain-positive and filamentous. Colonies are circular, umbonate, opaque and white colored after incubation for 5 days on R2A at 25°C. Positive for glucose fermentation, esculin hydrolysis, gelatin hydrolysis and cytochrome oxidase; but negative for nitrate reduction, indole production, arginine dihydrolase, urease and β -galactosidase. D-Glucose, L-arabinose, D-mannose, D-mannitol, *N*-acetyl-glucosamine, D-maltose, potassium gluconate, capric acid, adipic acid, malic acid, trisodium citrate and phenylacetic acid are not utilized as sole carbon sources. Strain SO314 (=NIBRBAC000506232) was isolated from a soil sample from Suncheon-si, Jeollanam-do.

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REFERENCES

- Amin, D.H., N.A. Abdallah, A. Abolmaaty, S. Tolba and E.M. Wellington. 2020. Microbiological and molecular insights on rare Actinobacteria harboring bioactive prospective. *Bull. Natl. Res. Cent.* 44:1-12.
- Felsenstein, J. 1985. Confidence limits on phylogenies: an approach using the bootstrap. *Evolution* 39:783-791.
- Goodfellow, M., P. Kämpfer, H.J. Busse, M.E. Trujillo, K.I. Suzuki, W. Ludwig and W.B. Whitman. 2012. *Bergey's Manual of Systematic Bacteriology*, 2nd edition, vol. 5, The Actinobacteria. Springer, New York.
- Jeon, Y.S., K. Lee, S.C. Park, B.S. Kim, Y.J. Cho, S.M. Ha and J. Chun. 2014. EzEditor: a versatile sequence alignment editor for both rRNA- and protein-coding genes. *Int. J. Syst. Evol. Microbiol.* 64:689-691.
- Jukes, T. and C. Cantor. 1969. Evolution of protein molecules. In: H.N. Munro (ed.), *Mammalian Protein Metabolism*, Academic Press, New York. pp. 21-132.
- Kim, M.S., S.B. Kim, C.J. Cha, W.T. Im, W.Y. Kim, M.K. Kim, H. Yi, H.R. Kim and C.N. Seong. 2021. Description of unrecorded bacterial species belonging to the phylum Actinobacteria in Korea. *J. Species Res.* 10:23-45.
- Kim, O.S., Y.J. Cho, K. Lee, S.H. Yoon, M. Kim, H. Na, S.C. Park, Y.S. Jeon, J.H. Lee, H. Yi, S. Won and J. Chun. 2012. Introducing EzTaxon-e: a prokaryotic 16S rRNA gene sequence database with phylotypes that represent uncultured species. *Int. J. Syst. Evol. Microbiol.* 62:716-721.

Kumar, S., G. Stecher and K. Tamura. 2016. MEGA7: Molecular Evolutionary Genetics Analysis version 7.0 for bigger datasets. *Mol. Biol. Evol.* 33:1870-1874.

Naikpatil, S.V. and J.L. Rathod. 2011. Selective isolation and antimicrobial activity of rare actinomycetes from mangrove sediment of Karwar. *J. Ecobiotechnol.* 3:48-53.

Saitou, N. and M. Nei. 1987. The neighbor-joining method: a new method for reconstructing phylogenetic trees. *Mol. Biol. Evol.* 4:406-425.

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