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Report on 31 unrecorded bacterial species in Korea that belong to the phylum *Actinobacteria*

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To discover and characterize indigenous species in Korea, a total of 31 bacterial strains that belong to the phylum *Actinobacteria* were isolated from various niches in Korea. Each strain showed the high sequence similarity (>99.1%) with the closest bacterial species, forming a robust phylogenetic clade. These strains have not been previously recorded in Korea. According to the recently updated taxonomy of the phylum *Actinobacteria* based upon 16S rRNA trees, we report 25 genera of 13 families within 5 orders of the class *Actinobacteria* as actinobacterial species found in Korea. Cellular morphology, Gram staining, basic biochemical characteristics are described in the species description.

Keywords: 16S rRNA gene, *Actinobacteria*, bacterial diversity, unrecorded species

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Introduction

Microorganisms are the most diverse and abundant groups of organisms on Earth, and they play important roles in various biogeochemical processes (Whitman *et al*., 1998; Fierer and Jackson, 2006). However, it was proposed that less than only 1% of total microbial cells were cultured on the planet (Amann *et al*., 1995). The diversity of uncultured microorganisms can offer the possibility of a vast number of novel microbial taxa to be still discovered. Thanks to innovative cultivation methods, increasing numbers of new names of prokaryotic species have been validly published (Oren and Garrity, 2014), and approximately 11,500 prokaryotic species have been validly published so far (Parte, 2014), although most of prokaryotic species have yet to be uncultured.

An increasing attention has been paid on the investigation of new or unrecorded prokaryotic species that are indigenous in Korea. A variety of novel bacterial species and unrecorded bacterial species were isolated from various environmental samples collected in Korea. As a result, the bacterial isolates were assigned to the phyla *Actinobacteria*, *Bacteroidetes*, *Deinococci*, *Firmicutes*, *Proteobacteria* and *Verrucomicrobia*.

The phylum *Actinobacteria* is comprised mainly of Gram-positive bacteria with a high G+C content (>55 mol% in genomic DNA), and constitutes one of the largest phyla within the Bacteria (Gao and Gupta, 2012). This phylum that contains >300 genera (Zhi *et al*., 2009) display enormous diversity in terms of their

morphology, physiology, and metabolic capabilities. The morphologies of actinobacterial species vary from coccoid to branched mycelia. Members of *Actinobacteria* are found in a wide range of environments such as soil, water, deep-sea, arctic ice, chemically contaminated sites, radioactive environments, gastrointestinal tracts of humans and animals, and plants (Gao and Gupta, 2012). Recently, the taxonomy of the phylum *Actinobacteria* based upon 16S rRNA trees was updated (Ludwig and Klenk, 2005.), which is the basis of the section on *Actinobacteria* in the Bergey's Manual of Systematic Bacteriology. Previously, the phylum *Actinobacteria* consisted of one class, 5 subclasses, and 9 orders. In the revised taxonomy, subclasses and suborders were eliminated, and elevated to the ranks of classes and orders, respectively; the phylum *Actinobacteria* is now divided into 6 classes and 22 orders. The largest class *Actinobacteria*, which accounts for >80% of all known actinobacterial families/genera, now contains a total of 15 orders, including both previously proposed orders *Actinomycetales* and *Bifidobacteriales* (Zhi *et al*., 2009). However, the order *Actinomycetales* is now restricted to the members of the family *Actinomycetaceae*, and the other suborders that were previously part of this order are now designated as distinct orders.

The species of this group also display tremendous physiological diversity, as revealed by their production of numerous extracellular enzymes (Chater *et al*., 2010) and secondary metabolites, many of which are antibiotics (Hopwood, 2007). The most extensively studied representatives of this group include soil-dwelling *Streptomyces* spp., which are the major producers of antibiotics (Chater, 2006; Ventura *et al*., 2007); the genus *Mycobacterium*, which are important human pathogens and responsible for the largest number of human deaths from bacterial infections; and the genus *Rhodococcus*, which possesses a high potential for industrial and environmental applications (Martínková *et al*., 2009; Yam *et al*., 2011).

As a part of results obtained from the research program supported by NIBR, the present report focuses on the description of bacterial species belonging to the *Actinobacteria* which have not been previously isolated in Korea. Here, we report 31 unrecorded bacterial species that belong to the phylum *Actinobacteria*.

Materials and Methods

A total of 31 bacterial strains were isolated from various environmental samples collected from soil, tidal flat, freshwater, seawater, wetland, and plant roots using various culture media (Table 1). The strains were isolated from each environmental sample by using the standard dilution plating technique on various culture media including R2A (BD), marine agar (MA; BD), nutrient agar (NA; BD), 1/10 PCA (BD), and minimal medium (MM; 30 g D-glucose, 1 g yeast extract, 0.5 g K_2HPO_4, 0.5 g KH_2PO_4, 2.2 g $(NH_4)_2SO_4$, 0.2 g $MgSO_4{\cdot}7H_2O$, 0.01 g $MnSO_4{\cdot}7H_2O$, 0.01 g $FeSO_4{\cdot}7H_2O$ and 0.01g NaCl in 1 L of water). The agar plates were incubated at 25-37°C for 2-10 days (Table 1). All isolates were sub-cultured, purified and stored as a stock culture in the same medium supplemented with 10-30% (v/v) glycerol. The designated strain IDs, sources, culture media, and incubation conditions are presented in Table 1.

Genomic DNA extraction, PCR amplification and 16S rRNA gene sequencing were carried out using standard procedures; the 16S rRNA gene of the isolates was amplified using the universal bacterial primer pair (27F and 1492R). The 16S rRNA gene sequences were compared with sequences in the ExTaxon-e server (http://eztaxon-e.ezbiocloud.net/) (Kim *et al*., 2012) and aligned with closely related species by using the CLUSTAL X program (Thompson *et al*., 1997). The phylogenetic relationships were evaluated by using neighbor-joining (Saitou and Nei, 1987) and maximum-likelihood (Felsen-stein, 1981) algorithms, and the trees were constructed using the Mega 6 program (Tamura *et al*., 2013). Evolutionary distances were calculated by the model of Jukes and Cantor (1969) and tree topologies were evaluated based on bootstrap analyses of 1,000 data sets.

Colony morphology was observed on agar plates after cells grew up to stationary phase, and cellular morphology and cell size were determined by either transmission electron microscopy or scanning electron microscopy using cells grown on various agar plates for 2-10 days. The Gram staining was carried out using the Gram-staining kit (Sigma-Aldrich), and enzyme activities and utilization of different carbon sources were assessed using API 20NE galleries (bioMérieux) according to the manufacturer's instructions.

Results and Discussion

Assignment of unrecorded strains to the validly published species of *Actinobacteria*

On the basis of 16S rRNA gene sequence comparisons and phylogenetic analyses, a total of 31 strains, which have not been previously isolated in Korea, were assigned to the phylum *Actinobacteria*. All of the 31 strains belonged to the class *Actinobacteria* and were distributed to 13 families of 5 orders; 8 strains of 7 genera in the family *Microbacteriaceae*, 4 strains of the genus *Rhodococcus* in the family *Nocardiaceae*, 4 strains of 3 genera in the family *Micrococcaceae*, 3 strains of 3 genera in the family *Intrasporangiaceae*, 3 strains of the

Table 1. List of strains belonging to the phylum *Actinobacteria* and their taxonomic assignment based on 16S rRNA gene analysis.

Class	Order	Family	Genus	Strain ID	NIBR ID	Most closely related species	Similarity (%)	Isolation source	Medium	Incubation conditions
Actinobacteria	*Corynebacteriales*	*Dietziaceae*	*Dietzia*	KYW853	NIBRBA0000114113	*Dietzia cinnamea*	99.9	Sea water	MA	25°C, 2d
		Mycobacteriaceae	*Mycobacterium*	SPE2-2	NIBRBA0000113880	*Mycobacterium frederiksbergense*	99.4	Plant root	R2A	30°C, 2d
		Nocardiaceae	*Rhodococcus*	UKS-28	NIBRBA0000113883	*Rhodococcus equi*	99.9	Wetland	R2A	25°C, 2d
			Rhodococcus	IK56	NIBRBA0000113918	*Rhodococcus canchipurensis*	99.2	Fresh water	1/10 PCA	25°C, 10d
			Rhodococcus	MK5-14	NIBRBA0000113950	*Rhodococcus jostii*	99.7	Soil	MA	30°C, 2d
			Rhodococcus	RS5-4	NIBRBA0000113951	*Rhodococcus maanshanensis*	99.3	Soil	R2A	30°C, 2d
		Tsukamurellaceae	*Tsukamurella*	mNW17	NIBRBA0000113963	*Tsukamurella tyrosinosolvens*	99.8	Soil	MM	25°C, 3d
	Micrococcales	*Cellulomonadaceae*	*Cellulomonas*	UKS-33	NIBRBA0000113882	*Cellulomonas xylanilytica*	99.6	Wetland	R2A	25°C, 2d
		Dermabacteraceae	*Brachybacterium*	HME8794	NIBRBA0000114094	*Brachybacterium muris*	99.6	Tidal flat	MA	30°C, 2d
		Intrasporangiaceae	*Humibacillus*	mNW13	NIBRBA0000113978	*Humibacillus xanthopallidus*	99.3	Soil	MM	25°C, 5d
			Serinicoccus	HWR24	NIBRBA0000113995	*Serinicoccus profundi*	99.9	Sea water	R2A	25°C, 3d
			Janibacter	AB7	NIBRBA0000114044	*Janibacter anophelis*	100	Tidal flat	MA	25°C, 2d
		Microbacteriaceae	*Plantibacter*	SPE-06	NIBRBA0000113879	*Plantibacter flavus*	99.4	Plant root	R2A	30°C, 2d
			Curtobacterium	MIC10	NIBRBA0000113848	*Curtobacterium citreum*	99.9	Fresh water	R2A	25°C, 2d
			Microbacterium	MS5-22	NIBRBA0000113944	*Microbacterium esteraromaticum*	99.3	Soil	MA	30°C, 2d
			Leifsonia	MA9	NIBRBA0000113962	*Leifsonia soli*	99.3	Soil	MA	25°C, 2d
			Microbacterium	MAT14	NIBRBA0000113972	*Microbacterium pumilum*	99.9	Soil	MA	25°C, 2d
			Microbacterium	WR-M1Y	NIBRBA0000113997	*Microbacterium oxydans*	99.7	Soil	MA	25°C, 3d
			Agrococcus	WT-RY7	NIBRBA0000114009	*Agrococcus baldri*	99.4	Plant	R2A	25°C, 3d
			Aquiluna	HME8543	NIBRBA0000114076	*Aquiluna rubra*	99.4	Fresh water	R2A	37°C, 2d
		Micrococcaceae	*Arthrobacter*	RK 4Y 5-1	NIBRBA0000113945	*Arthrobacter globiformis*	99.7	Soil	R2A	30°C, 2d
			Arthrobacter	mNW18	NIBRBA0000113977	*Arthrobacter scleromae*	99.1	Soil	MM	25°C, 3d

Table 1. Continued.

Class	Order	Family	Genus	Strain ID	NIBR ID	Most closely related species	Similarity (%)	Isolation source	Medium	Incubation conditions
Actinobacteria	*Micrococcales*	*Micrococcaceae*	*Kocuria*	HR39	NIBRBA0000113993	*Kocuria rosea*	99.6	Tidal flat	R2A	25°C, 3d
			Micrococcus	HME8781	NIBRBA0000114092	*Micrococcus flavus*	99.1	Tidal flat	MA	30°C, 2d
		Promicromonosporaceae	*Isoptericola*	RS5-5_B	NIBRBA0000113943	*Isoptericola nanjingensis*	100	Soil	R2A	30°C, 2d
			Cellulosimicrobium	NS3-4_B	NIBRBA0000113954	*Cellulosimicrobium funkei*	99.9	Soil	NA	30°C, 2d
	Propionibacteriales	*Propionibacteriaceae*	*Microlunatus*	Gsoil 950	NIBRBA0000113889	*Microlunatus soli*	99.2	Soil	R2A	25°C, 2d
	Streptomycetales	*Streptomycetaceae*	*Streptomyces*	UKS-24	NIBRBA0000113885	*Streptomyces avellaneus*	100	Wetland	R2A	25°C, 2d
			Streptomyces	MS5-13	NIBRBA0000113953	*Streptomyces omiyaensis*	100	Soil	MA	30°C, 2d
			Streptomyces	MA10	NIBRBA0000113970	*Streptomyces atratus*	100	Soil	MA	25°C, 2d
	Streptosporangiales	*Streptosporangiaceae*	*Microbispora*	UKS-23	NIBRBA0000113886	*Microbispora rosea* subsp. *rosea*	99.6	Wetland	R2A	25°C, 2d

genus *Streptomyces* in the family *Streptomycetaceae*, 2 strains of 2 genera in the family *Promicromonosporaceae*, and 7 strains of 7 genera in the respective families *Dietziaceae*, *Mycobacteriaceae*, *Tsukamurellaceae*, *Cellulomonadaceae*, *Dermabacteraceae*, *Propionibacteriaceae* and *Streptosporangiaceae* (Table 1). These strains were Gram-staining-positive, chemoheterotrophic, and rod-shaped, except for 6 strains that were Gram-staining-negative and 5 strains that were coccoid-shaped (Fig. 1). Colony size, morphology, and physiological characteristics are also shown in the species description.

Comparative sequence analyses of these strains revealed high 16S rRNA gene sequence similarities (> 99.1%) with the closest relatives (Table 1). The phylogenetic analyses showed that each strain formed a robust phylogenetic clade with the most closely related species. Eight strains belonging to the family *Microbacteriaceae* were phylogenetically assigned to *Plantibacter flavus* (Behrendt *et al*., 2002), *Curtobacterium citreum* (Yamada and Komagata, 1972), *Microbacterium esteraromaticum* (Takeuchi and Hatano, 1998), *Leifsonia soli* (Madhaiyan *et al*., 2010), *Microbacterium pumilum* (Kageyama *et al*., 2006), *Microbacterium oxydans* (Schumann *et al*., 1999), *Agrococcus baldri* (Zlamala *et al*., 2002) and *Aquiluna rubra* (Hahn, 2009), respectively. Four strains of the family *Micrococcaceae* were assigned to *Arthrobacter globiformis* (Conn and Dimmick, 1947), *Arthrobacter scleromae* (Ying Huang *et al*., 2005), *Kocuria rosea* (Stackebrandt *et al*., 1995) and *Micrococcus flavus* (Liu *et al*., 2007), respectively. Four strains were assigned to the genus *Rhodococcus* in the family *Nocardiaceae*; *R. equi* (Goodfellow and Alderson, 1977), *R. canchipurensis* (Nimaichand *et al*., 2013), *R. jostii* (Takeuchi *et al*., 2002) and *R. maanshanensis* (Zhang *et al*., 2002). Eight strains were assigned to *Humibacillus xanthopallidus* (Kageyama *et al*., 2008), *Serinicoccus profundi* (Xiao *et al*., 2011) and *Janibacter anophelis* (Peter Kämpfer *et al*., 2006) of the family *Intrasporangiaceae*, *Isoptericola nanjingensis* (Zhi Huang *et al*., 2012) and *Cellulosimicrobium funkei* (Brown *et al*., 2006) of the family *Promicromonosporaceae*, and *Streptomyces avellaneus* (Baldacci and Grein, 1966), *S. omiyaensis* (Umezawa *et al*., 1950) and *S. atratus* (Shibata *et al*., 1962) of the family *Streptomycetaceae*, respectively. Seven strains were assigned to *Dietzia cinnamea* (Yassin *et al*., 2006) of the family *Dietziaceae*, *Mycobacterium frederiksbergense* (Willumsen *et al*., 2001) of the family *Mycobacteriaceae*, *Tsukamurella tyrosinosolvens* (Yassin *et al*., 1997) of the family *Tsukamurellaceae*, *Cellulomonas xylanilytica* (Rivas *et al*., 2004) of the family *Cellulomonadaceae*, *Brachybacterium muris* (Buczolits *et al*., 2003) of the family *Dermabacteraceae*, *Microlunatus soli* (P. Kämpfer *et al*., 2010) of the family *Propionibacteriaceae*, and *Microbispora rosea* subsp. *rosea* (Miyadoh *et al*., 1990)

of the family *Streptosporangiaceae*, respectively (Fig. 2)

Description of *Dietzia cinnamea* KYW853

Cells are Gram-staining-negative, non-flagellated and rod-shaped. Colonies are opaque, round, smooth, convex and orange-colored after 2 days of incubation on MA at 25°C. Negative reactions are obtained for nitrate reduction, indole production, glucose fermentation, arginine dihydrolase, urease, esculin hydrolysis, gelatinase and β-galactosidase in API 20NE. Does not utilize 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. Strain KYW853 (=NIBRBA 0000114113) was isolated from a sea water sample, Gwangyang Bay, Gwangyang, Korea.

Description of *Mycobacterium frederiksbergense* SPE2-2

Cells are Gram-staining-positive, non-flagellated and rod-shaped. Colonies are round, smooth and yellow-colored after 2 days of incubation on R2A at 30°C. Positive reactions are obtained for nitrate reduction, glucose fermentation, esculin hydrolysis and β-galactosidase in API 20NE, and negative reactions are obtained for indole production, arginine dihydrolase, urease and gelatinase. D-glucose, L-arabinose, D-mannose, D-mannitol, *N*-acetyl-glucosamine, potassium gluconate, adipic acid, malic acid, trisodium citrate and phenylacetic acid are utilized. Does not utilize D-maltose and capric acid. Strain SPE2-2 (=NIBRBA0000113880) was isolated from a plant root sample, Chungnam National University, Daejeon, Korea.

Description of *Rhodococcus equi* UKS-28

Cells are Gram-staining-positive, non-flagellated and rod-shaped. Colonies are circular, raised, entire and yellow-colored after 2 days of incubation on R2A at 25°C. Positive reaction is obtained for nitrate reduction in API 20NE, and negative reactions are obtained for indole production, glucose fermentation, arginine dihydrolase, urease, esculin hydrolysis, gelatinase and β-galactosidase. Malic acid is utilized. Does not utilize 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 UKS-28 (=NIBRBA0000113883) was isolated from a wetland sample, Ungok-ri, Gochang, Chonbuk, Korea.

Description of *Rhodococcus canchipurensis* IK56

Cells are Gram-staining-positive, non-flagellated and rod-shaped. Colonies are smooth, circular, convex, opaque and pallid pink-colored after 10 days of incubation on 1/10 PCA at 25°C. Positive reactions are obtained for nitrate reduction, esculin hydrolysis and β-galactosidase in API 20NE, and negative reactions are obtained for indole production, glucose fermentation, arginine dihydrolase, urease and gelatinase. Malic acid is utilized. Does not utilize 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 IK56 (=NIBRBA0000113918) was isolated from a freshwater sample, Ingyeong lake, Incheon, Korea.

Description of *Rhodococcus jostii* MK5-14

Cells are Gram-staining-positive, non-flagellated and rod-shaped. Colonies are circular, entire, smooth and beige colored after 2 days on MA at 30°C. Negative reactions are obtained for nitrate reduction, indole production, glucose fermentation, arginine dihydrolase, urease, esculin hydrolysis, gelatinase and β-galactosidase in API 20NE. D-glucose, L-arabinose, D-mannose, D-mannitol, *N*-acetyl-glucosamine, potassium gluconate, adipic acid, malic acid, trisodium citrate and phenylacetic acid are utilized. Does not utilize D-maltose and capric acid. Strain MK5-14 (=NIBRBA0000113950) was isolated from a sample of ginseng field, Iljuk, Anseong, Korea.

Description of *Rhodococcus maanshanensis* RS5-4

Cells are Gram-staining-positive, non-flagellated, non-pigmented and rod-shaped. Colonies are circular, entire, smooth and white-colored after 2 days on R2A at 30°C. Positive reactions are obtained for nitrate reduction, urease, esculin hydrolysis, and β-galactosidase in API 20NE and negative reactions are obtained for indole production, glucose fermentation, arginine dihydrolase and gelatinase. D-glucose, L-arabinose, D-mannose, D-mannitol, *N*-acetyl-glucosamine, D-maltose, potassium gluconate, adipic acid, malic acid and trisodium citrate are utilized. Does not utilize capric acid and phenylacetic acid. Strain RS5-4 (=NIBRBA0000113951) was isolated from a sample of ginseng field, Iljuk, Anseong, Korea.

Description of *Tsukamurella tyrosinosolvens* mNW17

Cells are Gram-staining-positive, non-flagellated and rod-shaped. Colonies are circular and beige-colored after 3 days on minimal medium at 25°C. Positive reactions are obtained for glucose fermentation, urease, esculin hydrolysis, gelatinase and β-galactosidase in API 20NE, and negative reactions are obtained for nitrate reduction, indole production and arginine dihydrolase. L-arabinose,

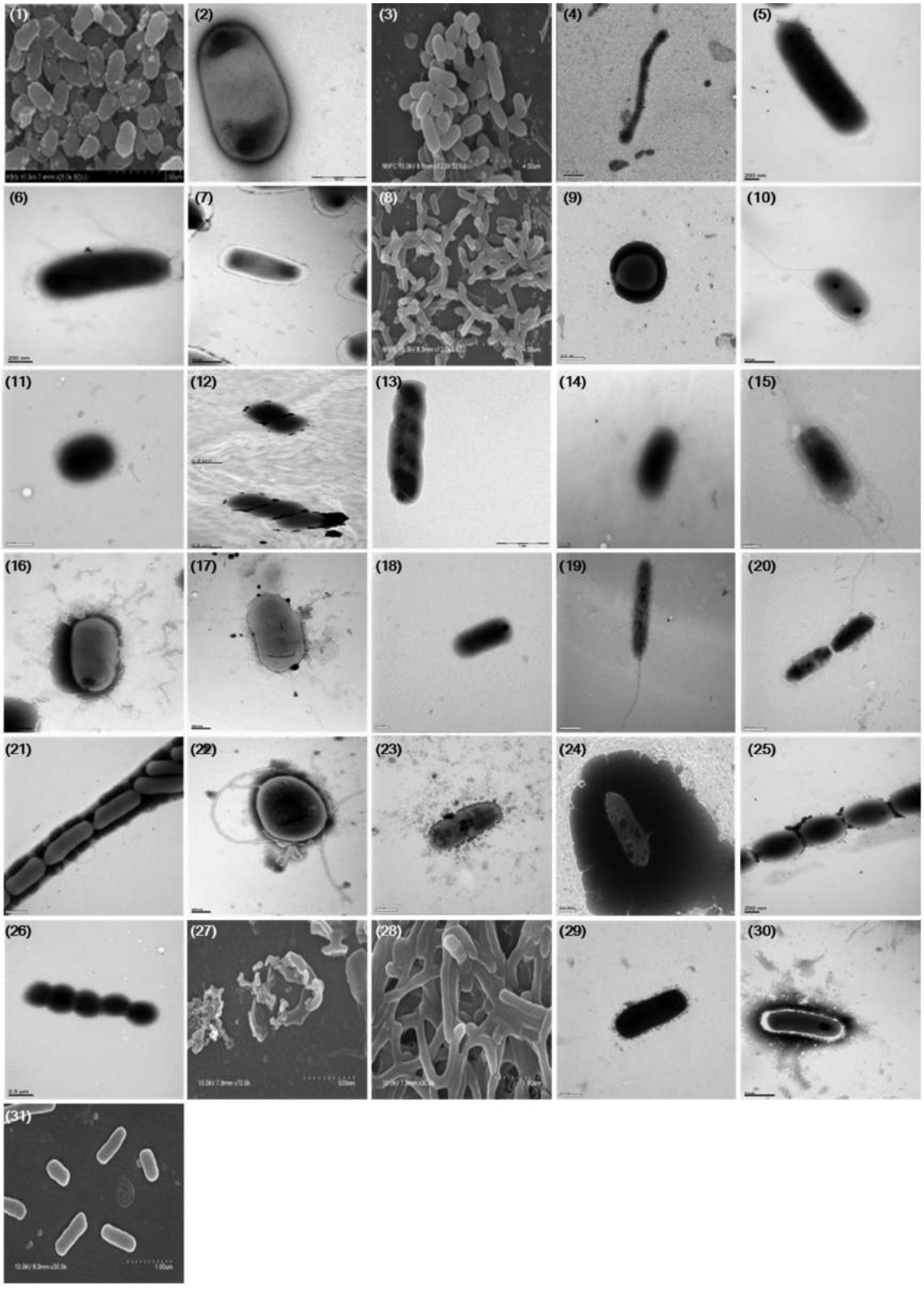

Fig. 1. Transmission electron micrographs or scanning electron micrographs of cells of the strains isolated in this study. Strains: 1, KYW853; 2, SPE2-2; 3, UKS-28; 4, IK56; 5, MK5-14; 6, RS5-4; 7, mNW17; 8, UKS-33; 9, HME8794; 10, mNW13; 11, HWR24; 12, AB7; 13, SPE-06; 14, MIC10; 15, MS5-22; 16, MA9; 17, MAT14; 18, WR-M1Y; 19, WT-RY7; 20, HME8543; 21, RK 4Y 5-1; 22, mNW18; 23, HR39; 24, HME8781; 25, RS5-5_B; 26, NS3-4_B; 27, Gsoil 950; 28, UKS-24; 29, MS5-13; 30, MA10; 31, UKS-23.

Fig. 2. Neighbor-joining phylogenetic tree based on 16S rRNA gene sequences, showing the relationship between the strains isolated in this study and their relatives of the phylum *Actinobacteria*. The numbers at nodes represent bootstrap values (>50%) obtained by neighbor-joining and maximum-likelihood methods, respectively. Closed circles indicate the nodes recovered by maximum-likelihood algorithm. The GenBank accession number of each species is enclosed in parentheses. Bar, 0.02 substitutions per nucleotide position.

D-mannitol, *N*-acetyl-glucosamine, D-maltose, potassium gluconate, trisodium citrate and phenylacetic acid are utilized. Does not utilize D-glucose, D-mannose, capric acid, adipic acid and malic acid. Strain mNW17 (=NIBRBA0000113963) was isolated from a forest soil sample, Gwanak Mountain, Seoul, Korea.

Description of *Cellulomonas xylanilytica* UKS-33

Cells are Gram-staining-positive, non-flagellated and rod-shaped. Colonies are circular, raised, entire and yellow-colored after 2 days on R2A at 25°C. Positive reactions are obtained for esculin hydrolysis and β-galactosidase in API 20NE, and negative reactions are obtained for nitrate reduction, indole production, glucose fermentation, arginine dihydrolase, urease and gelatinase. Does not utilize 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. Strain UKS-33 (=NIBRBA0000113882) was isolated from a wetland sample, Ungok-ri, Gochang, Korea.

Description of *Brachybacterium muris* HME8794

Cells are Gram-staining-positive, non-flagellated and rod-shaped. Colonies are circular, convex, entire and yellow-colored after 2 days on MA at 30°C. Positive reactions are obtained for esculin hydrolysis and β-galactosidase in API 20NE, and negative reactions are obtained for nitrate reduction, indole production, glucose fermentation, arginine dihydrolase, urease and gelatinase. D-glucose, D-mannose, D-maltose and potassium gluconate are utilized. Does not utilize L-arabinose, D-mannitol, *N*-acetyl-glucosamine, capric acid, adipic acid, malic acid, trisodium citrate and phenylacetic acid. Strain HME8794 (=NIBRBA0000114003) was isolated from a tidal flat sample, Sinan, Korea.

Description of *Humibacillus xanthopallidu* mNW13

Cells are flagellated and rod-shaped. Colonies are circular and orange-colored after 5 days on minimal medium at 25°C. Positive reactions are obtained for glucose fermentation, urease, esculin hydrolysis and β-galactosidase in API 20NE, and negative reactions are obtained for nitrate reduction, indole production, arginine dihydrolase and gelatinase. D-mannose, D-mannitol, D-maltose and malic acid are utilized. Does not utilize D-glucose, L-arabinose, *N*-acetyl-glucosamine, potassium gluconate, capric acid, adipic acid, trisodium citrate and phenylacetic acid. Strain mNW13 (=NIBRBA0000113978) was isolated from a forest soil sample, Gwanak Mountain, Seoul, Korea.

Description of *Serinicoccus profundi* HWR24

Cells are Gram-staining-positive, non-flagellated and coccoid-shaped. Colonies are circular, convex, glistening and yellow-colored after 3 days on R2A at 25°C. Positive reactions are obtained for nitrate reduction, esculin hydrolysis, gelatinase and β-galactosidase in API 20NE, and negative reactions are obtained for indole production, glucose fermentation, arginine dihydrolase and urease. D-glucose, D-mannose, D-maltose, potassium gluconate and malic acid are utilized. Does not utilize L-arabinose, D-mannitol, *N*-acetyl-glucosamine, capric acid, adipic acid, trisodium citrate and phenylacetic acid. Strain HWR24 (=NIBRBA0000113995) was isolated from a tidal flat sample, Hwango-ri, Anmyeon-eup, Taean, Korea.

Description of *Janibacter anopheles* AB7

Cells are Gram-staining-positive, non-flagellated and short-rod-shaped. Colonies are circular, raised, entire and ivory-colored after 2 days on MA at 25°C. Positive reactions are obtained for nitrate reduction, esculin hydrolysis and β-galactosidase in API 20NE and negative reactions are obtained for indole production, glucose fermentation, arginine dihydrolase, urease and gelatinase. D-glucose, L-arabinose, D-mannose, D-mannitol, *N*-acetyl-glucosamine, D-maltose, potassium gluconate, malic acid and trisodium citrate are utilized. Does not utilize capric acid, adipic acid and phenylacetic acid. Strain AB7 (=NIBRBA0000114044) was isolated from a tidal flat sample, Uihang-ri, Sowon-myeon, Taean, Korea.

Description of *Plantibacter flavus* SPE-06

Cells are Gram-staining-positive, non-flagellated and rod and spiral-shaped. Colonies are round, convex and yellow-colored after 2 days on R2A at 30°C. Positive reactions are obtained for glucose fermentation, esculin hydrolysis and β-galactosidase in API 20NE, and negative reactions are obtained for nitrate reduction, indole production, arginine dihydrolase, urease and gelatinase. D-glucose, L-arabinose, D-mannose, D-mannitol, *N*-acetyl-glucosamine, D-maltose, potassium gluconate and malic acid are utilized. Does not utilize capric acid, adipic acid, trisodium citrate and phenylacetic acid. Strain SPE-06 (=NIBRBA0000113879) was isolated from a plant root sample, Chungnam National University, Daejeon, Korea.

Description of *Curtobacterium citreum* MIC10

Cells are Gram-staining-negative, non-flagellated and rod-shaped. Colonies are circular and yellow-colored after 2 days on R2A at 25°C. Positive reactions are obtained for nitrate reduction, esculin hydrolysis, gelati-

nase and β-galactosidase in API 20NE, and negative reactions are obtained for indole production, glucose fermentation, arginine dihydrolase and urease. D-glucose, L-arabinose, D-mannose, D-mannitol, *N*-acetyl-glucosamine, D-maltose, and potassium gluconate are utilized. Does not utilize capric acid, adipic acid, malic acid, trisodium citrate and phenylacetic acid. Strain MIC10 (=NIBRBA0000113848) was isolated from a fresh water, Jeju island, Korea.

Description of *Microbacterium esteraromaticum* MS5-22

Cells are Gram-staining-negative, flagellated and rod-shaped. Colonies are circular, entire, smooth and yellow-colored after 2 days on MA at 30°C. Positive reactions are obtained for nitrate reduction and β-galactosidase in API 20NE, and negative reactions are obtained for indole production, glucose fermentation, arginine dihydrolase, urease, esculin hydrolysis and gelatinase. D-glucose, L-arabinose, D-mannose, D-mannitol, *N*-acetyl-glucosamine, D-maltose and potassium gluconate utilized. Does not utilize capric acid, adipic acid, malic acid, trisodium citrate and phenylacetic acid. Strain MS5-22 (=NIBRBA0000113944) was isolated from a sample of ginseng field, Iljuk, Anseong, Korea.

Description of *Leifsonia soli* MA9

Cells are flagellated and rod-shaped. Colonies are circular and yellow-colored after 2 days on MA at 25°C. Positive reactions are obtained for urease, esculin hydrolysis and β-galactosidase in API 20NE, and negative reactions are obtained for nitrate reduction, indole production, glucose fermentation, arginine dihydrolase and gelatinase. D-glucose and D-mannose are utilized. Does not utilize L-arabinose, D-mannitol, *N*-acetyl-glucosamine, D-maltose, potassium gluconate, capric acid, adipic acid, malic acid, trisodium citrate and phenylacetic acid. Strain MA9 (=NIBRBA0000113962) was isolated from a forest soil sample, Gwanak Mountain, Seoul, Korea.

Description of *Microbacterium pumilum* MAT14

Cells are Gram-staining-positive, flagellated and rod-shaped. Colonies are circular and orange-colored after 2 days on MA at 25°C. Positive reactions are obtained for glucose fermentation, urease, esculin hydrolysis and β-galactosidase in API 20NE, and negative reactions are obtained for nitrate reduction, indole production, arginine dihydrolase and gelatinase. Does not utilize 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. Strain MAT14 (=NIBRBA0000113972) was isolated from a forest soil sample, Gwanak Mountain, Seoul, Korea.

Description of *Microbacterium oxydans* WR-M1Y

Cells are Gram-staining-positive, non-flagellated and rod-shaped. Colonies are circular, convex, glistening and orange/yellow-colored after 3 days on MA at 25°C. Positive reactions are obtained for esculin hydrolysis, gelatinase and β-galactosidase in API 20NE, and negative reactions are obtained for nitrate reduction, indole production, glucose fermentation, arginine dihydrolase and urease. D-glucose, D-mannose, D-mannitol, D-maltose and potassium gluconate are utilized. Does not utilize L-arabinose, *N*-acetyl-glucosamine, capric acid, adipic acid, malic acid, trisodium citrate and phenylacetic acid. Strain WR-M1Y (=NIBRBA0000113997) was isolated from a soil sample in field of reeds, Wando, Korea.

Description of *Agrococcus baldri* WT-RY7

Cells are Gram-staining-positive, flagellated and rod-shaped. Colonies are circular, convex, glistening and vivid-yellow-colored after 3 days on R2A at 25°C. Positive reactions are obtained for esculin hydrolysis, gelatinase and β-galactosidase in API 20NE, and negative reactions are obtained for nitrate reduction, indole production, glucose fermentation, arginine dihydrolase and urease. Does not utilize 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. Strain WT-RY7 (=NIBRBA0000114009) was isolated from a plant sample, Wando, Korea.

Description of *Aquiluna rubra* HME8543

Cells are Gram-staining-positive, non-flagellated and rod-shaped. Colonies are circular, raised, entire and red-colored after 2 days on R2A at 37°C. Positive reaction is obtained for β-galactosidase in API 20NE, and negative reactions are obtained for nitrate reduction, indole production, glucose fermentation, arginine dihydrolase, urease, esculin hydrolysis and gelatinase. Does not utilize 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. Strain HME8543 (=NIBRBA0000114076) was isolated from a freshwater sample, Geongan Stream, Yongin, Korea.

Description of *Arthrobacter globiformis* RK 4Y 5-1

Cells are Gram-staining-negative, flagellated, non-pigmented and rod-shaped. Colonies are circular, entire,

smooth and white-colored after 2 days on R2A at 30°C. Positive reactions are obtained for gelatinase and β-galactosidase in API 20NE, and negative reactions are obtained for nitrate reduction, indole production, glucose fermentation, arginine dihydrolase, urease and esculin hydrolysis. D-glucose, L-arabinose, D-mannose, D-mannitol, *N*-acetyl-glucosamine, D-maltose, potassium gluconate, adipic acid, malic acid, trisodium citrate and phenylacetic acid are utilized. Dose not utilize capric acid. Strain RK 4Y 5-1 (=NIBRBA0000113994) was isolated from a sample of ginseng field, Iljuk, Anseong, Korea.

Description of *Arthrobacter scleromae* mNW18

Cells are Gram-staining-positive, flagellated, non-pigmented and coccoid-shaped. Colonies are circular and cream-colored after 3 days on minimal meduim at 25°C. Positive reactions are obtained for glucose fermentation, urease, esculin hydrolysis, gelatinase and β-galactosidase in API 20NE, and negative reactions are obtained for nitrate reduction, indole production and arginine dihydrolase. L-arabinose, D-mannitol, *N*-acetyl-glucosamine, D-maltose, potassium gluconate, trisodium citrate and phenylacetic acid are utilized. Does not utilize D-glucose, D-mannose, capric acid, adipic acid and malic acid. Strain mNW18 (=NIBRBA0000113977) was isolated from a forest soil sample, Gwanak Mountain, Seoul, Korea.

Description of *Kocuria rosea* HR39

Cells are Gram-staining-positive, non-flagellated and rod-shaped. Colonies are irregular, convex, glistening and pale orange/yellow-colored after 3 days on R2A at 25°C. Positive reactions are obtained for nitrate reduction and β-galactosidase in API 20NE, and negative reactions are obtained for indole production, glucose fermentation, arginine dihydrolase, urease, esculin hydrolysis and gelatinase. D-glucose, L-arabinose, D-mannitol, D-maltose, potassium gluconate, adipic acid and malic acid are utilized. Does not utilize D-mannose, *N*-acetyl-glucosamine, capric acid, trisodium citrate and phenylacetic acid. Strain HR39 (=NIBRBA0000113993) was isolated from a tidal flat sample, Hwango-ri, Anmyeon-eup, Taean, Korea.

Description of *Micrococcus flavus* HME8781

Cells are Gram-staining-positive, non-flagellated and rod-shaped. Colonies are circular, convex, entire and yellow-colored after 2 days on MA at 30°C. Negative reactions are obtained for nitrate reduction, indole production, glucose fermentation, arginine dihydrolase, urease, esculin hydrolysis, gelatinase and β-galactosidase in API 20NE. D-glucose D-maltose and malic acid are utilized. Does not utilize L-arabinose, D-mannose, D-mannitol, *N*-acetyl-glucosamine, potassium gluconate, capric acid, adipic acid, trisodium citrate and phenylacetic acid. Strain HME8781 (=NIBRBA0000114092) was isolated from a tidal flat sample, Sinan, Korea.

Description of *Isoptericola nanjingensis* RS5-5_B

Cells are Gram-staining-positive, non-flagellated and coccoid-shaped. Colonies are circular, entire, smooth and light yellow-colored after 2 days on R2A at 30°C. Positive reactions are obtained for nitrate reduction, esculin hydrolysis and β-galactosidase in API 20NE, and negative reactions are obtained for indole production, glucose fermentation, arginine dihydrolase, urease and gelatinase. D-glucose, L-arabinose, D-mannose, D-mannitol, *N*-acetyl-glucosamine, D-maltose and potassium gluconate are utilized. Does not utilize capric acid, adipic acid, malic acid, trisodium citrate and phenylacetic acid. Strain RS5-5_B (=NIBRBA0000113943) was isolated from a sample of ginseng field, Iljuk, Anseong, Korea.

Description of *Cellulosimicrobium funkei* NS3-4_B

Cells are Gram-staining-positive, non-flagellated and coccoid-shaped. Colonies are punctiform, entire, smooth and light yellow-colored after 2 days on NA at 30°C. Positive reactions are obtained for nitrate reduction, esculin hydrolysis and β-galactosidase in API 20NE, negative reactions are obtained for indole production, glucose fermentation, arginine dihydrolase, urease and gelatinase. D-glucose, L-arabinose, D-mannose, *N*-acetyl-glucosamine, D-maltose and potassium gluconate are utilized. Does not utilize D-mannitol, capric acid, adipic acid, malic acid, trisodium citrate and phenylacetic acid. Strain NS3-4_B (=NIBRBA0000113954) was isolated from a sample of ginseng field, Iljuk, Anseong, Korea.

Description of *Microlunatus soli* Gsoil 950

Cells are Gram-staining-positive, non-flagellated and rod-shaped. Colonies are circular, raised, entire and yellow-colored after 2 days on R2A at 25°C. Positive reactions are obtained for nitrate reduction, glucose fermentation, urease, esculin hydrolysis, gelatinase and β-galactosidase in API 20NE, and negative reactions are obtained for indole production and arginine dihydrolase. D-glucose, D-mannose, D-mannitol, *N*-acetyl-glucosamine, D-maltose and trisodium citrate are utilized. Does not utilize L-arabinose, potassium gluconate, capric acid, adipic acid, malic acid, and phenylacetic acid. Strain Gsoil 950 (=NIBRBA0000113889) was isolated from a sample of ginseng field, Pocheon, Korea.

Description of *Streptomyces avellaneus* UKS-24

Cells are Gram-staining-positive, non-flagellated and rod-shaped. Colonies are circle, raised, entire and yellow-colored after 2 days on R2A at 25°C Positive reaction is obtained for β-galactosidase in API 20NE, and negative reactions are obtained for nitrate reduction, indole production, glucose fermentation, arginine dihydrolase, urease, esculin hydrolysis and gelatinase. D-glucose, L-arabinose, *N*-acetyl-glucosamine, potassium gluconate and malic acid are utilized. Does not utilize D-mannose, D-mannitol, D-maltose, capric acid, adipic acid, trisodium citrate and phenylacetic acid. Strain UKS-24 (=NIBRBA0000113885) was isolated from a wetland sample, Ungok-ri, Gochang, Korea.

Description of *Streptomyces omiyaensis* MS5-13

Cells are Gram-staining-positive, non-flagellated and rod-shaped. Colonies are irregular, undulate, smooth and light yellow-colored after 2 days on MA at 30°C. Positive reactions are obtained for nitrate reduction, esculin hydrolysis and β-galactosidase in API 20NE, and negative reactions are obtained for indole production, glucose fermentation, arginine dihydrolase, urease and gelatinase. D-glucose, L-arabinose, D-mannose, D-mannitol, *N*-acetyl-glucosamine, D-maltose, potassium gluconate, adipic acid, malic acid and phenylacetic acid are utilized. Does not utilize capric acid and trisodium citrate. Strain MS5-13 (=NIBRBA0000113953) was isolated from a sample of ginseng field, Anseong, Korea.

Description of *Streptomyces atratus* MA10

Cells are Gram-staining-negative, non-flagellated and rod-shaped. Colonies are circle and yellow-colored after 2 days on MA at 25°C. Strain MA10 (=NIBRBA00 00113970) was isolated from a forest soil sample, Gwanak Mountain, Seoul, Korea.

Description of *Microbispora rosea* subsp. *rosea* UKS-23

Cells are Gram-staining-negative, non-flagellated and rod-shaped. Colonies are circular, raised, entire and yellow-colored after 2 day on R2A at 25°C. Positive reaction is obtained for esculin hydrolysis in API 20NE, and negative reactions are obtained for nitrate reduction, indole production, glucose fermentation, arginine dihydrolase, urease, gelatinase and β-galactosidase. D-glucose, L-arabinose, D-mannose, D-mannitol, *N*-acetyl-glucosamine, D-maltose, potassium gluconate, adipic acid and malic acid are utilized. Does not utilize capric acid, trisodium citrate and phenylacetic acid. Strain UKS-23 (=NIBRBA0000113886) was isolated from a wetland sample, Ungok-ri, Gochang, Korea.

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