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A report on 33 unrecorded bacterial species of Korea isolated in 2014, belonging to the class *Gammaproteobacteria*

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In 2014, as a subset study to discover indigenous prokaryotic species in Korea, a total of 33 bacterial strains assigned to the class *Gammaproteobacteria* were isolated from diverse environmental samples collected from soil, tidal flat, freshwater, seawater, oil-contaminated soil, and guts of animal. From the high 16S rRNA gene sequence similarity (>98.5%) and formation of a robust phylogenetic clade with the closest species, it was determined that each strain belonged to each independent and predefined bacterial species. There is no official report that these 33 species have been described in Korea; therefore, 1 strain of the *Aeromonadales*, 6 strains of the *Alteromonadales*, 3 strains of the *Chromatiales*, 5 strains of the *Enterobacteriales*, 4 strains of the *Oceanospirillales*, 11 strains of the *Pseudomonadales*, and 3 strains of the *Xanthomonadales* within the *Gammaproteobacteria* are described for unreported bacterial species in Korea. Gram reaction, colony and cell morphology, basic biochemical characteristics, and isolation sources are also described in the species description section.

Keywords: 16S rRNA, bacterial diversity, *Gammaproteobacteria*, unreported species

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Introduction

The class *Gammaproteobacteria* forms the largest phylogenetic group (at least 180 genera and over 750 species) in the phylum *Proteobacteia* and members of the class show diverse metabolic pathways and phenotypes (Garrity *et al*., 2005; Kersters *et al*., 2006; Williams *et al*., 2010). In general, all organisms in this class are unicellular and mostly rods, and are abundant in various freshwater habitats (Kim *et al*., 2012a; Zhang *et al*., 2014; Keller-Costa *et al*., 2014), seawater habitats (Giovannoni *et al*., 1990; Cho and Giovannoni, 2004; Du *et al*., 2009; Yan *et al*., 2011; Wang *et al*., 2012), and guts of animal (Kersters *et al*., 2006; Williams *et al*., 2010). The class *Gammaproteobacteria* contains photosynthetic purple sulfur bacteria (*Chromatiales*) together with a great number of familiar chemoheterotrophic groups, such as *Enterobacteriales*, *Legionellales*, *Pasteurellales*, *Pseudomonadales*, and also some chemolithotrophic sulfur and iron-oxidizing bacteria group (Kersters *et al*., 2006). Based on the branching pattern in the 16S rRNA gene trees, the class *Gammaproteobacteria* has been currently divided into 16 main orders (Parte, 2014): *Aeromonadales*, *Alteromonadales*, *Arenicellales*, *Cardiobacteriales*, *Chromatiales*, *Enterobacteriales*, *Legionellales*, *Methylococcales*, *Oceanospirillales*, *Orbales*, *Pasteurellales*, *Pseudomonadales*, *Salinisphaerales*, *Thiotrichales*, *Vibrionales*, *Xanthomonadales* (Garrity *et al*., 2005; Kersters *et al*., 2006; Williams *et al*., 2010).

As a part of the research program of 'The survey of Korean indigenous species', during 2014 we collected diverse environmental samples including ginseng field

soil, mud flats, freshwater, and seawaters and isolated hundreds of novel bacterial species and unreported bacterial species in Korea. Using the bacterial strains isolated during the study of 2014 research period, we have identified the strains based on 16S rRNA gene phylogeny and found that they belonged to the classes/phyla *Alphaproteobacteria*, *Betaproteobacteria*, *Gammaproteobacteria*, *Deltaproteobacteria*, *Bacteroidetes*, *Firmicutes*, *Actinobacteria*, *Deinococci*, and *Verrucomicrobia*. As a subset of this study, the present report focuses on the description of bacterial species belonging to the *Gammaproteobacteria* that have not officially reported in Korea. Here in the present study we report 33 unreported bacterial species in Korea belonging to 12 families of 7 orders in the *Gammaproteobacteria*.

Materials and Methods

All bacterial strains were isolated as pure cultures by a serial streaking onto different culture media. As a result, a total of 33 bacterial strains assigned to the class *Gammaproteobacteria* were isolated from various environmental habitats including gut of animals, ginseng soil, soil, mud flat, oil-contaminated lakes and soil, freshwater, and seawater. Each environmental sample was processed separately, spread onto diverse culture agar media including R2A agar (BD, USA), marine agar 2216 (MA; BD, USA), tryptic soy agar (TSA; BD, USA) and nutrient agar (NA; BD, USA), and incubated at 15-37°C for 1-5 days (Table 1). The strain designations, sources, culture media, and incubation conditions are summarized in Table 1. All strains were purified as single colonies and stored as 10-20% glycerol suspension at −80°C and as well as lyophilized ampoules.

Colony morphology of the strains were observed on agar plates with a magnifying glass after the cells were cultivated to their stationary phase. Cellular morphology and cell size were examined by either transmission electron microscopy or scanning electron microscopy (Fig. 1). Gram staining was performed using a Gram-staining kit or the standard procedures. Biochemical characteristics were tested by using API 20NE galleries (bioMérieux) according to the manufacturer's instructions.

Bacterial DNA extraction, PCR amplification, and 16S rRNA gene sequencing were performed using standard procedures described elsewhere. The 16S rRNA gene sequences of the strains assigned to the class *Gammaproteobacteria* were selected for subsequent analyses. The 16S rRNA gene sequences of gammaproteobacterial strains were compared with those of the validly-published type strains using the EzTaxon-e server (Kim *et al*., 2012b). The cutoff vale of 98.5% sequence similarity was applied for the identification of species. For phylogenetic analyses, multiple alignments between the 16S rRNA gene sequences of the isolates and those of the valid type strains were carried out using the ClustalW program and manually checked with the EzEditor (Jeon *et al*., 2014). Based on the sequences aligned, phylogenetic trees were generated by using neighbor-joining method (Saitou and Nei, 1987) that is programmed in MEGA 6.0 software (Tamura *et al*., 2013). The robustness of the phylogenetic trees was confirmed by bootstrap analyses based on 1,000 random replications (Felsenstein, 1985).

Results and Discussion

From the diverse habitats listed in Table 1, a total of 33 bacterial strains belonging to the class *Gammaproteobacteria* were isolated. The taxonomic composition and identification results of the isolates are summarized in Table 1. Based on 16S rRNA gene sequences and phylogenetic analyses, these 33 strains were assigned into 7 orders of the *Gammaproteobacteria*; 1 strain of the *Aeromonadales*, 6 strains of the *Alteromonadales*, 3 strains of the *Chromatiales*, 5 strains of the *Enterobacteriales*, 4 strains of the *Oceanospirillales*, 11 strains of the *Pseudomonadales*, and 3 strains of the *Xanthomonadales* (Table 1). All of these strains were Gram-staining-negative and rod-shaped or cocci-shaped bacteria (Fig. 1). Phylogenetic trees of bacterial strains assigned into the orders *Aeromonadales*, *Alteromonadales*, and *Chromatiales* (Fig. 2), *Enterobacteriales*, *Oceanospirillales*, and *Xanthomonadales* (Fig. 3), and *Pseudomonadales* (Fig. 4) are shown in Fig. 2, Fig. 3, and Fig. 4, respectively.

A single strain assigned to the family *Aeromonadaceae* of the order *Aeromonadales* was identified as being a member of *Aeromonas eucrenophila* (Fig. 2). Six strains in the order *Alteromonadales* belonged to 5 separate genera of 4 families: *Alteromonas* (1 species), *Marinobacter* (2 species), *Thalassotalea* (1 species), *Porticoccus* (1 species), and *Shewanella* (1 species). Three strains assigned to the family *Chromatiaceae* of the order *Chromatiales* belonged to the genus *Rheinheimera*. Five strains assigned to the family *Enterobacteriaceae* of the order *Enterobacteriales* belonged to the genera *Citrobacter*, *Kluyvera*, *Kosakonia*, *Kosakonia*, and *Pantoea* (Fig. 3). Four strains in the order *Oceanospirillales* belonged to 2 genera of 2 families: *Alcanivorax* (3 species) and *Kushneria* (1 species). Three strains assigned to the family *Xanthomonadaceae* belonged to the genera *Arenimonas*, *Pseudoxanthomonas*, and *Dyella* (Fig. 3). Finally, 11 strains in the order *Pseudomonadales* (Fig. 4) were found to belong to 2 genera (*Psychrobacter* and *Acinetobacter*) within the *Moraxellaceae* and the single genus *Pseudomonas* (8 strains) within the family *Pseudomonadaceae*.

Since there is no official report that these 33 species of

Table 1. Summary of strains isolated belonging to the *Gammaproteobacteria* and their taxonomic affiliations.

Order	Family	Genus	Strain ID	NIBR ID	Most closely related species (Type strain of species)	Similarity (%)	Isolation source	Medium	Incubation conditions
Aeromonadales	*Aeromonadaceae*	*Aeromonas*	MD2F 12	NIBRBA0000114772	*Aeromonas eucrenophila* (CECT 4224^T)	99.92%	Fresh water	R2A	25°C, 2 days
Alteromonadales	*Alteromonadaceae*	*Alteromonas*	HMF2613	NIBRBA0000114999	*Alteromonas australica* (H 17^T)	99.80%	Sea water	MA	37°C, 2 days
		Marinobacter	HME9674	NIBRBA0000114991	*Marinobacter lutaoensis* (JCM 11179^T)	99.30%	Sea water	MA	25°C, 3 days
		Marinobacter	IMCC12363	NIBRBA0000114859	*Marinobacter algicola* (DG893^T)	98.90%	Sea water	MA	15°C, 3 days
	Colwelliaceae	*Thalassotalea*	HME9272	NIBRBA0000114987	*Thalassotalea piscium* (T202^T)	99.00%	Water	MA	25°C 3 days
	Porticoccaceae	*Porticoccus*	IMCC12427	NIBRBA0000114867	*Porticoccus hydrocarbonoclasticus* (MCTG13d^T)	100.00%	Sea water	MA	15°C 3 days
	Shewanellaceae	*Shewanella*	61DPR3	NIBRBA0000114794	*Shewanella putrefaciens* (LMG 26268^T)	99.10%	Fresh water	R2A	25°C 2 days
Chromatiales	*Chromatiaceae*	*Rheinheimera*	HMF2735	NIBRBA0000115001	*Rheinheimera tangshanensis* (JA3-B52^T)	99.10%	Sea water	R2A	30°C 5 days
		Rheinheimera	MIH20	NIBRBA0000114775	*Rheinheimera arenilitoris* (J-MS1^T)	98.78%	Sea water	R2A	25°C 2 days
		Rheinheimera	Duck L2	NIBRBA0000114823	*Rheinheimera tangshanensis* (JA3-B52^T)	98.92%	Oil-contaminated lake	R2A	25°C 2 days
Enterobacteriales	*Enterobacteriaceae*	*Citrobacter*	WR25	NIBRBA0000114766	*Citrobacter freundii* (ATCC 8090^T)	99.43%	Fresh water	R2A	25°C 2 days
		Kluyvera	142-3	NIBRBA0000114788	*Kluyvera ascorbata* (ATCC 33433^T)	99.10%	Fresh water	TSA	25°C 2 days
		Kosakonia	HME8565	NIBRBA0000114985	*Kosakonia cowanii* (CIP 107300^T)	99.40%	Water	R2A	30°C 3 days
		Kosakonia	HMF2475	NIBRBA0000114997	*Kosakonia sacchari* (SP1^T)	99.70%	Water	R2A	25°C 3 days
		Pantoea	HMF2842	NIBRBA0000115006	*Pantoea vagans* (LMG 24199^T)	99.60%	Sediment	R2A	30°C 3 days
Oceanospirillales	*Alcanivoracaceae*	*Alcanivorax*	IMCC12288	NIBRBA0000114857	*Alcanivorax dieselolei* (B-5^T)	99.50%	Sea water	MA	15°C 5 days
		Alcanivorax	IMCC12372	NIBRBA0000114861	*Alcanivorax marinus* (R8-12^T)	100.00%	Sea water	MA	15°C 3 days
		Alcanivorax	IMCC12398	NIBRBA0000114865	*Alcanivorax venustensis* (ISO4^T)	100.00%	Sea water	MA	15°C 3 days
	Halomonadaceae	*Kushneria*	ET2115	NIBRBA0000114951	*Kushneria marisflavi* (SW32^T)	98.63%	Gut of *Eumicrotremus asperrimus*	TSA	25°C 3 days
Pseudomonadales	*Moraxellaceae*	*Psychrobacter*	ET131	NIBRBA0000114934	*Psychrobacter celer* (SW-238^T)	98.51%	Gut of *Eumicrotremus asperrimus*	TSA	25°C 3 days
		Psychrobacter	BM06	NIBRBA0000114949	*Psychrobacter arcticus* (273-4^T)	98.67%	Gut of *Anadara granosa*	MA	25°C 3 days
		Acinetobacter	145-5	NIBRBA0000114785	*Acinetobacter bouvetii* (DSM 14964^T)	100.00%	Fresh water	TSA	25°C 2 days
	Pseudomonadaceae	*Pseudomonas*	HMF2483	NIBRBA0000114998	*Pseudomonas tolaasii* (ATCC 33618^T)	99.70%	Soil	TSA	30°C 1 day

Table 1. Continued.

Order	Family	Genus	Strain ID	NIBR ID	Most closely related species (Type strain of species)	Similarity (%)	Isolation source	Medium	Incubation conditions
Pseudomonadales	*Pseudomonadaceae*	*Pseudomonas*	HMF2783	NIBRBA0000115003	*Pseudomonas mohnii* (Ipa-2^T)	99.80%	Sediment	R2A	30°C 3 days
		Pseudomonas	R1-18	NIBRBA0000114815	*Pseudomonas migulae* (CIP 1-5470^T)	99.28%	Soil	R2A	30°C 2 days
		Pseudomonas	KM2-7	NIBRBA0000114822	*Pseudomonas entomophila* (L48^T)	99.39%	Oil-contaminated soil	R2A	25°C 2 days
		Pseudomonas	SRYB1	NIBRBA0000114777	*Pseudomonas azotoformans* (1AM1603T^T)	99.07%	Fresh water	R2A	25°C 2 days
		Pseudomonas	B2-2	NIBRBA0000114881	*Pseudomonas alcaligenes* (NBRC 14159^T)	100.00%	Soil	R2A	30°C 2 days
		Pseudomonas	B11-1	NIBRBA0000114882	*Pseudomonas seleniipraecipitans* (CA5^T)	99.77%	Sediment	R2A	30°C 2 days
		Pseudomonas	B11-2	NIBRBA0000114883	*Pseudomonas marincola* (KMM 3042^T)	99.93%	Sediment	R2A	30°C 2 days
Xanthomonadales	*Xanthomonadaceae*	*Arenimonas*	WS26	NIBRBA0000115014	*Arenimonas aquaticum* (NA-09^T)	99.86%	Fresh water	R2A	25°C 3 days
		Pseudoxanthomonas	RK 6Y-4-1	NIBRBA0000114892	*Pseudoxanthomonas japonensis* (12-3^T)	99.65%	Soil	R2A	30°C 2 days
		Dyella	N1-1	NIBRBA0000114810	*Dyella japonica* (16301^T)	99.16%	Soil	NA	30°C 2 days

the *Gammaproteobacteria* have been isolated in Korea, here in this study the following 33 species are described as unreported species in Korea.

Description of *Aeromonas eucrenophila* MD2F 12

Cells are Gram-staining-negative, flagellated, non-pigmented, and rod-shaped. Colonies are circular, raised and entire, and white colored after 2 days of incubation on R2A at 25°C. Positive for urease in API 20NE, but negative for nitrate reduction, indole production, glucose fermentation, arginine dihydrolase, esculin hydrolysis, gelatinase and β-galactosidase. 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 MD2F 12 (=NIBRBA0000114772) was isolated from a freshwater sample, Jeonju, Korea

Description of *Alteromonas australica* HMF2613

Cells are Gram-staining-negative, flagellated, non-pigmented, and rod-shaped. Colonies are circular, convex and entire, and beige colored after 2 days of incubation on MA at 37°C. Positive for nitrate reduction in API 20NE, but negative for indole production, glucose fermentation, arginine dihydrolase, urease, esculin hydrolysis, gelatinase and β-galactosidase. 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 HMF2613 (=NIBRBA 0000114999) was isolated from a seawater sample, Gangneung, Korea.

Description of *Thalassotalea piscium* HME9272

Cells are Gram-staining-negative, flagellated, non-pigmented, and rod-shaped. Colonies are circular, convex and entire, and yellowish-white colored after 3 days of incubation on MA at 25°C. Positive for nitrate reduction in API 20NE, but negative for indole production, glucose fermentation, arginine dihydrolase, urease, esculin hydrolysis, gelatinase and β-galactosidase. 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 HME9272 (=NIBRBA0000114987) was isolated from a water sample, Gyeongpo-ho in Korea.

Description of *Marinobacter lutaoensis* HME9674

Cells are Gram-staining-negative, flagellated, non-pigmented, and rod-shaped. Colonies are circular, convex and entire, and beige-colored after 3 days of incubation on MA at 25°C. Positive for nitrate reduction in API

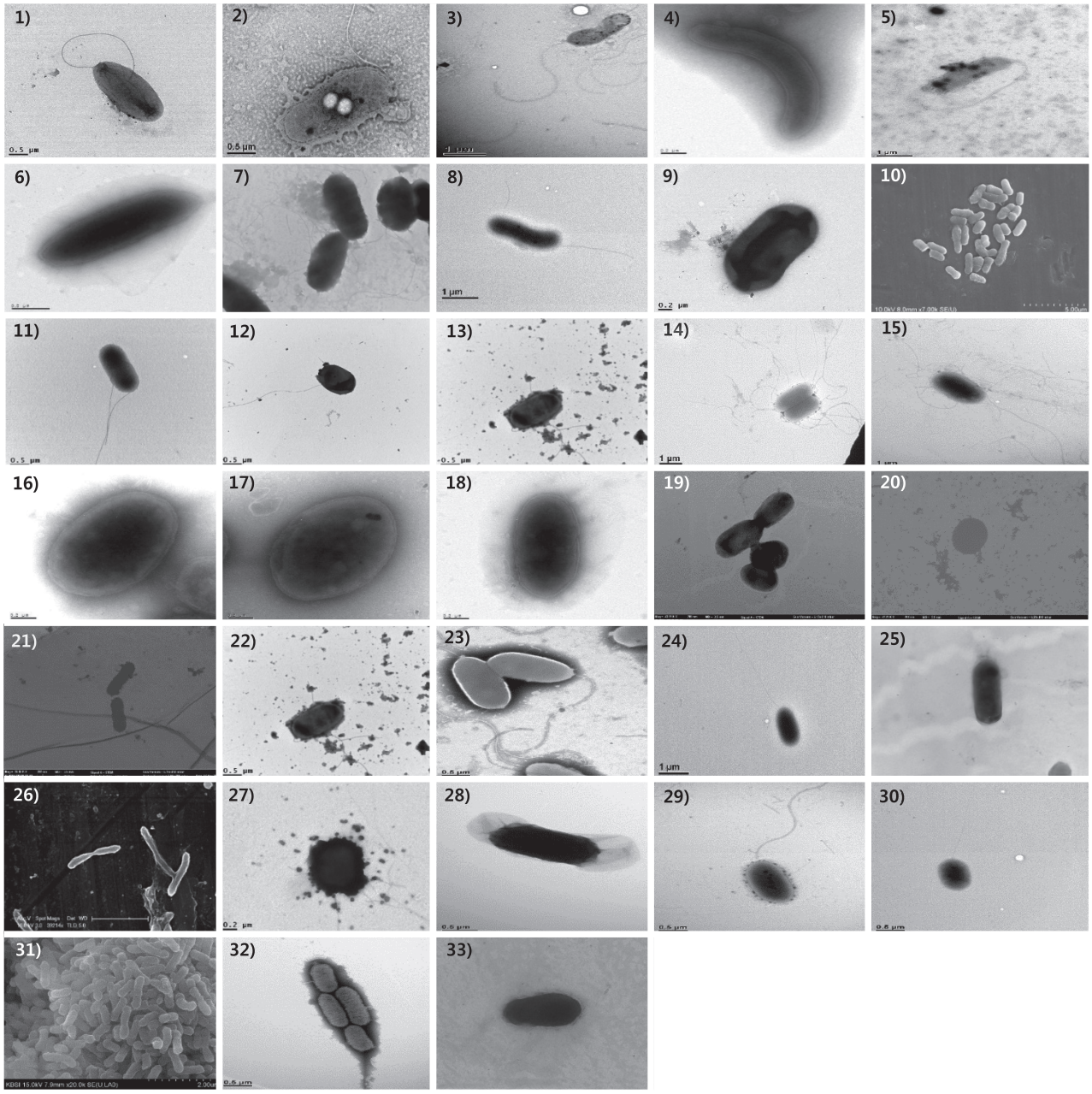

Fig. 1. Transmission electron micrographs or scanning electron micrographs of cells of the strains isolated in this study. Strains: 1, MD2F 12; 2, HMF2613; 3, HME9674; 4, IMCC12363; 5, HME9272; 6, IMCC12427; 7, 61DPR3; 8, HMF2735; 9, MIH20; 10, Duck L2; 11, WR25; 12, 142-3; 13, HME8565; 14, HMF2475; 15, HMF2842; 16, IMCC12288; 17, IMCC12372; 18, IMCC12398; 19, ET2115; 20, ET131; 21, BM06; 22, 145-5; 23, HMF2483; 24, HMF2783; 25, R1-18; 26, KM2-7; 27, SRYB1; 28, B2-2; 29, B11-1; 30, B11-2; 31, WS26; 32, RK 6Y-4-1; 33, N1-1.

20NE, but negative for indole production, glucose fermentation, arginine dihydrolase, urease, esculin hydrolysis, gelatinase and β-galactosidase. Capric acid and adipic acid are utilized. Does not utilize D-glucose, L-arabinose, D-mannose, D-mannitol, *N*-acetyl-glucosamine, D-maltose, potassium gluconate, malic acid, trisodium citrate and phenylacetic acid. Strain HME9674 (=NIBRBA0000114991) was isolated from a seawater sample, Gyeongpo-ho in Korea.

Description of *Marinobacter algicola* IMCC12363

Cells are Gram-staining-negative, non-flagellated, non-pigmented, and rod-shaped. Colonies are circular and convex, and cream colored after 3 days of incubation on MA at 15°C. Positive for nitrate reduction in API 20NE,

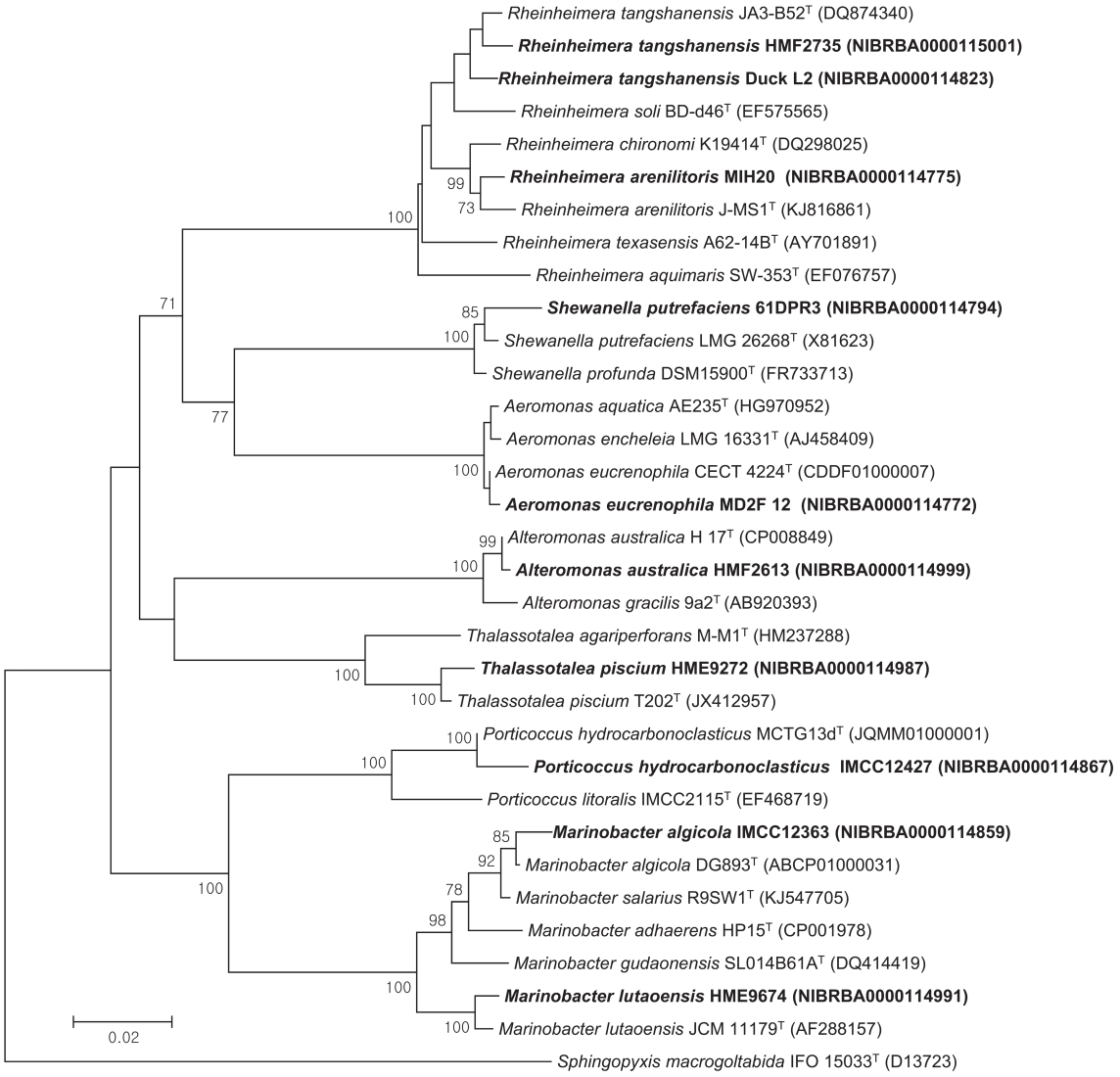

Fig. 2. Neighbor-joining phylogenetic tree, based on 16S rRNA gene sequences, showing the phylogenetic relationship between the strains isolated in this study and their relatives of the order *Aeromonadales*, *Alteromonadales* and *Chromatiales* in the class *Gammaproteobacteria*. *Sphingopyxis macrogoltabida* IFO 15033^T (D13723) was used as an outgroup. Bootstrap values (>70%) are shown above nodes for the neighbor-joining. Scale bar: 0.02 changes per nucleotide.

but negative for indole production, glucose fermentation, arginine dihydrolase, urease, esculin hydrolysis, gelatinase and β-galactosidase. 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 IMCC12363 (=NIBRBA0000114859) was isolated from a seawater sample, Sokcho, Korea.

Description of '*Porticoccus hydrocarbonoclasticus*' IMCC12427

Cells are Gram-staining-negative, non-flagellated, non-pigmented, and rod-shaped. Colonies are circular and convex, and cream colored after 3 days of incubation on MA at 15°C. Positive for nitrate reduction and β-galactosidase in API 20NE, but negative for indole production, glucose fermentation, arginine dihydrolase, urease, esculin hydrolysis and gelatinase. D-Mannose, *N*-acetyl-glucosamine and D-maltose are utilized. Does not utilize D-glucose, L-arabinose, D-mannitol, potassium gluconate, capric acid, adipic acid, malic acid, trisodium citrate and phenylacetic acid. Strain IMCC12427 (=NIBRBA 0000114867) was isolated from a seawater sample, Sokcho, Korea.

Description of '*Porticoccus hydrocarbonoclasticus*' 61DPR3

Cells are Gram-staining-negative, flagellated, non-pig-

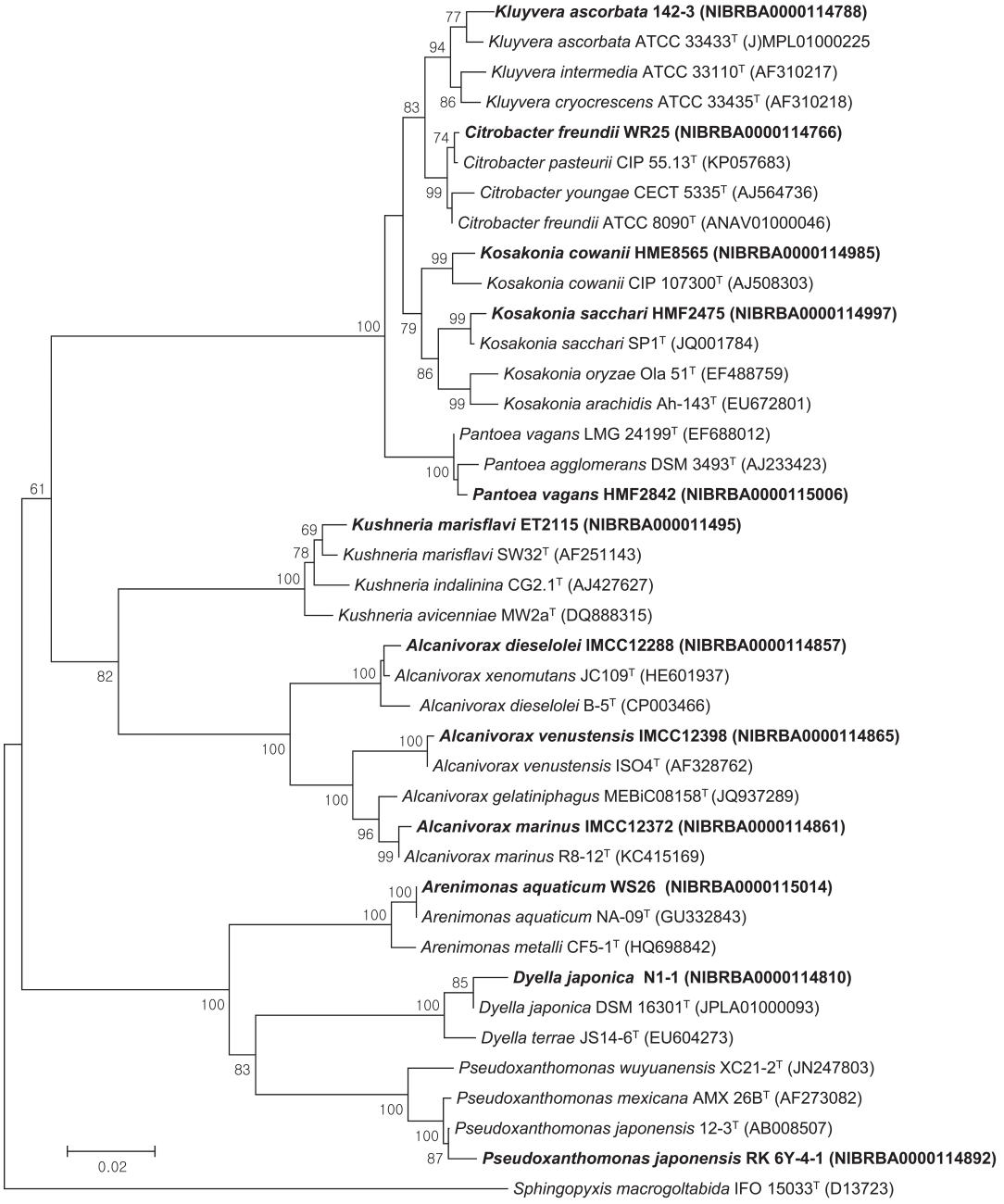

Fig. 3. Neighbor-joining phylogenetic tree, based on 16S rRNA gene sequences, showing the phylogenetic relationship between the strains isolated in this study and their relatives of the order *Enterobacteriales*, *Oceanospirillales* and *Xanthomonadales* in the class *Gammaproteobacteria*. *Sphingopyxis macrogoltabida* IFO 15033^T (D13723) was used as an outgroup. Bootstrap values (>70%) are shown above nodes for the neighbor-joining. Scale bar: 0.02 changes per nucleotide.

mented, and rod-shaped. Colonies are circular, smooth and flat, and bright brown colored after incubation on R2A at 25°C. Positive for esculin hydrolysis in API 20NE, but negative for nitrate reduction, indole production, glucose fermentation, arginine dihydrolase, urease, 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 61DPR3 (=NIBRBA0000114794) was isolated from a fresh water sample, Daejeon, Korea.

Description of *Rheinheimera tangshanensis* HMF2735

Cells are Gram-staining-negative, flagellated, non-pig-

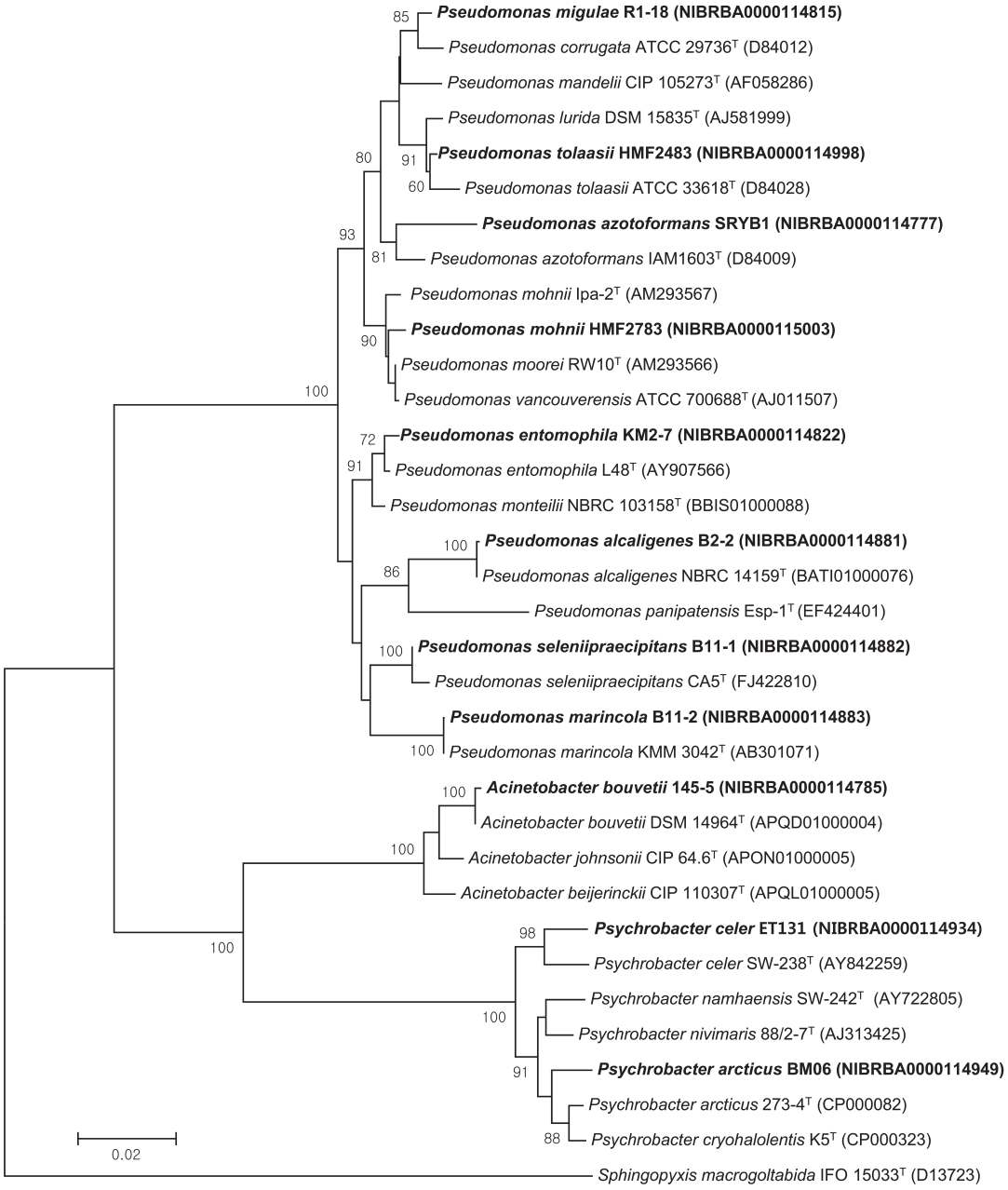

Fig. 4. Neighbor-joining phylogenetic tree, based on 16S rRNA gene sequences, showing the phylogenetic relationship between the strains isolated in this study and their relatives of the order *Pseudomonadales* in the class *Gammaproteobacteria*. *Sphingopyxis macrogoltabida* IFO 15033^T (D13723) was used as an outgroup. Bootstrap values (>70%) are shown above nodes for the neighbor-joining. Scale bar: 0.02 changes per nucleotide.

mented, and rod-shaped. Colonies are circular, raised and entire, and beige colored after 5 days of incubation on R2A at 30°C. Positive for nitrate reduction, arginine dihydrolase and gelatinase in API 20NE, but negative for indole production, glucose fermentation, arginine dihydrolase, urease, esculin hydrolysis, gelatinase and β-galactosidase. Capric 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, malic acid, trisodium citrate and phenylacetic acid. Strain HMF2735 (=NIBRBA 0000115001) was isolated from a seawater sample, Gangneung, Korea. The 16S rRNA gene sequence accession number is KP196826.

Description of *Rheinheimera arenilitoris* MIH20

Cells are Gram-staining-negative, non-flagellated, non-

pigmented, and rod-shaped. Colonies are circular, raised and entire, and brown colored after 2 days of incubation on R2A at 25°C. Positive for nitrate reduction in API 20 NE, but negative for indole production, glucose fermentation, arginine dihydrolase, urease, esculin hydrolysis, gelatinase and β-galactosidase. D-Glucose, D-mannitol and potassium gluconate are utilized. Does not utilize L-arabinose, D-mannose, *N*-acetyl-glucosamine, D-maltose, capric acid, adipic acid, malic acid, trisodium citrate and phenylacetic acid. Strain MIH20 (=NIBRBA0000114775) was isolated from a seawater sample, Marado, Korea.

Description of *Rheinheimera tangshanensis* Duck L2

Cells are Gram-staining-negative, non-flagellated, non-pigmented, and rod-shaped. Colonies are wrinkled and convex, and wavy white colored after 2 days of incubation on R2A at 25°C. Positive for esculin hydrolysis, gelatinase and β-galactosidase in API 20NE, but negative for nitrate reduction, indole production, glucose fermentation, arginine dihydrolase, urease and β-galactosidase. D-Glucose, L-arabinose, D-mannose, *N*-acetyl-glucosamine, D-maltose and malic acid are utilized. Does not utilize D-mannitol, potassium gluconate, capric acid, adipic acid, trisodium citrate and phenylacetic acid. Strain Duck L2 (=NIBRBA0000114823) was isolated from an oil-contaminated lake sample, Daejeon, Korea.

Description of *Citrobacter freundii* WR25

Cells are Gram-staining-negative, flagellated, non-pigmented, and rod-shaped. Colonies are circular, raised and entire, and white colored after 2 days of incubation on R2A at 25°C. Positive for urease and gelatinase in API 20NE, but negative for nitrate reduction, indole production, glucose fermentation, arginine dihydrolase, esculin hydrolysis and β-galactosidase. D-Mannitol, potassium gluconate, adipic acid, malic acid and phenylacetic acid are utilized. Does not utilize D-glucose, L-arabinose, D-mannose, *N*-acetyl-glucosamine, D-maltose, capric acid and trisodium citrate. Strain WR25 (=NIBRBA 0000114766) was isolated from a freshwater sample, Jeonju, Korea.

Description of *Kluyvera ascorbata* 142-3

Cells are Gram-staining-negative, flagellated, non-pigmented, and rod-shaped. Colonies are circular, raised and entire, and cream colored after 2 days of incubation on TSA at 25°C. Positive for nitrate reduction, esculin hydrolysis and β-galactosidase in API 20NE, but negative for indole production, glucose fermentation, arginine dihydrolase, urease and gelatinase. D-Glucose, D-mannose, D-mannitol, potassium gluconate and adipic acid are utilized. Does not utilize L-arabinose, N-acetyl-glucosamine, D-maltose, capric acid, malic acid, trisodium citrate and phenylacetic acid. Strain 142-3 (=NIBRBA 0000114788) was isolated from a freshwater sample, Cheongsong, Korea.

Description of *Kosakonia cowanii* HME8565

Cells are Gram-staining-negative, flagellated, non-pigmented, and rod-shaped. Colonies are circular, convex and entire, and cream colored after 3 days of incubation on R2A at 30°C. Positive for nitrate nitrate reduction, indole production, glucose fermentation, esculin hydrolysis and β-galactosidase in API 20NE, but negative for arginine dihydrolase, urease and gelatinase. D-Glucose, L-arabinose, D-mannose, D-mannitol, *N*-acetyl-glucosamine, D-maltose, potassium gluconate, malic acid, trisodium citrate and phenylacetic acid are utilized. Does not utilize capric acid and adipic acid. Strain HME8565 (=NIBRBA0000114985) was isolated from a freshwater sample, Yongin, Korea.

Description of *Kosakonia sacchari* HMF2475

Cells are Gram-staining-negative, flagellated, non-pigmented, and rod-shaped. Colonies are circular, convex and entire, and cream colored after 3 days of incubation on R2A at 25°C. Positive for nitrate reduction, esculin hydrolysis and β-galactosidase in API 20NE, but negative for indole production, glucose fermentation, arginine dihydrolase, urease and gelatinase. Adipic acid is utilized. Does not utilize D-glucose, L-arabinose, D-mannose, D-mannitol, *N*-acetyl-glucosamine, D-maltose, potassium gluconate, capric acid, malic acid, trisodium citrate and phenylacetic acid. Strain HMF2475 (=NIBRBA 0000114997) was isolated from a freshwater sample, Yongin, Korea.

Description of *Pantoea vagans* HMF2842

Cells are Gram-staining-negative, flagellated, non-pigmented, and short rod-shaped. Colonies are circular, convex and entire, and beige colored after 3 days of incubation on R2A at 30°C. Positive for nitrate reduction and arginine dihydrolase in API 20NE, but negative for indole production, glucose fermentation, urease, esculin hydrolysis, gelatinase and β-galactosidase. Capric acid, adipic acid, malic acid and trisodium citrate are utilized. Does not utilize D-glucose, L-arabinose, D-mannose, D-mannitol, *N*-acetyl-glucosamine, D-maltose, potassium gluconate and phenylacetic acid. Strain HMF2842 (=NIBRBA0000115006) was isolated from a sediment sample, Taebaek, Korea.

Description of *Alcanivorax dieselolei* IMCC12288

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

convex and opacity, and cream colored after 5 days of incubation on MA at 15°C. Positive for nitrate reduction and arginine dihydrolase in API 20NE, but negative for indole production, glucose fermentation, urease, esculin hydrolysis, gelatinase and β-galactosidase. D-Maltose and adipic acid are utilized. Does not utilize D-glucose, L-arabinose, D-mannose, D-mannitol, *N*-acetyl-glucosamine, potassium gluconate, capric acid, malic acid, trisodium citrate and phenylacetic acid. Strain IMCC12288 (=NIBRBA0000114857) was isolated from a seawater sample, Sokcho, Korea.

Description of *Alcanivorax marinus* IMCC12372

Cells are Gram-staining-negative, non-flagellated, non-pigmented, and rod-shaped. Colonies are circular, convex and opacity, and white colored after 3 days of incubation on MA at 15°C. Positive for gelatinase in API 20 NE, but negative for nitrate reduction, indole production, glucose fermentation, arginine dihydrolase, urease, esculin hydrolysis and β-galactosidase. 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 IMCC12372 (=NIBRBA0000114861) was isolated from a seawater sample, Sokcho, Korea.

Description of *Alcanivorax venustensis* IMCC12398

Cells are Gram-staining-negative, non-flagellated, non-pigmented, and rod-shaped. Colonies are punctiform, convex and dry, and white colored after 3 days of incubation on MA at 15°C. Positive for nitrate reduction, esculin hydrolysis and gelatinase in API 20NE, but negative for indole production, glucose fermentation, arginine dihydrolase, urease and β-galactosidase. *N*-Acetyl-glucosamine, D-maltose and malic acid are utilized. Does not utilize D-glucose, L-arabinose, D-mannose, D-mannitol, potassium gluconate, capric acid, adipic acid, trisodium citrate and phenylacetic acid. Strain IMCC 12398 (=NIBRBA0000114865) was isolated from a seawater sample, Sokcho, Korea.

Description of *Kushneria marisflavi* ET2115

Cells are Gram-staining-negative, non-flagellated, non-pigmented, and rod-shaped. Colonies are circular, and orange-yellow colored after 3 days of incubation on TSA at 25°C. Negative for nitrate reduction, indole production, glucose fermentation, arginine dihydrolase, urease, esculin hydrolysis, gelatinase and β-galactosidase in API 20NE. Capric acid, malic acid and trisodium citrate are utilized. Does not utilize D-glucose, L-arabinose, D-mannose, D-mannitol, *N*-acetyl-glucosamine, D-maltose, potassium gluconate, adipic acid and phenylacetic acid. Strain ET2115 (=NIBRBA0000114951) was isolated from gut of *Eumicrotremus asperrimus*.

Description of *Psychrobacter celer* ET131

Cells are Gram-staining-negative, non-flagellated, non-pigmented, and cocci-shaped. Colonies are circular, and cream colored after 3 days of incubation on TSA at 25°C. Positive for nitrate reduction, indole production, glucose fermentation, arginine dihydrolase, esculin hydrolysis, gelatinase and β-galactosidase in API 20NE, but negative for urease. D-Glucose, L-arabinose, D-mannose, D-mannitol, *N*-acetyl-glucosamine, D-maltose, potassium gluconate, capric acid, malic acid, trisodium citrate and phenylacetic acid are utilized. Does not utilize adipic acid. Strain ET131 (=NIBRBA0000114934) was isolated from gut of *Eumicrotremus asperrimus*.

Description of *Psychrobacter arcticus* BM06

Cells are Gram-staining-negative, non-flagellated, non-pigmented, and cocci-shaped. Colonies are circular, and cream colored after 3 days of incubation on MA at 25°C. Positive for nitrate reduction and arginine dihydrolase in API 20NE, but negative for indole production, glucose fermentation, urease, esculin hydrolysis, gelatinase and β-galactosidase. D-Glucose, D-mannose, *N*-acetyl-glucosamine, potassium gluconate, capric acid, malic acid, trisodium citrate and phenylacetic acid are utilized. Does not utilize L-arabinose, D-mannitol, D-maltose and adipic acid. Strain BM06 (=NIBRBA0000114949) was isolated from gut of *Anadara granosa*.

Description of *Acinetobacter bouvetii* 145-5

Cells are Gram-staining-negative, flagellated, non-pigmented, and rod-shaped. Colonies are circular, raised and entire, and cream colored after 2 days of incubation on TSA at 25°C. Positive for nitrate reduction, glucose fermentation, esculin hydrolysis and β-galactosidase in API 20NE, but negative for indole production, 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 145-5 (=NIBRBA 0000114785) was isolated from freshwater, Cheongsong, Korea.

Description of *Pseudomonas tolaasii* HMF2483

Cells are Gram-staining-negative, flagellated, non-pigmented, and rod-shaped. Colonies are circular, raised and entire, and beige colored after 1 day of incubation on TSA at 30°C. Positive for nitrate reduction in API 20NE, but negative for indole production, glucose fermentation, ar-

ginine dihydrolase, urease, esculin hydrolysis, gelatinase and β-galactosidase. 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 HMF2483 (=NIBRBA0000114998) was isolated from soil, Yongin, Korea.

Description of *Pseudomonas mohnii* HMF2783

Cells are Gram-staining-negative, flagellated, non-pigmented, and rod-shaped. Colonies are circular, raised and entire, and beige colored after 3 days of incubation on R2A at 30°C. Positive for nitrate reduction in API 20NE, but negative for indole production, glucose fermentation, arginine dihydrolase, urease, esculin hydrolysis, gelatinase and β-galactosidase. D-Glucose, L-arabinose, D-mannose, D-mannitol, potassium gluconate, capric acid, adipic acid, malic acid and trisodium citrate are utilized. Does not utilize *N*-acetyl-glucosamine, D-maltose and phenylacetic acid. Strain HMF2783 (=NIBRBA 0000115003) was isolated from sediment, Taebaek, Korea.

Description of *Pseudomonas migulae* R1-18

Cells are Gram-staining-negative, non-flagellated, non-pigmented, and rod-shaped. Colonies are slightly irregular, smooth and glistening, and white colored on R2A at 30°C. Positive for arginine dihydrolase in API 20NE, but negative for nitrate reduction, indole production, glucose fermentation, urease, esculin hydrolysis, gelatinase and β-galactosidase. D-Glucose, D-mannose, D-mannitol, *N*-acetyl-glucosamine, potassium gluconate, capric acid, malic acid and trisodium citrate are utilized. Does not utilize L-arabinose, D-maltose, adipic acid and phenylacetic acid. Strain R1-18 (=NIBRBA 0000114815) was isolated from soil, Daejeon, Korea.

Description of *Pseudomonas entomophila* KM2-7

Cells are Gram-staining-negative, non-flagellated, non-pigmented, and rod-shaped. Colonies are circular and raised, and transparent weak yellow colored after 2 days on R2A at 25°C. Positive for nitrate reduction, esculin hydrolysis and β-galactosidase in API 20NE, but negative 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, capric acid, malic acid, trisodium citrate and phenylacetic acid are utilized. Does not utilize adipic acid. Strain KM2-7 (=NIBRBA 0000114822) was isolated from oil-contaminated soil, Daejeon, Korea.

Description of *Pseudomonas azotoformans* SRYB1

Cells are Gram-staining-negative, flagellated, non-pigmented, and rod-shaped. Colonies are circular and raised, and white colored after 2 days on R2A at 25°C. Positive for esculin hydrolysis in API 20NE, but negative for nitrate reduction, indole production, glucose fermentation, arginine dihydrolase, urease, gelatinase and β-galactosidase. D-Glucose, D-mannose, D-mannitol, potassium gluconate, malic acid and trisodium citrate are utilized. Does not utilize L-arabinose, *N*-acetyl-glucosamine, D-maltose, capric acid, adipic acid and phenylacetic acid. Strain SRYB1 (=NIBRBA0000114777) was isolated from freshwater, Jeonju, Korea.

Description of *Pseudomonas alcaligenes* B2-2

Cells are Gram-staining-negative, non-flagellated, non-pigmented, and rod-shaped. Colonies are irregular, undulate and raised, and white colored after 2 days on R2A at 30°C. Positive for arginine dihydrolase in API 20NE, but negative for nitrate reduction, indole production, glucose fermentation, urease, esculin hydrolysis, gelatinase and β-galactosidase. D-Glucose, L-arabinose, D-mannose, D-mannitol, *N*-acetyl-glucosamine, potassium gluconate, capric acid, malic acid, trisodium citrate and phenylacetic acid are utilized. Does not utilize D-maltose and adipic acid. Strain B2-2 (=NIBRBA0000114881) was isolated from ginseng soil, Anseong, Korea.

Description of *Pseudomonas seleniipraecipitans* B11-1

Cells are Gram-staining-negative, flagellated, non-pigmented, and rod-shaped. Colonies are irregular, undulate and raised, and white colored after 2 days on R2A at 30 °C. Positive for arginine dihydrolase, urease and gelatinase in API 20NE, but negative for nitrate reduction, indole production, glucose fermentation, esculin hydrolysis and β-galactosidase. D-Glucose, D-mannose, potassium gluconate, capric acid, malic acid, trisodium citrate and phenylacetic acid are utilized. Does not utilize L-arabinose, D-mannitol, *N*-acetyl-glucosamine, D-maltose and adipic acid. Strain B11-1 (=NIBRBA0000114882) was isolated from flat sediment, Jebu island, Korea.

Description of *Pseudomonas marincola* B11-2

Cells are Gram-staining-negative, flagellated, non-pigmented, and rod-shaped. Colonies are irregular, undulate and raised, and yellow colored after 2 days on R2A at 30°C. Positive for urease,esculin hydrolysis, gelatinase and β-galactosidase in API 20NE, but negative for nitrate reduction, indole production, glucose fermentation and arginine dihydrolase. Does not utilize D-glucose, L-arabinose, D-mannose, D-mannitol, *N*-acetyl-glucosamine, D-maltose, potassium gluconate, capric acid, adi-

pic acid, malic acid, trisodium citrate and phenylacetic acid. Strain B11-2 (=NIBRBA0000114883) was isolated from flat sediment, Jebu island, Korea.

Description of *Arenimonas aquaticum* WS26

Cells are Gram-staining-negative, non-flagellated, non-pigmented, and rod-shaped. Colonies are circular, round and convex, and beige colored after 3 days on R2A at 25 °C. Positive for nitrate reduction, esculin hydrolysis and gelatinase in API 20NE, but negative for indole production, glucose fermentation, arginine dihydrolase, urease and β-galactosidase. D-Glucose, D-maltose, potassium gluconate and adipic acid are utilized. Does not utilize L-arabinose, D-mannose, D-mannitol, *N*-acetyl-glucosamine, capric acid, malic acid, trisodium citrate and phenylacetic acid. Strain WS26 (=NIBRBA0000115014) was isolated from a freshwater sample, Upo-wetland, Korea.

Description of *Pseudoxanthomonas japonensis* RK 6Y-4-1

Cells are Gram-staining-negative, non-flagellated, non-pigmented, and rod-shaped. Colonies are circular, entire and raised, and pale yellow colored after 2 days on R2A at 30°C. Positive for nitrate reduction, glucose fermentation, urease and β-galactosidase in API 20NE, but negative for indole production, arginine dihydrolase, esculin hydrolysis and gelatinase. D-Glucose, L-arabinose, D-mannose, D-mannitol, *N*-acetyl-glucosamine, D-maltose, potassium gluconate, malic acid, trisodium citrate and phenylacetic acid are utilized. Does not utilize capric acid and adipic acid. Strain RK 6Y-4-1 (=NIBRBA 0000114892) was isolated from ginseng soil, Anseong, Korea.

Description of *Dyella japonica* N1-1

Cells are Gram-staining-negative, non-flagellated, non-pigmented, and rod-shaped after 2 days on NA at 30°C. Positive for nitrate reduction, glucose fermentation, arginine dihydrolase, esculin hydrolysis and β-galactosidase in API 20NE, but negative for indole production, 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 N1-1 (=NIBRBA0000114810) was isolated from soil, Daejeon, Korea.

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