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The first record of nine bacterial species belonging to the phylum *Proteobacteria* in Korea

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As part of a larger study with the aim to discover indigenous prokaryotic species in Korea, nine bacterial strains were isolated and assigned to the phylum *Proteobacteria* in 2016. High 16S rRNA gene sequence similarity (>98.5%) and formation of a robust phylogenetic clades with known species indicated that each strain belongs to an independent and predefined bacterial species. This is the first report of these nine species in Korea: two strains of the *Methylobacterium*, two strains of the *Microvirga*, one strain of the *Pantoea*, and four strains of the *Psychrobacter*, all within the *Proteobacteria*. 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, *Proteobacteria*, unreported species

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Introduction

In 2016, we collected diverse soil samples and isolated unrecorded bacterial species in Korea. The identified bacterial species belonged to the phylum *Proteobacteria*. This report focuses on the isolation and description of unrecorded radiation-resistant species belonging to the phylum *Proteobacteria*.

Proteobacteria were established by Carl Woese (1987) and are one of the major phyla of Gram-negative bacteria. To date, the phylum *Proteobacteria* is known to be comprised of class *Alphaproteobacteria*, *Betaproteobacteria*, *Gammaproteobacteria*, *Deltaproteobacteria*, *Epsilonproteobacteria*, *Zetaproteobacteria* and *Oligoflexia* (List of Prokaryotic names with Standing in Nomenclature (LPSN); http://www.bacterio.net/-classifphyla.html #proteobacteria).

The *Alphaproteobacteria* with in the phylum *Proteobacteria* are a diverse class of organisms with many important biological roles including various metabolic strategies such as photosynthesis and nitrogen fixation. Moreover, they frequently adopt an intracellular life history as plant mutualists (Williams *et al*., 2007).

Gammaproteobacteria include the several ecologically, and scientifically important groups of bacteria like *Escherichia coli*, *Vibrio cholera* and *Salmonella* sp.. These bacteria play an important role as human pathogens. Although *Gammaproteobacteria* has only the taxonomic rank of class within the phylum *Proteobacteria*, it is richer in genera (~250) than all bacterial phyla except *Firmicutes* (Williams *et al*., 2010).

This report focuses on the description of bacterial species belonging to the *Proteobacteria* that are new records for Korea. This study provides the first reports in Korea of nine bacterial species belonging to three families of three orders in the *Proteobacteria*.

Materials and Methods

Various soil samples that isolated from Jeju were suspended on distilled water and serially diluted. R2A, TSA and MA agar media were inoculated with aliquots and the plates were incubated at 25°C for 3 days (Table 1). The designated strain IDs, sources, culture media, and incubation conditions are summarized in Table 1. All strains were purified as single colonies and stored as a 20 % glycerol suspension at −80°C as well as lyophilized ampoules.

Colony morphology and cell size of the strains were

Table 1. 16S rRNA gene sequence similarity, isolation source, medium, and incubation conditions of unrecorded strains belonging to the genus *Methylobacterium*

Strain ID	Most closely related species	Similarity (%)	Isolation source	Medium	Incubation conditions
15J10-8T5	*Methylobacterium hispanicum*	99.9	Soil of Jeju	R2A	25°C, 3d
KSM3-9	*Methylobacterium soli*	98.8	Soil of Jeju	R2A	25°C, 3d

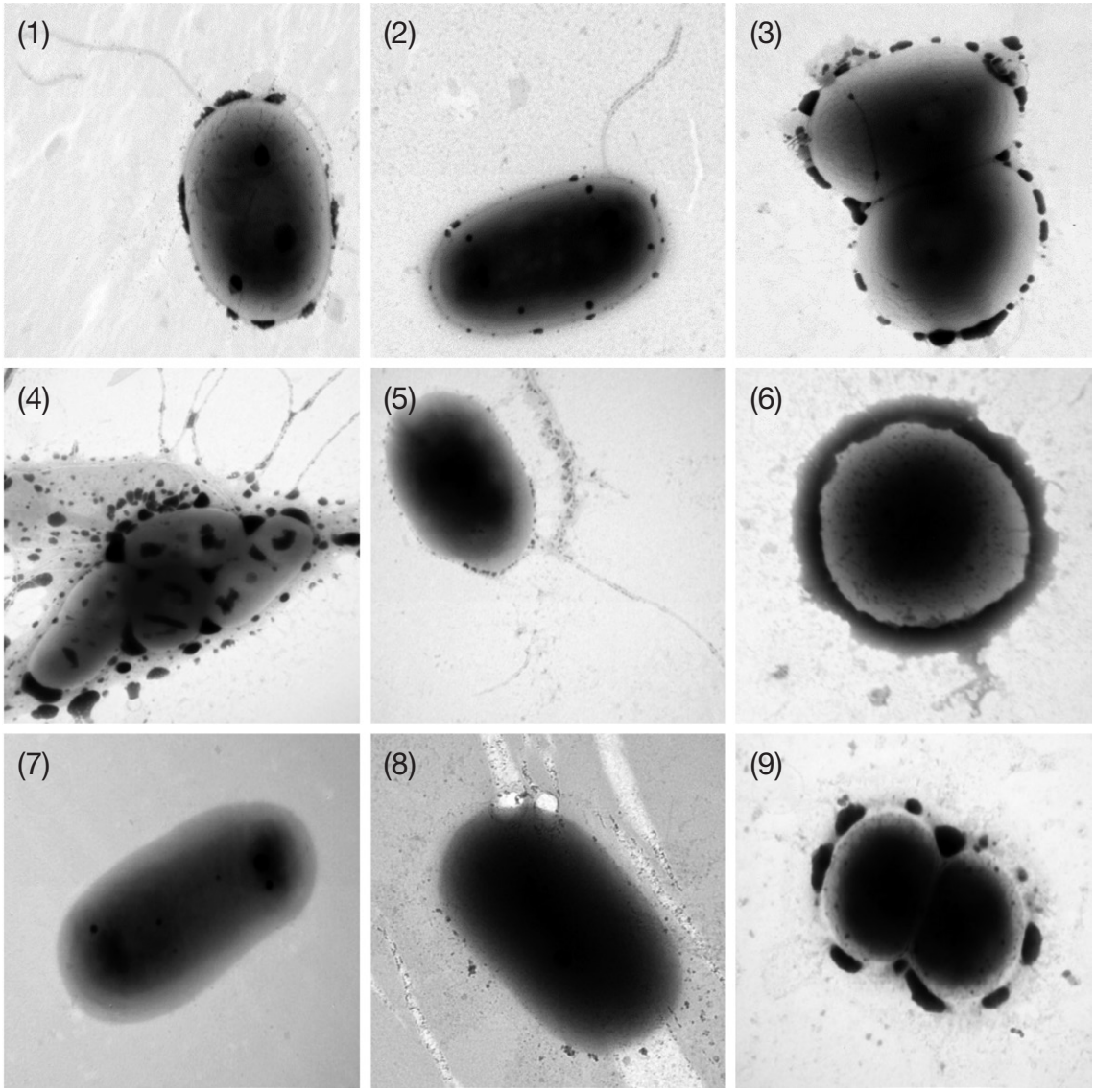

Fig. 1. Transmission electron micrographs of the strains isolated in this study. Strains: 1, 15J10-8T5; 2, KSM3-9; 3, 15J8-3; 4, 15J7-3T5; 5, 15J17K; 6, 15ML1C; 7, 216MGT3; 8, 216MGM6; 9, 16MFM4.

observed on R2A agar after cells were grown for 3 days at 25°C by using transmission electron microscopy (LIBRA 120, Carl Zeiss). Transmission electron micrographs of the strains are shown in Fig. 1. Gram reaction was performed according to the classic Gram procedure described by Doetsch (1981). Biochemical characteristics were tested using Biolog Microstation with the GEN III microplate system. A single colony was selected and emulsified into 'inoculating fluid A' (Biolog) for subsequent inoculation on to the MicroPlate test plate (Biolog). More fastidious organisms, including capnophilic strains, were cultured on alternative media, according to the manufacturer's instructions, and inocula were prepared to a specified transmittance using a turbidime-

ter, as specified in the user guide. For each isolate, each well of a MicroPlate was inoculated with 100 μL of the cell suspension using a multichannel pipette and incubated at 37°C for 24 h. MicroPlates were read in a MicroStation semi-automated reader after 24 h and results were interpreted by the identification system's software (GEN III database, version 5.2.1). The system indicated which isolates could not be identified after 20 h and required further incubation. Reading of these isolates were performed after 3 and 6 h incubation (Wragg *et al*., 2014). Amplification of the 16S rRNA gene was performed using 9F and 1492R universal bacterial primers (Weisburg *et al*., 1991). The 16SrRNA gene sequences of related taxa were obtained from EzTaxon-e (http://eztaxon-e.ezbiocloud.net) (Kim *et al*., 2012) and edited using BioEdit (Hall, 1999). Multiple alignments were performed with MUSCL (Edgar, 2004). To calculate the evolutionary distances, a two-parameter model was used (Kimura, 1983). Phylogenetic trees were constructed using the neighbor-joining method (Saitou and Nei, 1987) in MEGA5 (Tamura, 2011) with bootstrap values based on 1,000 replications (Felsenstein, 1985).

Results and Discussion

Based on the comparative 16S rRNA gene sequence analyses and phylogeny, nine strains, designated 15J10-8T5, KSM3-9, 15J8-3, 15J7-3T5, 15J17K, 15ML1C, 216MGT3, 216MGM6, 16MFM4 were assigned to the species level. Morphology and physiological characteristics are shown in the species description sections below.

Strains 15J10-8T5 and KSM3-9 were most closely related to *Methylobacterium hispanicum* GP34^T (AJ635304; 99.9% 16S rRNA gene sequence similarity), and *Methylobacterium soli* YIM 48816^T (EU860984; 98.8%), respectively (Table 1).

Strains 15J8-3 and 15J7-3T5 were most closely related to *Microvirga subterranea* DSM 14364^T (FR733708; 99.8% 16S rRNA gene sequence similarity), and *Microvirga zambiensis* WSM 3693^T (HM362433; 99.2%), respectively (Table 2).

Strains 15J17K was most closely related to *Pantoea eucrina* BD 872^T (EU216736; 99.8% 16S rRNA gene sequence similarity) (Table 3).

Strains 15ML1C, 216MGT3, 216MGM6 and 16MFM4 were most closely related to *Psychrobacter cryohalolentis* K5^T (AY660685; 99.4% 16S rRNA gene sequence similarity), *Psychrobacter maritimus* Pi2-20^T (AJ609272; 99.5%), *Psychrobacter okhotskensis* MD17^T (AB094794; 99.7%), and *Psychrobacter urativorans* ACAM 534^T (AJ609555; 99.7%), respectively (Table 4).

As expected from high 16S rRNA gene sequence similarities of the nine strains with their closest relatives, each strain formed a robust phylogenetic clade with the most closely related species (Figs. 2, 3). These results indicate that strains 15J10-8T5 and KSM3-9 are *Methylobacterium hispanicum* (Gallego *et al*., 2005), and *Methylobacterium soli* (Cao *et al*., 2011), repective-

Table 2. 16S rRNA gene sequence similarity, isolation source, medium, and incubation conditions of unrecorded strains belonging to the genus *Microvirga*

Strain ID	Most closely related species	Similarity (%)	Isolation source	Medium	Incubation conditions
15J8-3	*Microvirga subterranea*	99.8	Soil of Jeju	R2A	25°C, 3d
15J7-3T5	*Microvirga zambiensis*	99.2	Soil of Jeju	R2A	25°C, 3d

Table 3. 16S rRNA gene sequence similarity, isolation source, medium, and incubation conditions of unrecorded strain belonging to the genus *Pantoea*

Strain ID	Most closely related species	Similarity (%)	Isolation source	Medium	Incubation conditions
15J17K	*Pantoea eucrina*	99.8	Soil of Jeju	R2A	25°C, 3d

Table 4. 16S rRNA gene sequence similarity, isolation source, medium, and incubation conditions of unrecorded strains belonging to the genus *Psychrobacter*

Strain ID	Most closely related species	Similarity (%)	Isolation source	Medium	Incubation conditions
15ML1C	*Psychrobacter cryohalolentis*	99.4	Soil of Jeju	R2A	25°C, 3d
216MGT3	*Psychrobacter maritimus*	99.5	Soil of Jeju	TSA	25°C, 3d
216MGM6	*Psychrobacter okhotskensis*	99.7	Soil of Jeju	MA	25°C, 3d
16MFM4	*Psychrobacter urativorans*	99.7	Soil of Jeju	MA	25°C, 3d

TSA, Tryptic Soy Agar; MA, Marine Agar

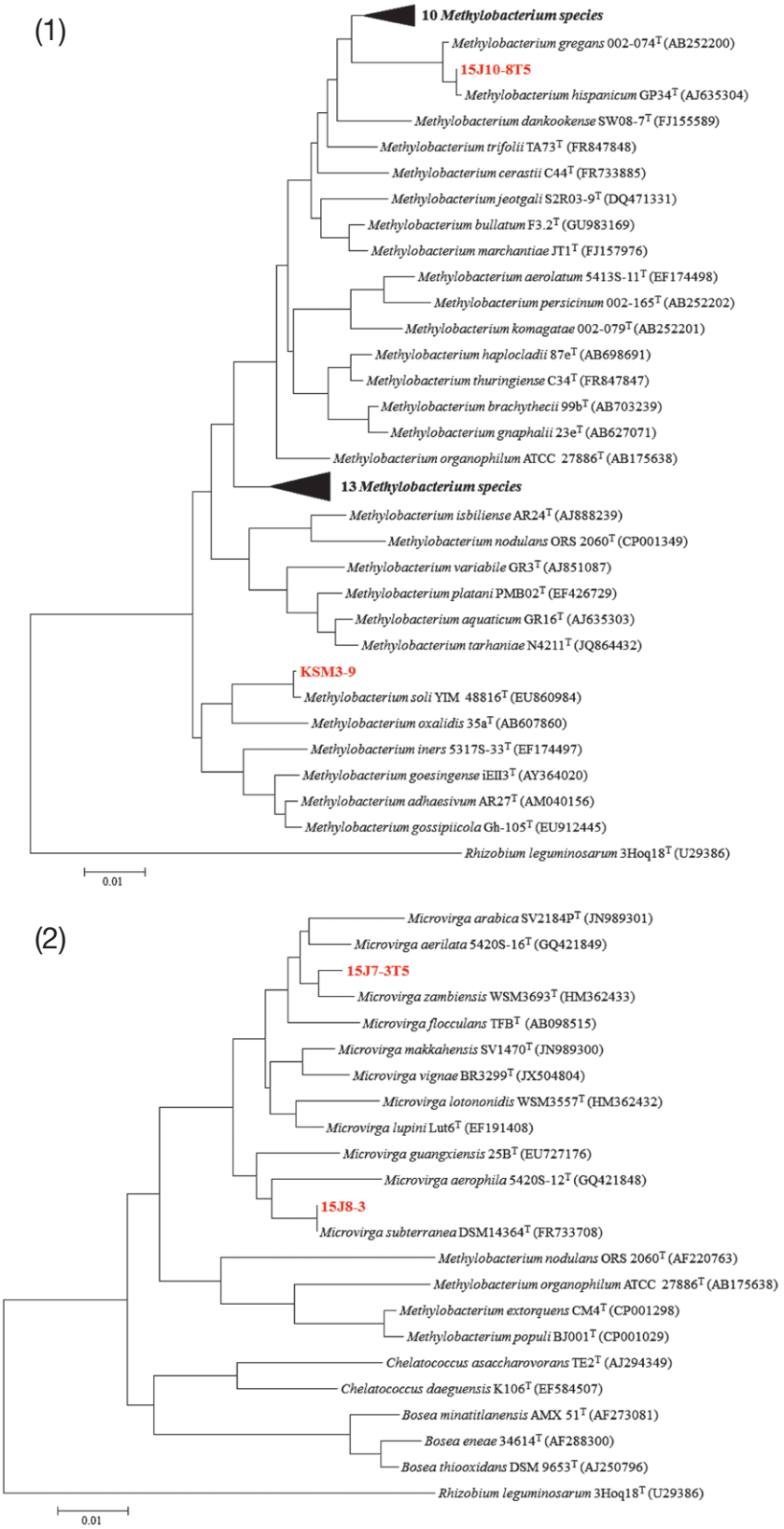

Fig. 2. Neighbor-joining phylogenetic tree based on 16S rRNA gene sequences shows the relationship between the strains isolated in this study and their relatives in the genera *Methylobacterium* (1) and *Microvirga* (2). Bootstrap values (>70%) are shown above nodes for the neighbor-joining methods. Bar: 0.01 substitutions per nucleotide position.

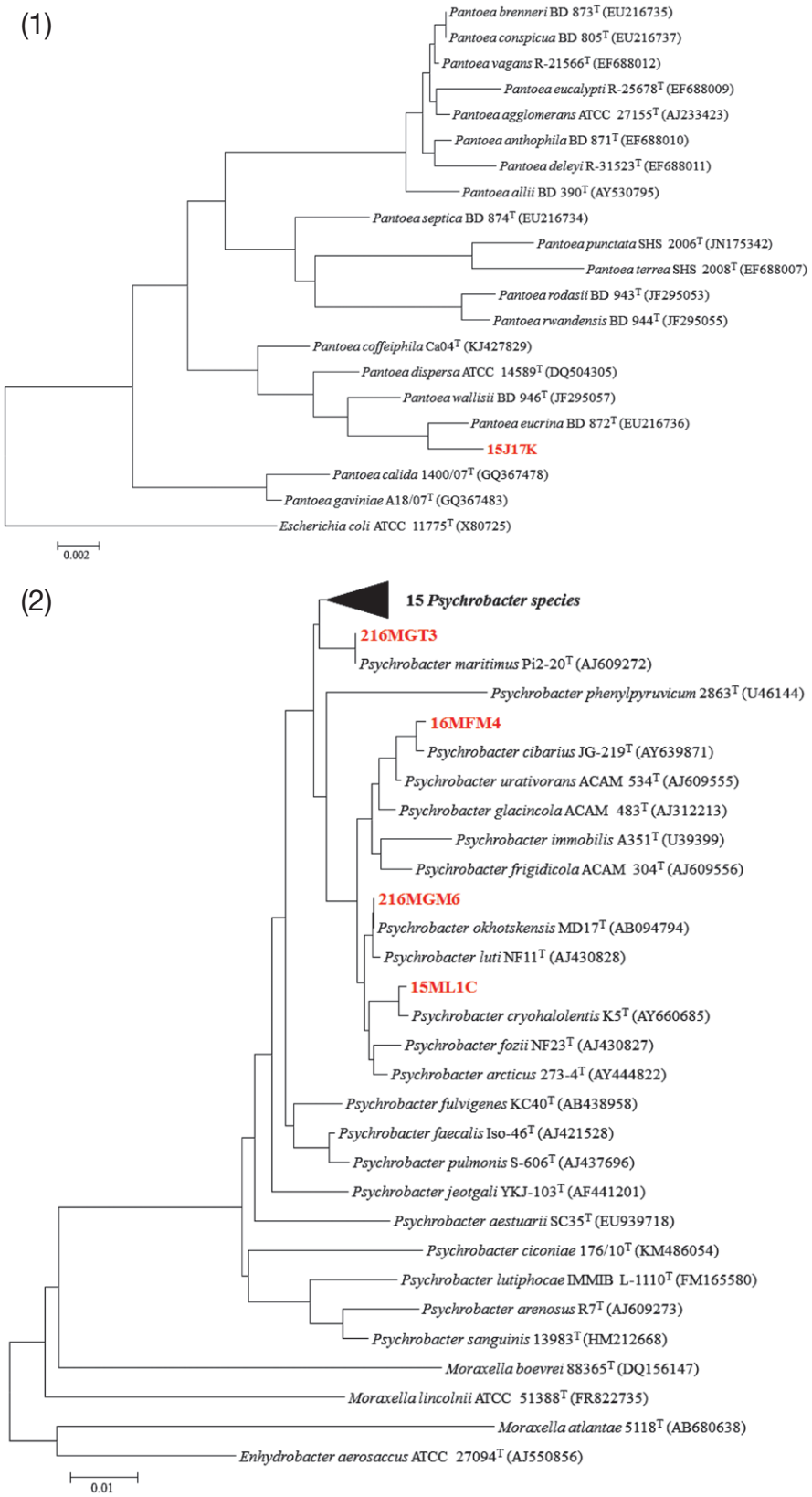

Fig. 3. Neighbor-joining phylogenetic tree based on 16S rRNA gene sequences shows the relationship between the strains isolated in this study and their relatives in the genera *Pantoea* (1) and *Psychrobacter* (2). Bootstrap values (>70%) are shown above nodes for the neighbor-joining methods. Bar: 0.002 and 0.01 substitutions per nucleotide position, respectively.

ly, and 15J8-3 and 15J7-3T5 are *Microvirga subterranea* (Kanso and Patel, 2011), and *Microvirga zambiensis* (Ardley *et al*., 2012), respectively (Fig. 2). 15J17K was identified as *Pantoea eucrina* (Brady *et al*., 2010) and 15ML1C, 216MGT3, 216MGM6 and 16MFM4 were identified as *Psychrobacter cryohalolentis* (Bakermans *et al*., 2006), *Psychrobacter maritimus* (Romanenko *et al*., 2004), *Psychrobacter okhotskensis* (Yumoto *et al*., 2003) and *Psychrobacter urativorans* (Bowman *et al*., 1996), respectively (Fig. 3).

Description of *Methylobacterium hispanicum* 15J10-8T5

Cell is Gram-stain-negative, flagellated, and rod-shaped. Colonies are pinkish-colored after 3 days of incubation on R2A at 25°C. In the GN3 microplates, acetic acid, acetoacetic acid, D-aspartic acid, bromo-succinic acid, citric acid, D-fructose, glucuronamide, L-glutamic acid, glycerol, β-hydroxy-D,L-butyric acid, α-keto-glutaric acid, L-lactic acid, D-malic acid, L-malic acid, methyl pyruvate, mucic acid, propionic acid and D-saccharic acid are utilized as sole carbon source. But *N*-acetyl-D-galactosamine, *N*-acetyl-D-mannosamine, *N*-acetyl-neuraminic acid, *N*-acetyl-D-glucosamine, L-alanine, γ-amino-butyric acid, D-arabitol, L-arginine, L-aspartic acid, D-cellobiose, dextrin, formic acid, D-fructose 6-PO4, D-fucose, L-fucose, L-galactonic acid lactone, D-galactose, D-galacturonic acid, gelatin, gentiobiose, D-gluconic acid, α-D-glucose, D-glucose-6-PO4, D-glucuronic acid, L-histidine, α-hydroxybutyric acid, p-hydroxy-phenylacetic acid, inosine, α-keto-butyric acid, D-lactic acid methyl ester, α-D-lactose, D-maltose, D-mannitol, D-mannose, D-melibiose, β-methyl-D-glucoside, 3-methyl glucose, myo-inositol, pectin, glycyl-L-proline, L-pyroglutamic acid, quinic acid, D-raffinose, L-rhamnose, D-salicin, D-serine, L-serine, D-sorbitol, stachyose, sucrose, D-trehalose, D-turanose and tween 40 are not utilized.

In sensitivity tests, the tetrazolium redox dye is reduced 1% sodium lactate, aztreonam, fusidic acid, lincomycin, nalidixic acid, pH 6, rifamycin SV, sodium butyrate, tetrazolium blue and tetrazolium violet. But not in the presence of 1% NaCl, 4% NaCl, 8% NaCl, guanidine HCl, lithium chloride, minocycline, niaproof, pH 5, potassium tellurite, D-serine, sodium bromate, troleandomycin and vancomycin. Strain 15J10-8T5 (=NIBRBAC 000499690) was isolated from a soil sample, Jeju, Korea.

Description of *Methylobacterium soli* KSM3-9

Cell is Gram-stain-negative, flagellated, and rod-shaped. Colonies are pinkish-colored after 3 days of incubation on R2A at 25°C. In the GN3 microplates, formic acid, D-fructose 6-PO4, D-fucose, L-fucose, D-galactose, glucuronamide, D-glucuronic acid, L-glutamic acid, glycerol, α-keto-glutaric acid, L-lactic acid, D-malic acid, L-malic acid, methyl pyruvate, L-rhamnose, D-saccharic acid and L-serine are utilized as sole carbon source. But acetic acid, acetoacetic acid, *N*-acetyl-D-galactosamine, *N*-acetyl-D-mannosamine, *N*-acetyl-neuraminic acid, *N*-acetyl-D-glucosamine, L-alanine, γ-amino-butryric acid, D-arabitol, L-arginine, D-aspartic acid, L-aspartic acid, bromo-succinic acid, D-cellobiose, citric acid, dextrin, D-fructose, L-galactonic acid lactone, D-galacturonic acid, gelatin, gentiobiose, D-gluconic acid, α-D-glucose, D-glucose-6-PO4, L-histidine, α-hydroxy-butyric acid, β-hydroxy-D,L-butyric acid, p-hydroxy-phenylacetic acid, inosine, α-keto-butyric acid, D-lactic acid methyl ester, α-D-lactose, D-maltose, D-mannitol, D-mannose, D-melibiose, β-methyl-D-glucoside, 3-methyl glucose, mucic acid, myo-inositol, pectin, propionic acid, glycyl-L-proline, L-pyroglutamic acid, quinic acid, D-raffinose, D-salicin, D-serine, D-sorbitol, stachyose, sucrose, D-trehalose, D-turanose and tween 40 are not utilized.

In sensitivity tests, the tetrazolium redox dye is reduced at 1% sodium lactate, aztreonam, fusidic acid, lincomycin, nalidixic acid, rifamycin SV, D-serine, sodium butyrate, tetrazolium blue, tetrazolium violet. But not in the presence of 1% NaCl, 4% NaCl, 8% NaCl, guanidine HCl, lithium chloride, minocycline, niaproof, pH 5, pH 6, potassium tellurite, sodium bromate, troleandomycin and vancomycin. Strain KSM3-9 (=NIBRBAC 000499691) was isolated from a soil sample, Jeju, Korea.

Description of *Microvirga subterranea* 15J8-3

Cell is Gram-stain-negative and rod-shaped. Colonies are pale yellow-colored after 3 days of incubation on R2A at 25°C. In the GN3 microplates, acetic acid, acetoacetic acid, *N*-acetyl-D-mannosamine, *N*-acetyl-D-glucosamine, D-cellobiose, dextrin, D-fructose, D-fructose 6-PO4, L-fucose, L-galactonic acid lactone, D-galactose, D-galacturonic acid, gelatin, gentiobiose, D-gluconic acid, α-D-glucose, D-glucose-6-PO4, glucuronamide, D-glucuronic acid, inosine, α-keto-glutaric acid, L-lactic acid, L-malic acid, D-maltose, D-mannitol, D-mannose, 3-methyl glucose, pectin, glycyl-L-proline, L-rhamnose, L-serine, D-sorbitol, stachyose, sucrose and D-turanose are utilized as sole carbon source. But *N*-acetyl-D-galactosamine, *N*-acetyl-neuraminic acid, L-alanine, γ-amino-butryric acid, D-arabitol, L-arginine, D-aspartic acid, L-aspartic acid, bromo-succinic acid, citric acid, formic acid, D-fucose, L-glutamic acid, glycerol, L-histidine, α-hydroxybutyric acid, β-hydroxy-D,L-butyric acid, p-hydroxy-phenylacetic acid, α-keto-butyric acid, D-lactic acid methyl ester, α-D-lactose, D-malic acid, D-melibiose, β-methyl-D-glucoside, methyl pyruvate, mucic

acid, myo-inositol, propionic acid, L-pyroglutamic acid, quinic acid, D-raffinose, D-saccharic acid, D-salicin, D-serine, D-trehalose and tween 40 are not utilized.

In sensitivity tests, the tetrazolium redox dye is reduced at 1% NaCl, 1% sodium lactate, fusidic acid, lincomycin, pH 6, potassium tellurite, sodium butyrate, tetrazolium blue, tetrazolium violet. But not in the presence of 4% NaCl, 8% NaCl, aztreonam, guanidine HCl, lithium chloride, minocycline, nalidixic acid, niaproof, pH 5, rifamycin SV, D-serine, sodium bromate, troleandomycin and vancomycin. Strain 15J8-3 (=NIBRBAC 000499692) was isolated from a soil sample, Jeju, Korea.

Description of *Microvirga zambiensis* 15J7-3T5

Cell is Gram-stain-negative, flagellated, and rod-shaped. Colonies are pale yellow-colored after 3 days of incubation on R2A at 25°C. In the GN3 microplates, *N*-acetyl-D-glucosamine, L-alanine, dextrin, D-fructose 6-PO4, D-fucose, L-fucose, L-galactonic acid lactone, D-galactose, D-galacturonic acid, glucuronamide, D-glucuronic acid, L-glutamic acid, glycerol, β-hydroxy-D,L-butyric acid, D-mannose, D-melibiose, methyl pyruvate, myo-inositol, L-rhamnose and D-sorbitol are utilized as sole carbon source. But acetic acid, acetoacetic acid, *N*-acetyl-D-galactosamine, *N*-acetyl-D-mannosamine, *N*-acetyl-neuraminic acid, γ-amino-butyric acid, D-arabitol, L-arginine, D-aspartic acid, L-aspartic acid, bromo-succinic acid, D-cellobiose, citric acid, formic acid, D-fructose, gelatin, gentiobiose, D-gluconic acid, α-D-glucose, D-glucose-6-PO4, L-histidine, α-hydroxybutyric acid, p-hydroxy-phenylacetic acid, inosine, α-keto-butyric acid, α-keto-glutaric acid, L-lactic acid, D-lactic acid methyl ester, α-D-lactose, D-malic acid, L-malic acid, D-maltose, D-mannitol, β-methyl-D-glucoside, 3-methyl glucose, mucic acid, pectin, propionic acid, glycyl-L-proline, L-pyroglutamic acid, quinic acid, D-raffinose, D-saccharic acid, D-salicin, D-serine, L-serine, stachyose, sucrose, D-trehalose, D-turanose and tween 40 are not utilized.

In sensitivity tests, the tetrazolium redox dye is reduced at 1% NaCl, aztreonam, fusidic acid, lincomycin, nalidixic acid, pH 6, rifamycin SV, sodium butyrate, tetrazolium blue and tetrazolium violet. But not in the presence of 1% sodium lactate, 4% NaCl, 8% NaCl, guanidine HCl, lithium chloride, minocycline, niaproof, pH 5, potassium tellurite, D-serine, sodium bromate, troleandomycin and vancomycin. Strain 15J7-3T5 (=NIBR BAC000499693) was isolated from a soil sample, Jeju, Korea.

Description of *Pantoea eucrina* 15J17K

Cell is Gram-stain-negative, flagellated, and rod-shaped. Colonies are pale lemon-colored after 3 days of incubation on R2A at 25°C. In the GN3 microplates, acetic acid, acetoacetic acid, *N*-acetyl-D-mannosamine, *N*-acetyl-D-glucosamine, L-alanine, γ-amino-butryric acid, L-arginine, L-aspartic acid, bromo-succinic acid, D-cellobiose, citric acid, dextrin, formic acid, D-fructose, D-fructose 6-PO4, D-fucose, L-fucose, L-galactonic acid lactone, D-galactose, D-galacturonic acid, gentiobiose, D-gluconic acid, α-D-glucose, D-glucose-6-PO4, glucuronamide, D-glucuronic acid, L-glutamic acid, glycerol, L-histidine, α-hydroxybutyric acid, inosine, α-keto-butyric acid, L-lactic acid, D-lactic acid methyl ester, α-D-lactose, D-malic acid, L-malic acid, D-maltose, D-mannitol, D-mannose, D-melibiose, β-methyl-D-glucoside, 3-methyl glucose, mucic acid, myo-inositol, propionic acid, glycyl-L-proline, quinic acid, L-rhamnose, D-saccharic acid, D-salicin, D-serine, L-serine, D-sorbitol, sucrose and D-trehalose are utilized as sole carbon source. But *N*-acetyl-D-galactosamine, *N*-acetyl-neuraminic acid, D-arabitol, D-aspartic acid, gelatin, β-hydroxy-D,L-butyric acid, p-hydroxy-phenylacetic acid, α-keto-glutaric acid, methyl pyruvate, pectin, L-pyroglutamic acid, D-raffinose, stachyose, D-turanose and tween 40 are not utilized.

In sensitivity tests, the tetrazolium redox dye is reduced at % NaCl, 1% sodium lactate, 4% NaCl, 8% NaCl, guanidine HCl, lincomycin, lithium chloride, niaproof, pH 5, pH 6, potassium tellurite, rifamycin SV, sodium bromate, tetrazolium blue, tetrazolium violet and troleandomycin. But not in the presence of aztreonam, fusidic acid, minocycline, nalidixic acid, D-serine, sodium butyrate and vancomycin. Strain 15J17K (=NIBRBAC 000499695) was isolated from a soil sample, Jeju, Korea.

Description of *Psychrobacter cryohalolentis* 15ML1C

Cell is Gram-stain-negative and coccus-shaped. Colonies are pale yellow-colored after 3 days of incubation on R2A at 25°C. In the GN3 microplates, acetic acid, *N*-acetyl-neuraminic acid, L-alanine, γ-amino-butryric acid, D-arabitol, L-aspartic acid, D-cellobiose, dextrin, D-fructose, D-fructose 6-PO4, D-fucose, L-fucose, D-galacturonic acid, gelatin, α-D-glucose, D-glucose-6-PO4, glucuronamide, L-glutamic acid, L-histidine, β-hydroxy-D,L-butyric acid, L-lactic acid, D-lactic acid methyl ester, α-D-lactose, D-malic acid, D-maltose, D-mannitol, D-mannose, D-melibiose, β-methyl-D-glucoside, 3-methyl glucose, myo-inositol, pectin, propionic acid, glycyl-L-proline, D-raffinose, L-rhamnose, L-serine, D-sorbitol, stachyose and tween 40 are utilized as sole carbon source. But acetoacetic acid, *N*-acetyl-D-galactosamine, *N*-acetyl-D-mannosamine, *N*-acetyl-D-glucosamine, L-arginine, D-aspartic acid, bromo-succinic acid, citric acid, formic acid, L-galactonic acid lactone, D-galactose, gentiobiose, D-gluconic acid, D-glucuronic

acid, glycerol, α-hydroxybutyric acid, p-hydroxy-phenylacetic acid, inosine, α-keto-butyric acid, α-keto-glutaric acid, L-malic acid, methyl pyruvate, mucic acid, L-pyroglutamic acid, quinic acid, D-saccharic acid, D-salicin, D-serine, sucrose, D-trehalose and D-turanose are not utilized.

In sensitivity tests, the tetrazolium redox dye is reduced at 1% NaCl, 1% sodium lactate, 4% NaCl, 8% NaCl, aztreonam, lincomycin, lithium chloride, nalidixic acid, pH 6, potassium tellurite, rifamycin SV, D-serine, sodium bromate, sodium butyrate, tetrazolium blue and troleandomycin. But not in the presence of fusidic acid, guanidine HCl, minocycline, niaproof, pH 5, tetrazolium violet and vancomycin. Strain 15ML1C (=NIBRBAC 000499696) was isolated from a soil sample, Jeju, Korea.

Description of *Psychrobacter maritimus* 216MGT3

Cell is Gram-stain-negative and rod-shaped. Colonies are pale yellow-colored after 3 days of incubation on Tryptic Soy Agar at 25°C. In the GN3 microplates, acetic acid, acetoacetic acid, *N*-acetyl-D-glucosamine, L-alanine, γ-amino-butryric acid, L-aspartic acid, bromo-succinic acid, citric acid, dextrin, formic acid, D-fructose, D-fructose 6-PO4, D-fucose, L-fucose, L-galactonic acid lactone, D-galacturonic acid, gelatin, gentiobiose, D-gluconic acid, α-D-glucose, glucuronamide, D-glucuronic acid, L-glutamic acid, L-histidine, α-hydroxybutyric acid, β-hydroxy-D,L-butyric acid, p-hydroxy-phenylacetic acid, α-keto-butyric acid, α-keto-glutaric acid, L-lactic acid, D-lactic acid methyl ester, α-D-lactose, D-malic acid, L-malic acid, D-maltose, D-mannitol, D-mannose, D-melibiose, β-methyl-D-glucoside, mucic acid, pectin, propionic acid, glycyl-L-proline, L-pyroglutamic acid, L-rhamnose, D-serine, L-serine, D-sorbitol, stachyose, sucrose, D-trehalose, D-turanose and tween 40 are utilized as sole carbon source. But *N*-acetyl-D-galactosamine, *N*-acetyl-D-mannosamine, *N*-acetyl-neuraminic acid, D-arabitol, L-arginine, D-aspartic acid, D-cellobiose, D-galactose, D-glucose-6-PO4, glycerol, inosine, 3-methyl glucose, methyl pyruvate, myo-inositol, quinic acid, D-raffinose, D-saccharic acid and D-salicin are not utilized.

In sensitivity tests, the tetrazolium redox dye is reduced at 1% NaCl, 1% sodium lactate, 4% NaCl, 8% NaCl, aztreonam, lincomycin, lithium chloride, pH 6, potassium tellurite, rifamycin SV, sodium butyrate and tetrazolium blue. But not in the presence of fusidic acid, guanidine HCl, minocycline, nalidixic acid, niaproof, pH 5, D-serine, sodium bromate, tetrazolium violet, troleandomycin and vancomycin. Strain 216MGT3 (=NIBR BAC000499697) was isolated from a soil sample, Jeju, Korea.

Description of *Psychrobacter okhotskensis* 216MGM6

Cell is Gram-stain-negative, flagellated, and rod-shaped. Colonies are pale yellow-colored after 3 days of incubation on Marine Agar at 25°C. In the GN3 microplates, acetic acid, acetoacetic acid, *N*-acetyl-D-glucosamine, L-alanine, γ-amino-butryric acid, D-arabitol, L-arginine, D-aspartic acid, L-aspartic acid, D-cellobiose, dextrin, D-fructose, D-fructose 6-PO4, D-fucose, L-fucose, L-galactonic acid lactone, D-galactose, D-galacturonic acid, gentiobiose, D-gluconic acid, α-D-glucose, D-glucose-6-PO4, glucuronamide, D-glucuronic acid, L-glutamic acid, glycerol, L-histidine, α-hydroxybutyric acid, β-hydroxy-D,L-butyric acid, α-keto-glutaric acid, L-lactic acid, D-lactic acid methyl ester, D-malic acid, L-malic acid, D-maltose, D-mannitol, D-mannose, D-melibiose, β-methyl-D-glucoside, 3-methyl glucose, methyl pyruvate, mucic acid, propionic acid, glycyl-L-proline, L-pyroglutamic acid, quinic acid, L-rhamnose, D-saccharic acid, D-salicin, L-serine, D-sorbitol, sucrose, D-trehalose, D-turanose and tween 40 are utilized as sole carbon source. But *N*-acetyl-D-galactosamine, *N*-acetyl-D-mannosamine, *N*-acetyl-neuraminic acid, bromo-succinic acid, citric acid, formic acid, gelatin, p-hydroxy-phenylacetic acid, inosine, α-keto-butyric acid, α-D-lactose, myo-inositol, pectin, D-raffinose, D-serine and stachyose are not utilized.

In sensitivity tests, the tetrazolium redox dye is reduced at 1% NaCl, 1% sodium lactate, 4% NaCl, 8% NaCl, lincomycin, lithium chloride, pH 6, potassium tellurite, rifamycin SV, sodium butyrate, tetrazolium blue and tetrazolium violet. But not in the presence of aztreonam, fusidic acid, guanidine HCl, minocycline, nalidixic acid, niaproof, pH 5, D-serine, sodium bromate, troleandomycin and vancomycin. Strain 216MGM6 (=NIBR BAC000499698) was isolated from a soil sample, Jeju, Korea.

Description of *Psychrobacter urativorans* 16MFM4

Cell is Gram-stain-negative and coccus-shaped. Colonies are pale yellow-colored after 3 days of incubation on Marine Agar at 25°C. In the GN3 microplates, acetic acid, acetoacetic acid, *N*-acetyl-D-galactosamine, L-alanine, γ-amino-butryric acid, D-arabitol, L-arginine, D-aspartic acid, L-aspartic acid, bromo-succinic acid, citric acid, dextrin, formic acid, D-fructose, D-fructose 6-PO4, D-fucose, L-fucose, L-galactonic acid lactone, D-galactose, D-galacturonic acid, gelatin, gentiobiose, D-gluconic acid, α-D-glucose, D-glucose-6-PO4, glucuronamide, D-glucuronic acid, L-glutamic acid, L-histidine, α-hydroxybutyric acid, β-hydroxy-D,L-butyric acid, α-keto-butyric acid, α-keto-glutaric acid, L-lactic acid, D-lactic acid methyl ester, α-D-lactose, D-malic acid, L-malic acid, D-maltose, D-mannose, D-melibiose,

β-methyl-D-glucoside, 3-methyl glucose, methyl pyruvate, mucic acid, pectin, propionic acid, glycyl-L-proline, L-pyroglutamic acid, quinic acid, D-raffinose, L-rhamnose, D-saccharic acid, D-serine, L-serine, stachyose and tween 40 are utilized as sole carbon source. But *N*-acetyl-D-mannosamine, *N*-acetyl-neuraminic acid, *N*-acetyl-D-glucosamine, D-cellobiose, glycerol, p-hydroxy-phenylacetic acid, inosine, D-mannitol, myo-inositol, D-salicin, D-sorbitol, sucrose, D-trehalose and D-turanose are not utilized.

In sensitivity tests, the tetrazolium redox dye is reduced at 1% NaCl, 1% sodium lactate, 4% NaCl, 8% NaCl, aztreonam, lincomycin, lithium chloride, pH 6, potassium tellurite, rifamycin SV, D-serine, sodium butyrate and tetrazolium blue. But not in the presence of fusidic acid, guanidine HCl, minocycline, nalidixic acid, niaproof, pH 5, sodium bromate, tetrazolium violet, troleandomycin and vancomycin. Strain 16MFM4 (=NIBR BAC000499699) was isolated from a soil sample, Jeju, Korea.

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