# A report of 11 unrecorded bacterial species in Korea isolated in 2017

Soohyun Maeng<sup>1</sup>, Ju-Young Kim<sup>2</sup>, Jun Hwee Jang<sup>2</sup>, Myung-Suk Kang<sup>3</sup> and Myung Kyum Kim<sup>2,\*</sup>

<sup>1</sup>Department of Public Health Sciences, Graduate School, Korea University, Seoul 02841, Republic of Korea <sup>2</sup>Department of Bio & Environmental Technology, College of Natural Science, Seoul Women's University, Seoul 01797, Republic of Korea <sup>3</sup>Migneorganium Beautrees Division National Institute of Biological Passaneon, Inches 22680, Beauthlis of Korea

<sup>3</sup>Microorganism Resources Division, National Institute of Biological Resources, Incheon 22689, Republic of Korea

\*Correspondent: biotech@swu.ac.kr

Eleven bacterial strains 17SD2\_15, 17Sr1\_23, 17SD2\_13, 17Sr1\_31, 17gy\_18, 16B15D, 16B02D, 16B04G, 16B01D, 17U4-2 and 17J28-10 assigned to the phylum *Proteobacteria* were isolated from soil samples collected from Seoul Women's University, in South Korea. The *Belnapia* species, strain 17SD2\_15 was cocci-shaped and pink-colored. The *Methylobacterium* species, strain 17Sr1\_23, 17SD2\_13, 17Sr1\_31, and 16B15D were short rod-shaped and pink-colored. The *Oxalicibacterium* species, strain 16B04G was short rod-shaped and pink-colored. The *Oxalicibacterium* species, strain 16B04G was short rod-shaped and pink-colored. The *Oxalicibacterium* species, strain 16B04G was short rod-shaped and pink-colored. The *Oxalicibacterium* species, strain 16B04G was short rod-shaped and pink-colored. The *Oxalicibacterium* species, strain 16B04G was short rod-shaped and pink-colored. The *Oxalicibacterium* species, strain 16B04G was short rod-shaped and pink-colored. The *Oxalicibacterium* species, strain 16B04G was short rod-shaped and pink-colored. The *Oxalicibacterium* species, strain 16B04G was short rod-shaped and pink-colored. The *Oxalicibacterium* species, strain 16B04G was short rod-shaped and pink-colored. The *Oxalicibacterium* species, strain 16B04G was short rod-shaped and pink-colored. The *Variovorax* species, strain 17U4-2 was cocci-shaped and yellow-colored. The *Paracoccus* species, 17J28-10 was cocci-shaped and orange-colored. Phylogenetic analysis based on 16S rRNA gene sequence showed that strains 17SD2\_15, 17Sr1\_23, 17SD2\_13, 17Sr1\_31, 17gy\_18, 16B15D, 16B02D, 16B04G, 16B01D, 17U4-2 and 17J28-10 were most closely related to *Belnapia soli* (with 99.9% similarity), *Methylobacterium gregans* (99.1%), *Methylobacterium isbiliense* (99.6%), *Methylobacterium oxalidis* (99.9%), *Microvirga aerilata* (98.7%), *Methylobacterium aerolatum* (99.0%), *Microvirga vignae* (100.0%), *Noviherbaspirillum canariense* (100.0%), *Sphingomonas desiccabilis* (100.0%), *Variovorax humicola* (99.6%), and *Paracoccus acridae* (99.1

Keywords: 16S rRNA, bacterial diversity, Proteobacteria, unreported species

© 2018 National Institute of Biological Resources DOI:10.12651/JSR.2018.7.2.135

### INTRODUCTION

In 2017, eleven unreported species were isolated from diverse soil samples collected in Korea. The current report focuses on the description of eleven unreported bacterial species belonging to phylum *Proteobacteria* that have not officially reported in Korea.

Six bacterial strains 17Sr1\_23, 17SD2\_13, 17Sr1\_31, 17gy\_18, 16B15D and 16B02D belong to the family *Methylobacteriaceae*. The family *Methylobacteriaceae* is one of the large family of *Alphaproteobacteria* (Garrity *et al.*, 2005a; Kelly *et al.*, 2014) and at present contains three genera, *Methylobacterium*, *Microvirga* and *Meganema* (http://www.bacterio.net/). The genus *Methylobacterium* is one of the largest genus containing 44 validated species. They are facultative methylotrophs able to grow on methanol and other one-carbon compounds as sources

of energy and carbon (Gallego *et al.*, 2005; Konovalova *et al.*, 2007; Cao *et al.*, 2011). The family *Methylobacteriaceae* are pink-pigmented and contain ubiquinone Q-10. The eight species of *Microvirga* and the single species of *Meganema* are not methylotrophic (Kanso and Patel 2003; Weon *et al.*, 2010; Ardley *et al.*, 2012).

Remaining five Strains 17SD2\_15, 16B04G, 16B01D, 17U4-2 and 17J28-10 belonged to the family *Acetobacteraceae*, *Oxalobacteraceae*, *Sphingomonadaceae*, *Comamonadaceae*, and *Rhodobacteraceae*, respectively. *Acetobacteraceae* are included in the order *Rhodospirillales* and 32 genera are validly published (http://www.bacterio.net/-classifphyla.html#rhodospirillales). The genera in the family *Acetobacteraceae* are basically classified into two groups, an acetous group and an acidophilic group, in the light of application, ecology, and phylogeny (Komagata *et al.*, 2014).

| Strain ID | Most closely related species | Similarity (%) | Isolation source | Medium | Incubation conditions |
|-----------|------------------------------|----------------|------------------|--------|-----------------------|
| 17SD2_15  | Belnapia soli                | 99.9           | Soil             | R2A    | 25°C, 3d              |
| 17Sr1_23  | Methylobacterium gregans     | 99.1           | Soil             | R2A    | 25°C, 3d              |
| 17SD2_13  | Methylobacterium isbiliense  | 99.6           | Soil             | R2A    | 25°C, 3d              |
| 17Sr1_31  | Methylobacterium oxalidis    | 99.9           | Soil             | R2A    | 25°C, 3d              |
| 17gy_18   | Microvirga aerilata          | 98.7           | Soil             | R2A    | 25°C, 3d              |
| 16B15D    | Methylobacterium aerolatum   | 99.0           | Soil             | R2A    | 25°C, 3d              |
| 16B02D    | Microvirga vignae            | 100.0          | Soil             | R2A    | 25°C, 3d              |
| 16B04G    | Oxalicibacterium flavum      | 100.0          | Soil             | R2A    | 25°C, 3d              |
| 16B01D    | Sphingomonas desiccabilis    | 100.0          | Soil             | R2A    | 25°C, 3d              |
| 17U4-2    | Variovorax humicola          | 99.6           | Soil             | R2A    | 25°C, 3d              |
| 17J28-10  | Paracoccus acridae           | 99.1           | Soil             | R2A    | 25°C, 3d              |

Table 1.16S rRNA gene sequence similarity, isolation source, medium, and incubation conditions of unrecorded strains.

The family Oxalobacteraceae which contains the 13 genera (Xu et al., 2005) is belonged to the order Burkholderiales. The family Sphingomonadaceae was proposed based on the 16S rRNA gene sequence phylogeny and the presence of 2'hydroxymyristol dihydrosphingosine 1-glucuronic acid (SGL-1) as major sphingoglycolipid in the cellular lipids (Kosako et al., 2000). Sphingomonadaceae is belonged to the order Sphingomonadales which contains the 18 genera at the time of writing (http://www.bacterio.net/-classifphyla.html#sphingomonadaceae). The Sphingomonadaceae family was proposed based on the 16S rRNA gene sequence phylogeny and the presence of 2'hydroxymyristol dihydrosphingosine 1-glucuronic acid (SGL-1) as major sphingoglycolipid in the cellular lipids (Kosako et al. 2000).

The family *Comamonadaceae* is one of the biggest family belonging to the order *Burkholderiales* in which contains 37 genera (http://www.bacterio.net/-classifphy-la.html#comamonadaceae). These genera form a phylogenetic cluster based on 16S rRNA gene sequence similarity of 93-97% yet, have a diverse phenotypic characteristic that includes aerobic organotrophs, anaerobic denitrifiers, Fe<sup>3+</sup>-reducing bacteria, hydrogen oxidizers, photoautotrophic and photoheterotrophic bacteria, and fermentative bacteria (Willems *et al.*, 1991).

The family *Rhodobacteraceae* are basically, aquatic bacteria that frequently thrive in marine environments. They include mainly aerobic photo- and chemohetero-trophs but also purple non-sulfur bacteria which perform photosynthesis in anaerobic environments. They are deeply involved in sulfur and carbon biogeochemical cycling and symbiosis with aquatic micro- and macroorganisms (Garrity *et al.*, 2005b).

# **MATERIAL AND METHODS**

Different soil samples were collected and suspended

in distilled water and serially diluted. The 100  $\mu$ L of the aliquot was inoculated onto R2A agar and incubated at 25°C for 3 days. The designated strain IDs, isolation sources, growth media, and incubation conditions are summarized in Table 1. All strains were purified as single colonies and stored as 20% glycerol suspension at -80°C as well as freeze-dried ampoules.

The colony morphology and cell size of the strains were observed by transmission electron microscopy (LI BRA 120, Carl Zeiss) using cells grown for 3 days at 25°C on R2A agar. Transmission electron micrograph of the strains are shown in Fig. 1. Gram reaction was performed following the classic Gram procedure described by Doetsch (1981). Biochemical characteristics were tested by using Biolog Microstation with 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 turbidimeter, as specified in the user guide. For each isolate, 100 µL of the cell suspension was inoculated into each well of the microplate, using a multichannel pipette and incubated at 37°C for 24 h, according to growth characteristics. Microplates were read in the MicroStation semi-automated reader after 24 h and results interpreted by the identification system's software (GEN III database, version 5.2.1). Whereas the system which indicates that the isolates could not be identified after 20 h and required further incubation. Such isolates were re-incubated and re-read between 3 and 6 h later (Wragg et al., 2014). Genomic DNA was extracted and 16S rRNA gene was amplified by PCR with 9F and 1492R universal bacterial primers (Weisburg et al., 1991). The 16SrRNA gene sequences of the closely related strains were obtained from EzTaxon-e (http:// eztaxon-e.ezbiocloud.net) (Kim et al., 2012) and edited



**Fig. 1.** Transmission electron micrographs of the strains isolated in this study. Strains: 1, 17SD2\_15; 2, 17Sr1\_23; 3, 17SD2\_13; 4, 17Sr1\_31; 5, 17gy\_18; 6, 16B15D; 7, 16B02D; 8, 16B04G; 9, 16B01D; 10, 17U4-2; 11, 17J28-10.

using the BioEdit program (Hall, 1999). Multiple alignments were performed with the MUSCL program (Edgar, 2004). Using the two-parameter model (Kimura, 1983) calculated the evolutionary distances. Phylogenetic trees were constructed using the neighbor-joining (Saitou and Nei, 1987) in the MEGA5 program (Tamura, 2011) with bootstrap values based on 1,000 replications (Felsenstein, 1985).

## **RESULTS AND DISCUSSION**

Based on 16S rRNA gene sequence similarity, 11 of previously unreported bacterial species were identified. The taxonomic composition and identification results are summarized in Table 1. The 11 strains were assigned to the family *Acetobacteraceae* (1 strain), *Methylobacteriaceae* (6 strain), *Oxalobacteraceae* (1 strain), and *Sphingomonadaceae* (1 strain), *Comamonadaceae* (1 strain), *Rhodobacteraceae* (1 strain) of the phylum *Proteobacteria*. At the generic level, the strains belong to 7 different genera: *Belnapia* (1 species), *Methylobacterium* (4 species), *Microvirga* (2 species), *Sphingomonas* (1 species), *Oxalicibacterium* (1 species), *Variovorax* (1 species), and *Paracoccus* (1 species). The identification of the isolates based on sequence similarity was supported by the phylogenetic trees. The neighbor-joining trees show the close relationship of the isolates and type strains of validly published species (Fig. 1). The detailed morphological and physiological characteristics are given in the strain descriptions.

As an outcome of this study, the diversity of radiation-resistant bacterial species is not reported previously in Korean ecosystems was discovered. The 11 isolates were identified as unreported species, and their phenotypic characteristics were examined through polyphasic study. Accordingly, the following 11 species are unreported species in Korea.

#### Description of Belnapia soli 17SD2\_15

Cells are Gram-stain-negative, non-flagellated, and



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 of the genus *Belnapia*. Bootstrap values (>70%) are shown above nodes for the neighbor-joining methods. Bar: 0.01 and 0.02 substitutions per nucleotide position, respectively.



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 of the genus *Methylobacterium*. Bootstrap values (>70%) are shown above nodes for the neighbor-joining methods. Bar: 0.01 and 0.02 substitutions per nucleotide position, respectively.

coccus-shaped. Colonies are pale pinkish-colored after 3 days of incubation on R2A at 25°C. In the BIOLOG GEN III, D-glucuronic acid was utilized as a sole carbon source. But acetoacetic acid, *N*-acetyl-D-mannosamine, L-alanine, L-arginine, L-aspartic acid, D-cellobiose, dextrin, D-fructose, D-fructose 6-PO<sub>4</sub>, L-galactonic acid lactone, D-galactose, D-galacturonic acid, gelatin, gentiobiose, D-gluconic acid,  $\alpha$ -D-glucose, glucuronamide, L-glutamic acid, inosine,  $\alpha$ -keto-glutaric acid, L-malic acid, D-maltose, D-mannitol, D-mannose, D-melibiose,  $\beta$ -methyl-D-glucoside, 3-methyl glucose, myo-inositol, pectin, propionic acid, D-raffinose, L-rhamnose, L-serine, stachyose, sucrose, D-trehalose, D-turanose, acetic acid, *N*-acetyl-D-galactosamine, *N*-acetyl-neuraminic acid, *N*-acetyl-D-glucosamine,  $\gamma$ -amino-butryric acid, D-arabitol, D-aspartic acid, bromo-succinic acid, citric acid, formic acid, D-fucose, L-fucose, D-glucose-6-PO<sub>4</sub>, glycerol, L-histidine,  $\alpha$ -hydroxybutyric acid,  $\beta$ -hydroxy-D, L-butyric acid, *p*-hydroxy-phenylacetic acid,  $\alpha$ -keto-butyric acid, L-lactic acid, D-lactic acid methyl ester,  $\alpha$ -D-lactose, D-malic acid, methyl pyruvate, mucic acid, D-saccharic acid, D-salicin, D-serine, D-sorbitol, and tween 40 were not utilized. In sensitivity tests, the tetrazolium redox dye was reduced at pH 6 but not at 1% NaCl, 1% sodium lactate, 4% NaCl, 8% NaCl, guanidine HCl, lithium chloride, pH 6, potassium tellurite, D-serine, sodium butyrate, aztreonam, fusidic acid,



**Fig. 4.** Neighbor-joining phylogenetic tree based on 16S rRNA gene sequences shows the relationship between the strains isolated in this study and their relatives of the genus *Methylobacterium* (1), (2). Bootstrap values (>70%) are shown above nodes for the neighbor-joining methods. Bar: 0.01 and 0.02 substitutions per nucleotide position, respectively.

lincomycin, minocycline, nalidixic acid, niaproof 4, pH 5, rifamycin SV, sodium bromate, tetrazolium blue, troleandomycin, and vancomycin.

In API 20NE system, positive for arginine dihydrolase and urease. Negative for nitrate reduction, indole production, glucose fermentation, esculin hydrolysis, gelatin hydrolysis, and  $\beta$ -galactosidase. Uses D-glucose, potassium gluconate, adipic acid, malic acid, and trisodium citrate but not L-arabinose, D-mannose, D-mannitol, *N*-acetyl-glucosamine, capric acid, D-maltose, and phenyl acetic acid as carbon sources.

In API 32GN system, positive for itaconic acid, suberic acid, sodium malonate, sodium acetate, lactic acid, L-serine, propionic acid, capric acid, potassium 2-ketogluconate, 3-hydroxybutyric acid, 4-hydroxybenzoic acid, and L-proline. Negative for L-rhamnose, *N*-acetyl-glucosamine, D-ribose, inositol, D-saccharose (sucrose), D-maltose, L-alanine, potassium 5-ketogluconate, glycogen, 3-hydroxybenzoic acid, D-mannitol, D-glucose, salicin, D-melibiose, D-fucose, D-sorbitol, L-arabinose, valeric acid, trisodium citrate, and L-histidine. Strain 17SD2\_15 (=NIBRBAC000500510) was isolated from a soil sample.

#### Description of Methylobacterium gregans 17Sr1\_23

Cells are Gram-stain-negative, non-flagellated, and short rod-shaped. Colonies are pink-colored after 3 days of incubation on R2A at 25°C. In the BIOLOG GEN III, D-glucuronic acid and formic acid is utilized as a sole carbon source. But acetoacetic acid, *N*-acetyl-D-mannosamine, L-alanine, L-arginine, L-aspartic acid, D-cel-



Fig. 5. Neighbor-joining phylogenetic tree based on 16S rRNA gene sequences shows the relationship between the strains isolated in this study and their relatives of the genus *Microvirga*. Bootstrap values (>70%) are shown above nodes for the neighbor-joining methods. Bar: 0.01 and 0.02 substitutions per nucleotide position, respectively.

lobiose, dextrin, D-fructose, D-fructose 6-PO<sub>4</sub>, L-galactonic acid lactone, D-galactose, D-galacturonic acid, gelatin, gentiobiose, D-gluconic acid,  $\alpha$ -D-glucose, glucuronamide, L-glutamic acid, inosine,  $\alpha$ -keto-glutaric acid, L-malic acid, D-maltose, D-mannitol, D-mannose, D-melibiose,  $\beta$ -methyl-D-glucoside, 3-methyl glucose, myo-inositol, pectin, propionic acid, glycyl-L-proline, L-pyroglutamic acid, quinic acid, D-raffinose, L-rhamnose, L-serine, stachyose, sucrose, D-trehalose, D-turanose acetic acid, N-acetyl-D-galactosamine, N-acetyl-neuraminic acid, N-acetyl-D-glucosamine,  $\gamma$ -amino-butryric acid, D-arabitol, D-aspartic acid, bromo-succinic acid, citric acid, D-fucose, L-fucose, D-gluco se-6-PO<sub>4</sub>, glycerol, L-histidine,  $\alpha$ -hydroxybutyric acid,  $\beta$ -hydroxy-D,L-butyric acid, p-hydroxy-phenylacetic acid,  $\alpha$ -keto-butyric acid, L-lactic acid, D-lactic acid methyl ester, a-D-lactose, D-malic acid, methyl pyruvate, mucic acid, D-saccharic acid, D-salicin, D-serine, D-sorbitol, and tween 40 were not utilized. In sensitivity tests, the tetrazolium redox dye is reduced in the presence of lincomycin, vancomycin, tetrazolium blue, nalidixic acid, potassium tellurite, and aztreonam; but not in the presence of 1% NaCl, 1% sodium lactate, 4% NaCl, 8% NaCl, guanidine HCl, lithium chloride, pH 6, D-serine, sodium butyrate, tetrazolium violet fusidic acid, minocycline, niaproof 4, pH 5, rifamycin SV, sodium bromate, and troleandomycin.

In API 20NE system, positive for the reduction of nitrates (NO<sub>3</sub>) to nitrogen (N<sub>2</sub>), arginine dihydrolase, urease, esculin hydrolysis,  $\beta$ -galactosidase, D-mannose, and D-maltose. Negative for the reduction of nitrates (NO<sub>3</sub>) to nitrite (NO<sub>2</sub><sup>-</sup>), indole production on tryptophan, glucose fermentation, gelatin hydrolysis, D-glucose, L-arabinose, D-mannitol, *N*-acetyl-D-glucosamine, potassium gluconate, capric acid, adipic acid, malic acid, trisodium citrate, and phenylacetic acid.

In API 32GN system, positive for itaconic acid, suberic acid, lactic acid, L-alanine, propionic acid, trisodium citrate, and 3-hydroxybutyric acid. Negative for L-rhamnose, *N*-acetyl-glucosamine, D-ribose, inositol, D-saccharose (sucrose), D-maltose, sodium malonate, sodium acetate, potassium 5-ketogluconate, glycogen, 3-hydroxybenzoic acid, L-serine, D-mannitol, D-glucose, salicin, D-melibiose, D-fucose, D-sorbitol, L-arabinose, capric acid, valeric acid, L-histidine, potassium 2-ketogluconate, 4-hydroxybenzoic acid, and L-proline. Strain 17Sr1\_23 (=NIBRBAC000500515) was isolated from a soil sample.

#### Description of Methylobacterium isbiliense 17SD2\_13

Cells are Gram-stain-negative, non-flagellated, and short rod-shaped. Colonies are pink-colored after 3 days of incubation on R2A at 25°C. In the BIOLOG GEN III acetoacetic acid, *N*-acetyl-D-mannosamine, L-alanine, L-arginine, L-aspartic acid, D-cellobiose, dextrin, D-fructose, D-fructose 6-PO<sub>4</sub>, L-galactonic acid lactone, D-galactose, D-galacturonic acid, gelatin, gentiobiose, D-gluconic acid,  $\alpha$ -D-glucose, glucuronamide, D-glucuronic acid, L-glutamic acid, inosine,  $\alpha$ -keto-glutaric acid, L-malic acid, D-maltose, D-mannitol, D-mannose, D-melibiose,  $\beta$ -methyl-D-glucoside, 3-methyl glucose, myo-inositol, pectin, propionic acid, glycyl-L-proline, L-pyroglutamic acid, quinic acid, D-raffinose, L-rham-



**Fig. 6.** Neighbor-joining phylogenetic tree based on 16S rRNA gene sequences shows the relationship between the strains isolated in this study and their relatives of the genus *Methylobacterium*. Bootstrap values (>70%) are shown above nodes for the neighbor-joining methods. Bar: 0.01 and 0.02 substitutions per nucleotide position, respectively.

nose, L-serine, stachyose, sucrose, D-trehalose, D-turanose, acetic acid, N-acetyl-D-galactosamine, N-acetyl-neuraminic acid, N-acetyl-D-glucosamine,  $\gamma$ -amino-butryric acid, D-arabitol, D-aspartic acid, bromo-succinic acid, citric acid, formic acid, D-fucose, L-fucose, D-glucose-6-PO<sub>4</sub>, glycerol, L-histidine,  $\alpha$ -hydroxybutyric acid,  $\beta$ -hydroxy-D, L-butyric acid, p-hydroxy-phenylacetic acid,  $\alpha$ -keto-butyric acid, L-lactic acid, D-lactic acid methyl ester,  $\alpha$ -D-lactose, D-malic acid, methyl pyruvate, mucic acid, D-saccharic acid, D-salicin, D-serine, D-sorbitol, and tween 40 were not utilized. In sensitivity tests, the tetrazolium redox dye is reduced in the presence of tetrazolium violet, tetrazolium blue, and aztreonam; but not in the presence of 1% NaCl, 1% sodium lactate, 4% NaCl, 8% NaCl, guanidine HCl, lithium chloride, pH 6, potassium tellurite, D-serine, sodium butyrate, fusidic acid, lincomycin, minocycline, nalidixic acid, niaproof 4, pH 5, rifamycin SV, sodium bromate, troleandomycin, and vancomycin.

In API 20NE system, positive for the reduction of nitrates (NO<sub>3</sub>) to nitrite (NO<sub>2</sub><sup>-</sup>), arginine dihydrolase, urease, esculin hydrolysis,  $\beta$ -galactosidase, D-glucose, L-arabinose, D-mannose, D-mannitol, *N*-acetyl-D-glucosamine, D-maltose, potassium gluconate, adipic acid, malic acid, trisodium citrate, and phenylacetic acid. Weak positive for gelatin hydrolysis. Negative for the reduction of nitrates (NO<sub>3</sub>) to nitrogen (N<sub>2</sub>), indole production on tryptophan, glucose fermentation, and capric acid.

In API 32GN system, positive for L-rhamnose, D-ribose, itaconic acid, suberic acid, sodium malonate, sodium acetate, lactic acid, potassium 2-ketogluconate, 3-hydroxybutyric acid, L-serine, D-glucose, L-arabinose, propionic acid, capric acid, valeric acid, trisodium citrate,



Fig. 7. Neighbor-joining phylogenetic tree based on 16S rRNA gene sequences shows the relationship between the strains isolated in this study and their relatives of the genus *Microvirga*. Bootstrap values (>70%) are shown above nodes for the neighbor-joining methods. Bar: 0.01 and 0.02 substitutions per nucleotide position, respectively.

4-hydroxybenzoic acid, L-proline, and 3-hydroxybenzoic acid. Negative for *N*-acetyl-glucosamine, inositol, D-saccharose (sucrose), D-maltose, L-alanine, potassium 5-ketogluconate, glycogen, L-serine, D-mannitol, salicin, D-melibiose, D-fucose, D-sorbitol, and L-histidine. Strain 17SD2\_13 (= NIBRBAC000500516) was isolated from a soil sample.

#### Description of Methylobacterium oxalidis 17Sr1\_31

Cells are Gram-stain-negative, non-flagellated, and short rod-shaped. Colonies are orange-colored after 3 days of incubation on R2A at 25°C. In the BIOLOG GEN III, L-malic acid and acetic acid is utilized as a sole carbon source. But acetoacetic acid, N-acetyl-D-mannosamine, L-alanine, L-arginine, L-aspartic acid, D-cellobiose, dextrin, D-fructose, D-fructose 6-PO<sub>4</sub>, L-galactonic acid lactone, D-galactose, D-galacturonic acid, gelatin, gentiobiose, D-gluconic acid,  $\alpha$ -D-glucose, glucuronamide, D-glucuronic acid, L-glutamic acid, inosine,  $\alpha$ -keto-glutaric acid, D-maltose, D-mannitol, D-mannose, D-melibiose,  $\beta$ -methyl-D-glucoside, 3-methyl glucose, myo-inositol, pectin, propionic acid, glycyl-L-proline, L-pyroglutamic acid, quinic acid, D-raffinose, L-rhamnose, L-serine, stachyose, sucrose, D-trehalose, D-turanose, N-acetyl-D-galactosamine, N-acetyl-neuraminic acid, N-acetyl-D-glucosamine,  $\gamma$ -amino-butryric acid, D-arabitol, D-aspartic acid, bromo-succinic acid, citric acid, formic acid, D-fucose, L-fucose, D-glucose-6-PO<sub>4</sub>, glycerol, L-histidine,  $\alpha$ -hydroxybutyric acid,  $\beta$ -hydroxy-D, L-butyric acid, *p*-hydroxy-phenylacetic acid,  $\alpha$ -keto-butyric acid, L-lactic acid, D-lactic acid methyl ester,  $\alpha$ -D-lactose, D-malic acid, methyl pyruvate, mucic acid, D-saccharic acid, D-salicin, D-serine, D-sorbitol, and tween 40 were not utilized. In sensitivity tests, the tetrazolium redox dye is reduced in the presence of lincomycin, tetrazolium blue, aztreonam; but not in the presence of 1% NaCl, 1% sodium lactate, 4% NaCl, 8% NaCl, guanidine HCl, lithium chloride, pH 6, potassium tellurite, D-serine, sodium butyrate, tetrazolium violet fusidic acid, minocycline, nalidixic acid, niaproof 4, pH 5, rifamycin SV, sodium bromate, troleandomycin, and vancomycin.

In API 20NE system, positive for the reduction of nitrates (NO<sub>3</sub>) to nitrite (NO<sub>2</sub><sup>-</sup>), arginine dihydrolase, urease, esculin hydrolysis, gelatin hydrolysis,  $\beta$ -galactosidase, D-glucose, L-arabinose, D-mannose, D-mannitol *N*-acetyl-D-glucosamine, D-maltose, potassium gluconate, adipic acid, malic acid, and trisodium citrate. Weak positive for phenylacetic acid. Negative for the reduction of nitrates (NO<sub>3</sub>) to nitrogen (N<sub>2</sub>), indole production on tryptophan, glucose fermentation, and capric acid.

In API 32GN system, positive for D-mannitol, D-glucose, D-sorbitol, valeric acid, and 3-hydroxybutyric acid. Negative for L-rhamnose, *N*-acetyl-glucosamine, D-ribose, inositol, D-saccharose (sucrose), D-maltose, itaconic acid, suberic acid, sodium malonate, sodium acetate, lactic acid, L-alanine, potassium 5-ketogluconate, glycogen, 3-hydroxybenzoic acid, L-serine, salicin, D-melibiose, D-fucose, L-arabinose, propionic acid, capric acid, 63

Herminiimonas glaciei UMB49



**Fig. 8.** Neighbor-joining phylogenetic tree based on 16S rRNA gene sequences shows the relationship between the strains isolated in this study and their relatives of the genus *Noviherbaspirillum*. Bootstrap values (>70%) are shown above nodes for the neighbor-joining methods. Bar: 0.01 and 0.02 substitutions per nucleotide position, respectively.

valeric acid, trisodium citrate, L-histidine, potassium 2-ketogluconate, 3-hydroxybutyric acid, L-serine, 4-hydroxybenzoic acid, and L-proline. Strain 17Sr1\_31 (=NI BRBAC000500517) was isolated from a soil sample.

#### Description of Microvirga aerilata 17gy\_18

Cells are Gram-stain-negative, non-flagellated, and short rod-shaped. Colonies are pink-colored after 3 days of incubation on R2A at 25°C. In the BIOLOG GEN III, acetoacetic acid, *N*-acetyl-D-mannosamine, L-alanine, L-arginine, L-aspartic acid, D-cellobiose, dextrin, D-fructose, D-fructose 6-PO<sub>4</sub>, L-galactonic acid lactone, D-galactose, D-galacturonic acid, gelatin, gentiobiose, D-gluconic acid,  $\alpha$ -D-glucose, glucuronamide, D-glucuronic acid, L-glutamic acid, inosine,  $\alpha$ -keto-glutaric acid, L-malic acid, D-maltose, D-mannitol, D-mannose, D-melibiose,  $\beta$ -methyl-D-glucoside, 3-methyl glucose, myo-inositol, pectin, propionic acid, glycyl-L-proline, L-pyroglutamic acid, quinic acid, D-raffinose, L-rhamnose, L-serine, stachyose, sucrose, D-trehalose, D-turanose acetic acid, *N*-acetyl-D-galactosamine, *N*-acetyl-neuraminic acid, *N*-acetyl-D-glucosamine,  $\gamma$ -amino-butryric acid, D-arabitol, D-aspartic acid, bromo-succinic acid, citric acid, formic acid, D-fucose, L-fucose, D-glucose-6-PO<sub>4</sub>, glycerol, L-histidine,  $\alpha$ -hydroxybu-



**Fig. 9.** Neighbor-joining phylogenetic tree based on 16S rRNA gene sequences shows the relationship between the strains isolated in this study and their relatives of the genus *Sphingomonas*. Bootstrap values (>70%) are shown above nodes for the neighbor-joining methods. Bar: 0.01 and 0.02 substitutions per nucleotide position, respectively.

tyric acid,  $\beta$ -hydroxy-D, L-butyric acid, p-hydroxy-phenylacetic acid,  $\alpha$ -keto-butyric acid, L-lactic acid, D-lactic acid methyl ester,  $\alpha$ -D-lactose, D-malic acid, methyl pyruvate, mucic acid, D-saccharic acid, D-salicin, D-serine, D-sorbitol, and tween 40 were not utilized. In sensitivity tests, the tetrazolium redox dye is reduced in the presence of tetrazolium violet; but not in the presence of 1% NaCl, 1% sodium lactate, 4% NaCl, 8% NaCl, guanidine HCl, lithium chloride, pH 6, potassium tellurite, D-serine, sodium butyrate, aztreonam, fusidic acid, lincomycin, minocycline, nalidixic acid, niaproof 4, pH 5, rifamycin SV, sodium bromate, tetrazolium blue, troleandomycin, and vancomycin.

In API 20NE system, positive for the reduction of nitrates (NO<sub>3</sub>) to nitrite (NO<sub>2</sub><sup>-</sup>), urease, esculin hydrolysis, D-glucose, L-arabinose, D-mannose, *N*-acetyl-D-glucosamine, adipic acid, phenylacetic acid, potassium 2-ketogluconate, and 3-hydroxybutyric acid. Negative for the reduction of nitrates (NO<sub>3</sub>) to nitrogen (N<sub>2</sub>), indole production on tryptophan, glucose fermentation, arginine dihydrolase, gelatin hydrolysis,  $\beta$ -galactosidase, D-mannitol, D-maltose, potassium gluconate, capric acid, malic acid, trisodium citrate.

In API 32GN system, positive for *N*-acetyl-glucosamine, D-saccharose (sucrose), itaconic acid, sodium malonate, lactic acid, D-glucose, D-fucose, and L-arabinose. Negative for L-rhamnose, D-ribose, inositol, D-maltose, suberic acid, L-alanine, potassium 5-ketogluconate, glycogen, 3-hydroxybenzoic acid, L-serine, D-mannitol, sodium acetate, salicin, D-melibiose, D-sorbitol, propionic acid, capric acid, valeric acid, trisodium citrate, L-histidine, 4-hydroxybenzoic acid, and L-proline. Strain 17gy\_18 (=NIBRBAC000500518) was isolated from a soil sample.

#### Description of Methylobacterium aerolatum 16B15D

Cells are Gram-stain-negative, non-flagellated, and



**Fig. 10.** Neighbor-joining phylogenetic tree based on 16S rRNA gene sequences shows the relationship between the strains isolated in this study and their relatives of the genus *Variovorax*. Bootstrap values (>70%) are shown above nodes for the neighbor-joining methods. Bar: 0.01 and 0.02 substitutions per nucleotide position, respectively.

short rod-shaped. Colonies are pink-colored after 3 days of incubation on R2A at 25°C. In the BIOLOG GEN III, acetoacetic acid, N-acetyl-D-mannosamine, L-alanine, L-arginine, L-aspartic acid, D-cellobiose, dextrin, D-fructose, D-fructose 6-PO<sub>4</sub>, L-galactonic acid lactone, D-galactose, D-galacturonic acid, gelatin, gentiobiose, D-gluconic acid,  $\alpha$ -D-glucose, glucuronamide, D-glucuronic acid, L-glutamic acid, inosine,  $\alpha$ -keto-glutaric acid, L-malic acid, D-maltose, D-mannitol, D-mannose, D-melibiose,  $\beta$ -methyl-D-glucoside, 3-methyl glucose, myo-inositol, pectin, propionic acid, glycyl-L-proline, L-pyroglutamic acid, quinic acid, D-raffinose, L-rhamnose, L-serine, stachyose, sucrose, D-trehalose, D-turanose acetic acid, N-acetyl-D-galactosamine, *N*-acetyl-neuraminic acid, *N*-acetyl-D-glucosamine,  $\gamma$ -amino-butryric acid, D-arabitol, D-aspartic acid, bromo-succinic acid, citric acid, formic acid, D-fucose, L-fucose, D-glucose-6-PO<sub>4</sub>, glycerol, L-histidine,  $\alpha$ -hydroxybutyric acid,  $\beta$ -hydroxy-D, L-butyric acid, p-hydroxy-phenylacetic acid,  $\alpha$ -keto-butyric acid, L-lactic acid, D-lactic acid methyl ester,  $\alpha$ -D-lactose, D-malic

acid, methyl pyruvate, mucic acid, D-saccharic acid, D-salicin, D-serine, D-sorbitol, and tween 40 were not utilized. In sensitivity tests, the tetrazolium redox dye is reduced in the presence tetrazolium violet; but not in the presence of 1% NaCl, 1% sodium lactate, 4% NaCl, 8% NaCl, guanidine HCl, lithium chloride, pH 6, potassium tellurite, D-serine, sodium butyrate, aztreonam, fusidic acid, lincomycin, minocycline, nalidixic acid, niaproof 4, pH 5, rifamycin SV, sodium bromate, tetrazolium blue, troleandomycin, and vancomycin.

In API 20NE system, positive for the reduction of nitrates (NO<sub>3</sub>) to nitrite (NO<sub>2</sub><sup>-</sup>), arginine dihydrolase, urease, D-glucose, L-arabinose, D-mannose, D-mannitol, D-maltose, potassium gluconate, adipic acid, and malic acid. Weak positive for trisodium citrate. Negative for the reduction of nitrates (NO<sub>3</sub>) to nitrogen (N<sub>2</sub>), indole production on tryptophan, glucose fermentation, esculin hydrolysis, gelatin hydrolysis,  $\beta$ -galactosidase, *N*-acetyl-D-glucosamine, capric acid, and phenylacetic acid.

In API 32GN system, positive for D-ribose, itaconic acid, D-glucose, and L-arabinose. Negative for L-rham-



Fig. 11. Neighbor-joining phylogenetic tree based on 16S rRNA gene sequences shows the relationship between the strains isolated in this study and their relatives of the genus *Paracoccus*. Bootstrap values (>70%) are shown above nodes for the neighbor-joining methods. Bar: 0.01 and 0.02 substitutions per nucleotide position, respectively.

nose, inositol, *N*-acetyl-glucosamine, D-saccharose (sucrose), D-maltose, suberic acid, sodium malonate, sodium acetate, lactic acid, L-alanine, potassium 5-ketogluconate, glycogen, 3-hydroxybenzoic acid, L-serine, D-mannitol, salicin, D-melibiose, D-fucose, D-sorbitol, propionic acid, capric acid, valeric acid, trisodium citrate, L-histidine, potassium 2-ketogluconate, 3-hydroxybutyric acid, L-serine, 4-hydroxybenzoic acid, and Lproline. Strain 16B15D (=NIBRBAC000500519) was isolated from a soil sample.

#### Description of Microvirga vignae 16B02D

Cells are Gram-stain-negative, non-flagellated, and short rod-shaped. Colonies are pink-colored after 3 days of incubation on R2A at 25°C. In the BIOLOG GEN III, acetoacetic acid, *N*-acetyl-D-mannosamine, L-alanine, L-arginine, L-aspartic acid, D-cellobiose, dextrin, D-fructose, D-fructose 6-PO<sub>4</sub>, L-galactonic acid lactone, D-galactose, D-galacturonic acid, gelatin, gentiobiose, D-gluconic acid, *a*-D-glucose, glucuronamide, D-glucuronic acid, L-glutamic acid, inosine,  $\alpha$ -keto-glutaric acid, L-malic acid, D-maltose, D-mannitol, D-mannose, D-melibiose,  $\beta$ -methyl-D-glucoside, 3-methyl glucose, myo-inositol, pectin, propionic acid, glycyl-L-proline, L-pyroglutamic acid, quinic acid, D-raffinose, L-rhamnose, L-serine, stachyose, sucrose, D-trehalose, D-turanose acetic acid, N-acetyl-D-galactosamine, N-acetyl-neuraminic acid, N-acetyl-D-glucosamine,  $\gamma$ -amino-butryric acid, D-arabitol, D-aspartic acid, bromo-succinic acid, citric acid, formic acid, D-fucose, L-fucose, D-glucose-6-PO<sub>4</sub>, glycerol, L-histidine,  $\alpha$ -hydroxybutyric acid,  $\beta$ -hydroxy-D, L-butyric acid, p-hydroxy-phenylacetic acid,  $\alpha$ -keto-butyric acid, L-lactic acid, D-lactic acid methyl ester,  $\alpha$ -D-lactose, D-malic acid, methyl pyruvate, mucic acid, D-saccharic acid, D-salicin, D-serine, D-sorbitol, and tween 40 were not utilized. In sensitivity tests, the tetrazolium redox dye is reduced in the presence tetrazolium violet; but not in the presence of 1% NaCl, 1% sodium lactate, 4% NaCl, 8% NaCl, guanidine HCl, lithium chloride, pH 6, potassium tellurite, D-serine, sodium butyrate, aztreonam, fusidic acid, lincomycin, minocycline, nalidixic acid, niaproof 4, pH 5, rifamycin SV, sodium bromate, tetrazolium blue, troleandomycin, and vancomycin.

In API 20NE system, positive for arginine dihydrolase, urease,  $\beta$ -galactosidase, D-glucose, L-arabinose, D-mannose, malic acid, and trisodium citrate. Weak positive for D-mannitol. Negative for the reduction of nitrates (NO<sub>3</sub>) to nitrite (NO<sub>2</sub><sup>-</sup>), reduction of nitrates (NO<sub>3</sub>) to nitrogen (N<sub>2</sub>), indole production on tryptophan, glucose fermentation, esculin hydrolysis, gelatin hydrolysis, *N*-acetyl-D-glucosamine, D-maltose, potassium gluconate, capric acid, adipic acid, and phenylacetic acid.

In API 32GN system, positive for *N*-acetyl-glucosamine, inoitol, L-alanine, D-mannitol, D-glucose, salicin, D-sorbitol, L-arabinose, propionic acid, potassium 2-ketogluconate, 4-hydroxybenzoic acid, and L-proline. Negative for L-rhamnose, D-ribose, D-saccharose (sucrose), D-maltose, itaconic acid, suberic acid, sodium malonate, sodium acetate, lactic acid, potassium 5-ketogluconate, glycogen, 3-hydroxybenzoic acid, L-serine, D-melibiose, D-fucose, capric acid, valeric acid, trisodium citrate, L-histidine, and 3-hydroxybutyric acid. Strain 16B02D (=NIBRBAC000500520) was isolated from a soil sample.

#### Description of Oxalicibacterium flavum 16B04G

Cells are Gram-stain-negative and short rod-shaped. Colonies are pink-colored after 3 days of incubation on R2A at 25°C. In the BIOLOG GEN III, acetoacetic acid, N-acetyl-D-mannosamine, L-aspartic acid, D-cellobiose, dextrin, D-fructose, D-fructose 6-PO<sub>4</sub>, L-galactonic acid lactone, D-galactose, D-galacturonic acid, gentiobiose, D-gluconic acid,  $\alpha$ -D-glucose, D-glucuronic acid, L-glutamic acid, a-keto-glutaric acid, L-malic acid, D-maltose, D-mannitol, D-mannose,  $\beta$ -methyl-D-glucoside, 3-methyl glucose, myo-inositol, pectin, propionic acid, glycyl-L-proline, L-pyroglutamic acid, L-rhamnose, L-serine, stachyose, sucrose, D-trehalose, D-turanose, acetic acid, N-acetyl-neuraminic acid, N-acetyl-D-glucosamine, y-amino-butryric acid, D-arabitol, D-aspartic acid, bromo-succinic acid, citric acid, formic acid, D-fucose, L-fucose, D-glucose-6-PO<sub>4</sub>, glycerol, L-histidine,  $\alpha$ -hydroxybutyric acid,  $\beta$ -hydroxy-D,L-butyric acid, *p*-hydroxy-phenylacetic acid,  $\alpha$ -keto-butyric acid, L-lactic acid, D-lactic acid methyl ester,  $\alpha$ -D-lactose, D-malic acid, methyl pyruvate, mucic acid, D-saccharic acid, D-sorbitol, tween 40 are utilized as sole carbon source. But D-raffinose, D-melibiose, D-salicin, N-acetyl-D-galactosamine, inosine, D-serine, L-alanine, L-arginine, glucuronamide, and quinic acid were not utilized. In sensitivity tests, the tetrazolium redox dye is reduced in the presence of 1% NaCl, 1% sodium lactate, 4% NaCl, lithium chloride, pH 6, potassium tellurite, D-serine, sodium butyrate, aztreonam, nalidixic acid, niaproof 4, sodium bromate; but not in the presence of pH 5, 8% NaCl, fusidic acid, troleandomycin, rifamycin SV, minocycline, gelatin, lincomycin, guanidine HCl, vancomycin tetrazolium violet, tetrazolium blue.

In API 20NE system, positive for arginine dihydrolase, urease, esculin hydrolysis, D-glucose, D-mannose, D-mannitol, *N*-acetyl-D-glucosamine, adipic acid, malic acid, trisodium citrate, and potassium gluconate. Negative for the reduction of nitrates (NO<sub>3</sub>) to nitrite (NO<sub>2</sub><sup>-</sup>), reduction of nitrates (NO<sub>3</sub>) to nitrogen (N<sub>2</sub>), indole production on tryptophan, glucose fermentation, gelatin hydrolysis,  $\beta$ -galactosidase, L-arabinose, D-maltose, capric acid, and phenylacetic acid.

In API 32GN system, positive for D-ribose, inositol, D-saccharose (sucrose), *N*-acetyl-glucosamine, suberic acid, lactic acid, D-mannitol, D-glucose, salicin, L-arabinose, and 4-hydroxybenzoic acid. Negative for L-rhamnose, D-maltose, itaconic acid, sodium malonate, sodium acetate, L-alanine, potassium 5-ketogluconate, glycogen, 3-hydroxybenzoic acid, L-serine, D-melibiose, D-fucose, D-sorbitol, propionic acid, capric acid, valeric acid, trisodium citrate, L-histidine, potassium 2-ketogluconate, 3-hydroxybutyric acid, and L-proline. Strain 16B04G (=NIBRBAC000500521) was isolated from a soil sample.

#### Description of Sphingomonas desiccabilis 16B01D

Cells are Gram-stain-negative and short rod-shaped. Colonies are yellow-colored after 3 days of incubation on R2A at 25°C. In the BIOLOG GEN III, dextrin, D-maltose, D-trehalose, D-cellobiose, gentiobiose, sucrose, D-turanose, D-raffinose,  $\alpha$ -D-lactose, D-melibiose,  $\beta$ methyl-D-glucoside, D-salicin, D-mannose, D-galactose, D-galacturonic acid, and D-glucuronic acid were utilized as a sole carbon source. But acetoacetic acid, N-acetyl-D-mannosamine, L-alanine, L-arginine, L-aspartic acid, D-fructose, D-fructose 6-PO<sub>4</sub>, L-galactonic acid lactone, gelatin, D-gluconic acid,  $\alpha$ -D-glucose, glucuronamide, L-glutamic acid, inosine,  $\alpha$ -keto-glutaric acid, L-malic acid, D-mannitol, 3-methyl glucose, myo-inositol, pectin, propionic acid, glycyl-L-proline, L-pyroglutamic acid, quinic acid, L-rhamnose, L-serine, stachyose, acetic acid, N-acetyl-D-galactosamine, N-acetyl-neuraminic acid, N-acetyl-D-glucosamine,  $\gamma$ -amino-butryric acid, D-arabitol, D-aspartic acid, bromo-succinic acid, citric acid, formic acid, D-fucose, L-fucose, D-glucose-6-PO<sub>4</sub>, glycerol, L-histidine,  $\alpha$ -hydroxybutyric acid,  $\beta$ -hydroxy-D, L-butyric acid, p-hydroxy-phenylacetic acid,  $\alpha$ -keto-butyric acid, L-lactic acid, D-lactic acid methyl ester, D-malic acid, methyl pyruvate, mucic acid, D-saccharic acid, D-serine, D-sorbitol, and tween 40 were not utilized. In sensitivity tests, the tetrazolium redox dye is reduced at pH 6 but not at pH 5; the dye is reduced in the presence of pH 6, 1% NaCl, 1% sodium lactate, rifamycin SV, tetrazolium blue; but not in the presence of sodium lactate, 4% NaCl, 8% NaCl, guanidine HCl, lithium chloride, potassium tellurite, D-serine, sodium butyrate, tetrazolium violet aztreonam, fusidic acid, lincomycin, minocycline, nalidixic acid, niaproof 4, pH 5, sodium bromate, troleandomycin, and vancomycin.

In API 20NE system, positive for of arginine dihydrolase, urease, esculin hydrolysis,  $\beta$ -galactosidase, D-glucose, L-arabinose, D-mannose, N-acetyl-D-glucosamine, and D-maltose. Weak positive for D-mannitol, potassium gluconate, adipic acid, malic acid, and trisodium citrate. Negative for the reduction of nitrates (NO<sub>3</sub>) to nitrite (NO<sub>2</sub><sup>-</sup>), reduction of nitrates (NO<sub>3</sub>) to nitrogen (N<sub>2</sub>), indole production on tryptophan, glucose fermentation, gelatin hydrolysis, capric acid, and phenylacetic acid.

In API 32GN system, positive for *N*-acetyl-glucosamine, D-saccharose (sucrose), D-maltose, suberic acid, sodium malonate, sodium acetate, lactic acid, L-alanine, potassium 5-ketogluconate, glycogen, 3-hydroxybenzoic acid, L-serine, D-glucose, salicin, D-melibiose, D-sorbitol, L-arabinose, valeric acid, trisodium citrate, L-histidine, potassium 2-ketogluconate, 3-hydroxybutyric acid, 4-hydroxybenzoic acid, and L-proline. Negative for L-rhamnose, D-ribose, inositol, itaconic acid, D-mannitol, D-fucose, propionic acid, and capric acid. Strain 16B01D (=NIBRBAC000500522) was isolated from a soil sample.

#### Description of Variovorax humicola 17U4-2

Cells are Gram-stain-positive and short rod-shaped. Colonies are yellow-colored after 3 days of incubation on R2A at 25°C. In the BIOLOG GEN III, D-maltose, D-trehalose, D-cellobiolose, gentiobiose, sucrose, D-turanose, stachyose, D-raffinose, D-melibiose,  $\beta$ -D-glucoside, D-salicin, neuraminic acid,  $\alpha$ -D-glucose, D-fructose, D-mannitol, glycerol, L-histidine, D-gluconic acid, and L-malic acid were utilized as sole carbon source. But dextrin,  $\alpha$ -D-lactose, N-glucosamine,  $\beta$ -mannosamin, N-galactosamin, D-mannose, D-galactose, 6-methyl-glucose, D-fucose, L-fucose, L-rhamnose, inosine, D-sorbitol, D-arabitol, myo-inositol, D-glucose, D-fructose, D-aspartic acid, D-serine, gelatin, glycyl-L-proline, L-alanine, L-arginine, L-aspartic acid, L-glutamic acid, pyroglutamic acid, L-serine, pectin, galacturonic acid, L-galactonic lactone, D-glucuronic acid, glucuronamide, mucic acid, quinic acid, D-saccharic acid, phenylacetic

acid, methyl pyruvate, D-lactic acid, L-lactic acid, citric acid,  $\alpha$ -glutaric acid, D-malic acid, succinic acid, tween 40,  $\gamma$ -butryric acid,  $\alpha$ -butryric acid,  $\beta$ -butryric acid, acetoacetic acid, propionic acid, acetic acid, and formic acid were not utilized. In sensitivity tests, the tetrazolium redox dye is reduced at pH 6 but not at pH 5; the dye is reduced in the presence of 1% NaCl, 4% NaCl, sodium lactate, D-serine, tetrazolium violet, nalidixic acid, lithium chloride, potassium tellurite, aztreonam, and sodium butyrate; but not in the presence of 8% NaCl, fusidic acid, troleandomycin, rifamycin SV, minocycline, lincomycin, guanidine HCl, niaproof 4, vancomycin, tetrazolium blue, and sodium bromate.

In API 20NE system, positive for of reduction of nitrates (NO<sub>3</sub>) to nitrogen (N<sub>2</sub>), arginine dihydrolase, urease, esculin hydrolysis,  $\beta$ -galactosidase, D-glucose, Larabinose, D-mannose, D-mannitol, *N*-acetyl-D-glucosamine, D-maltose, potassium gluconate, adipic acid, malic acid, trisodium citrate, and phenylacetic acid. Negative for reduction of nitrates (NO<sub>3</sub>) to nitrite (NO<sub>2</sub><sup>-</sup>), indole production on tryptophan, glucose fermentation, gelatin hydrolysis, and capric acid.

In API 32GN system, positive for L-rhamnose, *N*-acetyl-glucosamine, D-ribose, D-saccharose (sucrose), D-maltose, suberic acid, sodium acetate, lactic acid, Lalanine, potassium 5-ketogluconate, 3-hydroxybenzoic acid, D-mannitol, D-glucose, salicin, D-melibiose, D-sorbitol, L-arabinose, propionic acid, valeric acid, L-histidine, potassium 2-ketogluconate, 3-hydroxybutyric acid, 4-hydroxybenzoic acid, and L-proline. Negative for inositol, itaconic acid, sodium malonate, glycogen, L-serine, D-fucose, capric acid, and trisodium citrate. Strain 17U4-2 (=NIBRBAC000500500) was isolated from a soil sample.

#### Description of Paracoccus acridae 17J28-10

Cells are Gram-stain-negative and cocci-shaped. Colonies are orange-colored after 3 days of incubation on R2A at 25°C. In the BIOLOG GEN III, L-alanine, pyroglutamic acid, L-lactic acid, L-malic acid, acetoacetic acid, and acetic acid is utilized as sole carbon source. But D-maltose, D-trehalose, D-cellobiolose, gentiobiose, sucrose, D-turanose, stachyose, D-raffinose, Dmelibiose,  $\beta$ -D-glucoside, D-salicin, neuraminic acid,  $\alpha$ -D-glucose, D-fructose, D-mannitol, glycerol, L-histidine, D-gluconic acid, dextrin,  $\alpha$ -D-lactose, N-glucosamine,  $\beta$ -mannosamin, N-galactosamin, D-mannose, D-galactose, 6-methyl-glucose, D-fucose, L-fucose, L-rhamnose, inosine, D-sorbitol, D-arabitol, myo-inositol, D-glucose, D-fructose, D-aspartic acid, D-serine, gelatin, glycyl-L-proline, L-arginine, L-aspartic acid, L-glutamic acid, L-serine, pectin, galacturonic acid, L-galactonic lactone, D-glucuronic acid, glucuronamide, mucic acid, uinic acid, D-saccharic acid, pheylacetic acid, methyl pyruvate, D-lactic acid, citric acid,  $\alpha$ -glutaric acid, D-malic acid, succinic acid, tween40,  $\gamma$ -butryric acid,  $\alpha$ -butryric acid,  $\beta$ -butryric acid, propionic acid, and formic acid were not utilized. In sensitivity tests, the tetrazolium redox dye is reduced in the presence of pH 6, 1% NaCl, sodium lactate, and lithium chloride; but not in the presence of pH 5, 4% NaCl, 8% NaCl, fusidic acid, D-serine, troleandomycin, rifamycin SV, minocycline, lincomycin, guanidine HCl, niaproof 4, vancomycin, tetrazolium violet, tetrazolium Blue, nalidixic acid, potassium tellurite, aztreonam, sodium butyrate, and sodium bromate.

In API 20NE system, positive for of reduction of nitrates (NO<sub>3</sub>) to nitrogen (N<sub>2</sub>), arginine dihydrolase, urease, D-glucose, L-arabinose, D-mannose, D-mannitol, and phenylacetic acid. Negative for reduction of nitrates (NO<sub>3</sub>) to nitrite (NO<sub>2</sub><sup>-</sup>), indole production on tryptophan, glucose fermentation, esculin hydrolysis, gelatin hydrolysis,  $\beta$ -galactosidase, *N*-acetyl-D-glucosamine, D-maltose, potassium gluconate, capric acid, adipic acid, malic acid, and trisodium citrate.

In API 32GN system, positive for L-rhamnose, D-ribose, inositol, lactic acid, L-alanine, potassium 5-ketogluconate, 3-hydroxybenzoic acid, L-serine, D-mannitol, D-glucose, salicin, D-sorbitol, propionic acid, L-histidine, 3-hydroxybutyric acid, 4-hydroxybenzoi acid, and L-proline. Negative for *N*-acetyl-glucosamine, D-saccharose (sucrose), D-maltose, itaconic acid, suberic acid, sodium malonate, sodium acetate, glycogen, D-melibiose, D-fucose, L-arabinose, capric acid, valeric acid, trisodium citrate, and potassium 2-ketogluconate. Strain 17J28-10 (=NIBRBAC000500501) was isolated from a soil sample.

#### **ACKNOWLEDGMENTS**

This work was supported by a research grant from Seoul Women's University (2018) and by a grant from the National Institute of Biological Resources (NIBR), funded by the Ministry of Environment (MOE) of the Republic of Korea (NIBR201701107).

#### REFERENCES

Ardley, J.K. et al. 2012. Microvirga lupini sp. nov., Microvirga lotononidis sp. nov. and Microvirga zambiensis sp. nov. are alphaproteobacterial root-nodule bacteria that specifically nodulate and fix nitrogen with geographically and taxonomically separate legume hosts. International Journal of Systematic and Evolutionary Microbiology 62:2579-2588.

- Cao, Y.-R. et al. 2011. Methylobacterium soli sp. nov. a methanol-utilizing bacterium isolated from the forest soil. Antonie Leeuwenhoek 99:629-634.
- Doetsch, R. 1981. Determinative methods of light microscopy. Manual of methods for general bacteriology. pp. 21-33.
- Edgar, R.C. 2004. MUSCLE: multiple sequence alignment with high accuracy and high throughput. Nucleic Acids Res. 32:1792-1797.
- Felsenstein, J. 1985. Confidence limits on phylogenies: an approach using the bootstrap. Evolution 39:783-791.
- Gallego, V., M.T. García and A. Ventosa. 2005. Methylobacterium hispanicum sp. nov. and Methylobacterium aquaticum sp. nov., isolated from drinking water. International Journal of Systematic and Evolutionary Microbiology 55:281-287.
- Garrity G.M., J.A. Bell and T. Lilburn. 2005a. Family IX. *Methylobacteriaceae* fam. nov. In: Brenner, D.J., N.R. Krieg, J.T. Staley, G.M. Garrity (eds.), Bergey's manual of systematic bacteriology, vol 2, Part C: the alpha- beta-, delta- and *Epsilonproteobacteria*. Springer, New York, pp. 567-571.
- Garrity, G.M., J.A. Bell and T. Lilburn. 2005b. Family I. *Rhodobacteraceae* fam. nov. In: Brenner, D.J., N.R. Krieg, J.T. Staley and G.M. Garrity (eds.), Bergey's manual of systematic bacteriology, vol 2, 2nd edn, The *Proteobacteria*, Part C. The Alpha-, Beta-, Delta-, and *Epsilonproteobacteria*. Springer, New York, pp. 161-228.
- Hall, T.A. BioEdit: a user-friendly biological sequence alignment editor and analysis program for Windows 95/98/ NT. In: Nucleic acids symposium series, 1999. vol 41. [London]: Information Retrieval Ltd., c1979-c2000., pp. 95-98.
- Kanso, S. and B.K. Patel. 2003. *Microvirga subterranea* gen. nov., sp. nov., a moderate thermophile from a deep subsurface Australian thermal aquifer. International Journal of Systematic and Evolutionary Microbiology 53:401-406.
- Kelly, D.P., I.R. McDonald and A.P. Wood. 2014. The Family *Methylobacteriaceae*. In: E. Rosenberg, E.F. DeLong, S. Lory, E. Stackebrandt, and F. Thompson (eds.), The Prokaryotes. Springer, Berlin, Heidelberg.
- Kim, O.-S. *et al.* 2012. Introducing EzTaxon-e: a prokaryotic 16S rRNA gene sequence database with phylotypes that represent uncultured species. International Journal of Systematic and Evolutionary Microbiology 62:716-721.
- Kimura, M. 1983. The neutral theory of molecular evolution. Cambridge University Press.
- Komagata, K., T. Iino and Y. Yamada. 2014. The Family Acetobacteraceae. In: E. Rosenberg, E.F. DeLong, S. Lory, E. Stackebrandt and F. Thompson (eds.), The Prokaryotes. Springer, Berlin, Heidelberg. pp. 3-78.
- Konovalova, A.M., S.O. Shylin and P.V. Rokytko. 2007. Characteristics of carotinoids of methylotrophic bacte-

ria of the *Methylobacterium* genus. Mikrobiol Z (Kiev) 69:35-41 (in Ukrainian).

- Kosako, Y., E. Yabuuchi, T. Naka, N. Fujiwara and K. Kobayashi. 2000. Proposal of Sphingomonadaceae fam. nov., consisting of Sphingomonas Yabuuchi et al. 1990, Erythrobacter Shiba and Shimidu. 1982, Erythromicrobium Yurkov et al. 1994, Porphyrobacter Fuerst et al. 1993, Zymomonas Kluyver and van Niel. 1936, and Sandaracinobacter Yurkov et al. 1997, with the type genus Sphingomonas Yabuuchi et al. 1990. Microbiology and Immunology 44:563-575.
- Saitou, N. and M. Nei. 1987. The neighbor-joining method: a new method for reconstructing phylogenetic trees. Molecular Biology and Evolution 4:406-425.
- Tamura, K., D. Peterson, N. Peterson, G. Stecher, M. Nei and S. Kumar. 2011. MEGA5: molecular evolutionary genetics analysis using maximum likelihood, evolutionary distance, and maximum parsimony methods. Molecular Biology and Evolution 28:2731-2739.
- Weisburg, W.G., S.M. Barns, D.A. Pelletier and D.J. Lane. 1991. 16S ribosomal DNA amplification for phylogenetic study. Journal of Bacteriology 173:697-703.
- Weon, H.Y., S.W. Kwon, J.A. Son, E.H. Jo, S.J. Kim, Y.S. Kim, B.Y. Kim and J.O. Ka. 2010. Description of *Microvirga aerophila* sp. nov., and *Microvirga aerilata* sp. nov., isolated from air, reclassification of *Balneimonas flocculans* Takeda *et al.* 2004 as *Microvirga flocculans*

comb. nov., and emended description of the genus *Microvirga*. International Journal of Systematic and Evolutionary Microbiology 60:2596-2600.

- Willems, A., J. De Ley, M. Gillis and K. Kersters. 1991. Comamonadaceae, a new family encompassing the acidovorans rRNA complex, including Variovorax paradoxus gen. nov., comb. nov., for Alcaligenes paradoxus (Davis) 1969. International Journal of Systematic Bacteriology 41:445-450.
- Wragg, P., L. Randall and A.M. Whatmore. 2014. Comparison of Biolog GEN III MicroStation semi-automated bacterial identification system with matrix-assisted laser desorption ionization-time of flightmass spectrometry and 16S ribosomal RNA gene sequencing for the identification of bacteria of veterinary interest. Journal of Microbiological Methods 105:16-21.
- Xu, P., W.J. Li, S.K. Tang, Y.Q. Zhang, G.Z. Chen, H.H. Chen, L.H. Xu and C.L. Jiang. 2005. *Naxibacter alkalitolerans* gen. nov., sp. nov., a novel member of the family "*Oxalobacteraceae*" isolated from China. International Journal of Systematic and Evolutionary Microbiology 55:1149-1153.

Submitted: March 8, 2018 Revised: April 14, 2018 Accepted: May 14, 2018