

A report of 35 unreported bacterial species in Korea, belonging to the phylum *Firmicutes*

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In an investigation of indigenous prokaryotic species in Korea, a total of 35 bacterial strains assigned to the phylum *Firmicutes* were isolated from diverse habitats including natural and artificial environments. Based on their high 16S rRNA gene sequence similarity (>98.7%) and formation of robust phylogenetic clades with species of validly published names, the isolates were identified as 35 species belonging to the orders *Bacillales* (the family *Bacillaceae*, *Paenibacillaceae*, *Planococcaceae*, and *Staphylococcaceae*) and *Lactobacillales* (*Aerococcaceae*, *Enterococcaceae*, *Lactobacillaceae*, *Leuconostocaceae*, and *Streptococcaceae*). Since these 35 species in Korean environments has not been reported in any official report, we identified them as unrecorded bacterial species and investigated them taxonomically. The newly found unrecorded species belong to 20 species in the order *Bacillales* and 15 species in the order *Lactobacillales*. The morphological, cultural, physiological, and biochemical properties of the isolates were examined and the descriptive information of the 35 previously unrecorded species is provided here.

Keywords: 16S rRNA, *Firmicutes*, unrecorded species

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INTRODUCTION

The phylum *Firmicutes* (Gibbons and Murray, 1978) is a large group consisting of 7 classes (*Bacilli*, *Clostridia*, *Erysipelotrichia*, *Limnochordia*, *Negativicutes*, *Thermolithobacteria*, and *Tissierellia*), and at least 39 families and 274 genera. The taxonomy of this phylum is the subject of numerous recent rearrangements because it is phenotypically and phylogenetically diverse. Traditionally *Firmicutes* has been known as gram-positive with low G+C (mol% G+C content of DNA is <50), but some gram-negative members are also present in the phylum (e.g., *Vellonellaceae*, *Syntrophomonadaceae*). The cell walls are rigid by containing muramic acid, but some con-

tain teichoic acid instead (De Vos *et al.*, 2011). Respiration is aerobic, anaerobic, or facultative anaerobic. Cells are spherical-, straight-, curved-, helical-, or filamentous-shaped. Most members are chemoorganotrophs, but a few are anoxygenic photoheterotrophs (De Vos *et al.*, 2011).

The orders *Bacillales* and *Lactobacillales* are the largest and most popular members of the *Firmicutes*. The order *Bacillales* is generally characterized by the formation of endospores and possession of menaquinone 7, and contains 9 families named *Bacillaceae*, *Alicyclobacillaceae*, *Listeriaceae*, *Paenibacillaceae*, *Pasteuriaceae*, *Planococcaceae*, *Sporolactobacillaceae*, *Staphylococcaceae*, and *Thermoactinomycetaceae* (De Vos *et al.*, 2011). The

order *Lactobacillales* is generally characterized by being facultative anaerobic respiration without forming an endospore, and contains 7 families named *Lactobacillaceae*, *Aerococcaceae*, *Carnobacteriaceae*, *Enterococcaceae*, *Leuconostocaceae*, *Streptococcaceae*, and *Clostridiaceae* (De Vos *et al.*, 2011).

According to the recent microbiome studies, *Firmicutes* has a significant impact on human health and disease, such as obesity, autoimmune disease, allergic diseases, and intestinal disease. For example, in irritable bowel syndrome, the increased *Firmicutes* abundance was positively correlated with the disease severity (Rajilić-Stojanović *et al.*, 2011; Jeffery *et al.*, 2012). In obesity, the abundance of *Firmicutes* increases proportionally because the bacteria leads to decrease in overall metabolic diversity in the human gut (Bäckhed *et al.*, 2004; Ley *et al.*, 2005; Turnbaugh *et al.*, 2009).

During the indigenous bacterial diversity survey performed by National Institute of Biological Resources in Korea, numerous bacterial strains belonging to the phylum *Firmicutes* were isolated from various sources including natural and artificial environments, as well as animals and human. As a result of preliminary phylogenetic analysis using partial 16S rRNA gene sequences, 35 of them were identified to belong to the orders *Bacillales* and *Lactobacillales* with a possibility to be recognized as unrecorded bacterial species. Thus, the taxonomy of the new isolates was investigated and the presence of 35 indigenous species in Korea is reported here.

MATERIALS AND METHODS

As sources of bacteria, samples of lichen, sea salt, sea water, sea sand, soil, moss, mouse gut, mine drainage, human sputum, human feces, and wood were collected from natural and artificial environments in Republic of Korea (Table 1). Bacterial culture media were used for standard dilution plating technique, including Reasoner's 2A (R2A; BD), Marine Agar 2216 (MA; BD), Mueller Hinton Agar (MH; BD), Luria-Bertani Agar (LB; BD), Glucose Yeast Extract Agar (GYE; BD), Anaerobe Basal Agar (AB; Oxoid), Tryptic Soy Yeast Agar (TSY; BD), and Lactobacilli MRS Agar (MRS; BD). The spread plates were incubated at 25–37°C for 1–7 days and subjected to single colony isolation. The pure culture of bacterial isolates were preserved at –80°C as a suspension in liquid culture media containing 10–20% glycerol (w/v) and also preserved by lyophilization. The 35 bacterial species designations, isolation sources, culture media, and incubation conditions are summarized in Table 1.

The 16S rRNA gene was amplified and sequenced using standard PCR and Sanger sequencing methods with universal primers 27F and 1492R. The determined sequences

were compared with sequences of validly published type strains using the EzBioCloud server (Yoon *et al.*, 2017). Based on pairwise sequence similarity, an isolate showing 98.7% or higher similarity to any type strain of the phylum *Firmicutes*, but whose presence has not been reported in Korea, was identified as an unrecorded *Firmicutes* species. The sequences were aligned using EzEditor (Jeon *et al.*, 2014) and phylogenetic trees were reconstructed using maximum-likelihood and neighbor-joining methods implemented in MEGA7 (Kumar *et al.*, 2016).

Cellular morphology and cell size were examined by either transmission electron microscopy or scanning electron microscopy. Gram staining was performed using a Gram-staining kit. Biochemical characteristics were tested by using API 20NE or API 20A kits (bioMérieux) according to the manufacturer's instructions. Oxidase activity was evaluated using 1% tetramethyl-*p*-phenylenediamine (bioMérieux).

RESULTS AND DISCUSSION

Thirty-five *Firmicutes* strains were isolated from various environmental habitats and identified as unrecorded bacterial species by having 16s rRNA gene sequence similarity 98.7% or higher but never been reported in Korea. The similarity-based identification was also supported by phylogenetic trees (Fig. 1). Each isolate formed a monophyletic clade with the type strain of identified bacterial species, confirming the proper assignment of the isolate to the species with published names. The tree topologies of neighbor-joining and maximum likelihood trees were almost identical showing the close relationship of the isolates and valid type strains of the family *Firmicutes*. The taxonomic affiliations of the isolates are summarized in Table 1.

The 35 isolates belonged to 2 orders of the *Firmicutes*; 20 isolates to the order *Bacillales*, 15 isolates to the order *Lactobacillales*. In the order *Bacillales*, the 20 isolates further grouped into 4 bacterial families; 5 isolates (HMF5940, GA079, 18JY44-4, LPB0245, and CAU 1495) of the family *Bacillaceae*; 8 isolates (18S2L7, CAU 1503, HMF7158, 18H7A6, MMS18-CY120, HMF7562, GY1, and 18JY2-4) of the family *Paenibacillaceae*; 5 isolates (CAU 1497, CAU 1499, 18N1A7, CAU 1496, and 18H3T14) of the family *Planococcaceae*; 2 isolates (LPB0217 and LPB0242) of the family *Staphylococcaceae*. In the order *Lactobacillales*, the 15 isolates further grouped into 5 bacterial families; 1 isolate (LPB0228) of the family *Aerococcaceae*; 2 isolates (CAU 1515 and LPB0216) of the family *Enterococcaceae*; 4 isolates (CAU 1514, LPB0236, LPB0237, and LPB0238) of the family *Lactobacillaceae*; 3 isolates (LPB0215, LPB0235, and LPB0239) of the family *Leuconostocaceae*; 5 iso-

Table 1. Strain information and taxonomic affiliation of the 35 unrecorded *Firmicutes* species.

Order	Family	Strain designation	NIBR ID	Most closely related species	16S rRNA gene sequence similarity (%)	Isolation source (GPS)	Medium	Incubation condition
<i>Bacillales</i>	<i>Bacillaceae</i>	HMF5940	NIBRBAC000502507	<i>Anaerobacillus macyae</i>	98.9	Sea salt (N36°44'57.6"; E126°18'22.7")	MA	30°C, 3 days
		GA079	NIBRBAC000502569	<i>Bacillus cucumis</i>	99.2	Soil (N36°21'22.5"; E127°19'52.8")	R2A	30°C, 3 days
		181Y44-4	NIBRBAC000502331	<i>Bacillus psychrosaccharolyticus</i>	99.7	Soil (unknown)	R2A	25°C, 4 days
		LPB0245	NIBRBAC000502432	<i>Bacillus solisilvae</i>	99.4	Soil (N37°29'47.44"; E130°53'30.8")	R2A	25°C, 3 days
		CAU 1495	NIBRBAC000502365	<i>Halobacillus dabaenensis</i>	99.1	Soil (N37°32'03.1"; E126°24'42.9")	MA	30°C, 1–2 days
		18S2L7	NIBRBAC000502535	<i>Paenibacillus anaericanus</i>	99.2	Soil (N37°0'22.96"; E127°59'2.4")	MH	30°C, 2 days
		CAU 1503	NIBRBAC000502370	<i>Paenibacillus lautus</i>	99.2	Sea water (N33°25'22.0"; E126°55'47.1")	MA	30°C, 2–3 days
		HMF7158	NIBRBAC000502511	<i>Paenibacillus methanolicus</i>	98.7	Moss (N37°20'08.9"; E127°15'41.2")	R2A	30°C, 3 days
		18H7A6	NIBRBAC000502542	<i>Paenibacillus polysaccharolyticus</i>	99.6	Soil (N37°39'6.73"; E126°43'17.93")	MH	30°C, 2 days
		MMS18-CY120	NIBRBAC000502557	<i>Paenibacillus salinicani</i>	98.8	Soil (N36°24'49.2"; E126°47'57.3")	R2A	30°C, 3 days
<i>Bacillales</i>	<i>Planococcaceae</i>	HMF7562	NIBRBAC000502516	<i>Paenibacillus shirakamiensis</i>	99.9	Wood (N36°13'07.4"; E127°20'24.6")	R2A	30°C, 3 days
		GY1	NIBRBAC000502403	<i>Paenibacillus silage</i>	99.6	Lichen (N33°32'59"; E126°45'26")	LB	25°C, 3 days
		181Y2-4	NIBRBAC000502333	<i>Paenibacillus tibetensis</i>	99.6	Soil (unknown)	R2A	25°C, 4 days
		CAU 1497	NIBRBAC000502362	<i>Lysinibacillus contaminans</i>	99.2	Mine drainage (N37°47'37.5"; E127°19'20.8")	MA	30°C, 1–2 days
		CAU 1499	NIBRBAC000502361	<i>Psychrobacillus lasitcapitis</i>	99.0	Sea sand (N35°09'32.6"; E129°09'44.2")	GYE	30°C, 2–3 days
		18N1A7	NIBRBAC000502551	<i>Psychrobacillus psychrodurans</i>	99.7	Soil (N38°5'14.92"; E127°44.62")	MH	30°C, 2 days
		CAU 1496	NIBRBAC000502369	<i>Rummelibacillus pycnus</i>	99.2	Mine drainage (N37°47'37.5"; E127°19'20.8")	GYE	30°C, 1–2 days
		18H3T14	NIBRBAC000502547	<i>Solibacillus istrionensis</i>	99.8	Soil (N37°32'02.4"; E127°01'58.2")	MH	30°C, 2 days
		LPB0217	NIBRBAC000502414	<i>Staphylococcus argenteus</i>	99.9	Mouse gut (N36°23'57.4"; E127°23'39.53")	AB	30°C, 3 days
		LPB0242	NIBRBAC000502440	<i>Staphylococcus lentus</i>	99.9	Human sputum (N37°34'42.78"; E126°59'59.21")	TSY	37°C, 3 days

Table 1. Continued.

Order	Family	Strain designation	NIBR ID	Most closely related species	16S rRNA gene sequence similarity (%)	Isolation source (GPS)	Medium	Incubation condition
<i>Aerococcaceae</i>		LPB0228	NIBRBAC000502439	<i>Gramulicatella adiacens</i>	99.3	Human sputum (N37°34'42.78"; E126°59'59.21")	TSY	37°C, 3 days
	<i>Enterococcaceae</i>	CAU 1515	NIBRBAC000502382	<i>Enterococcus avium</i>	99.8	Human feces (N37°30'20.1"; E126°57'32.6")	MRS	37°C, 1–2 days
		LPB0216	NIBRBAC000502413	<i>Enterococcus lactis</i>	99.6	Mouse gut (N36°23'57.4"; E127°23'39.53")	AB	30°C, 3 days
<i>Lactobacillaceae</i>		CAU 1514	NIBRBAC000502381	<i>Lactobacillus paracasei</i> subsp. <i>tolerans</i>	100	Human feces (N37°30'20.1"; E126°57'32.6")	MRS	37°C, 1–2 days
		LPB0238	NIBRBAC000502429	<i>Lactobacillus murinus</i>	100	Mouse gut (N36°19'57.54"; E127°27'40.82")	MRS	30°C, 3 days
		LPB0236	NIBRBAC000502427	<i>Lactobacillus ruminis</i>	99.9	Mouse gut (N36°23'57.4"; E127°23'39.53")	MRS	30°C, 3 days
		LPB0237	NIBRBAC000502428	<i>Pediococcus acidilactici</i>	99.9	Mouse gut (N36°23'57.4"; E127°23'39.53")	MRS	30°C, 3 days
<i>Lactobacillales</i>		LPB0215	NIBRBAC000502437	<i>Weissella cibaria</i>	99.9	Mouse gut (N36°23'57.4"; E127°23'39.53")	MRS	30°C, 3 days
	<i>Leuconostocaceae</i>	LPB0235	NIBRBAC000502426	<i>Weissella confuse</i>	100	Mouse gut (N36°19'57.54"; E127°27'40.82")	MRS	30°C, 3 days
		LPB0239	NIBRBAC000502430	<i>Weissella paramesenteroides</i>	100	Mouse gut (N36°16'57.54"; E127°27'40.82")	MRS	30°C, 3 days
<i>Streptococcaceae</i>		LPB0222	NIBRBAC000502419	<i>Streptococcus anginosus</i> subsp. <i>whiteyi</i>	99.8	Human sputum (N37°34'42.78"; E126°59'59.21")	TSY	37°C, 3 days
		LPB0219	NIBRBAC000502416	<i>Streptococcus cristatus</i>	99.3	Human sputum (N37°34'42.78"; E126°59'59.21")	TSY	37°C, 3 days
		LPB0221	NIBRBAC000502418	<i>Streptococcus gordonii</i>	99.8	Human sputum (N37°34'42.78"; E126°59'59.21")	TSY	37°C, 3 days
		LPB0225	NIBRBAC000502421	<i>Streptococcus mutans</i>	99.6	Human sputum (N37°34'42.78"; E126°59'59.21")	TSY	37°C, 3 days
		LPB0227	NIBRBAC000502422	<i>Streptococcus sanguinis</i>	99.9	Human sputum (N37°34'42.78"; E126°59'59.21")	TSY	37°C, 3 days

Abbreviations: MA, Marine agar; MH, Mueller Hinton agar; GYE, Glucose Yeast Extract agar; MRS, Lactobacilli MRS agar; TSY, Trypticase Soy Yeast agar; L.B, Luria-Bertani agar; AB, Anaerobe basal agar.

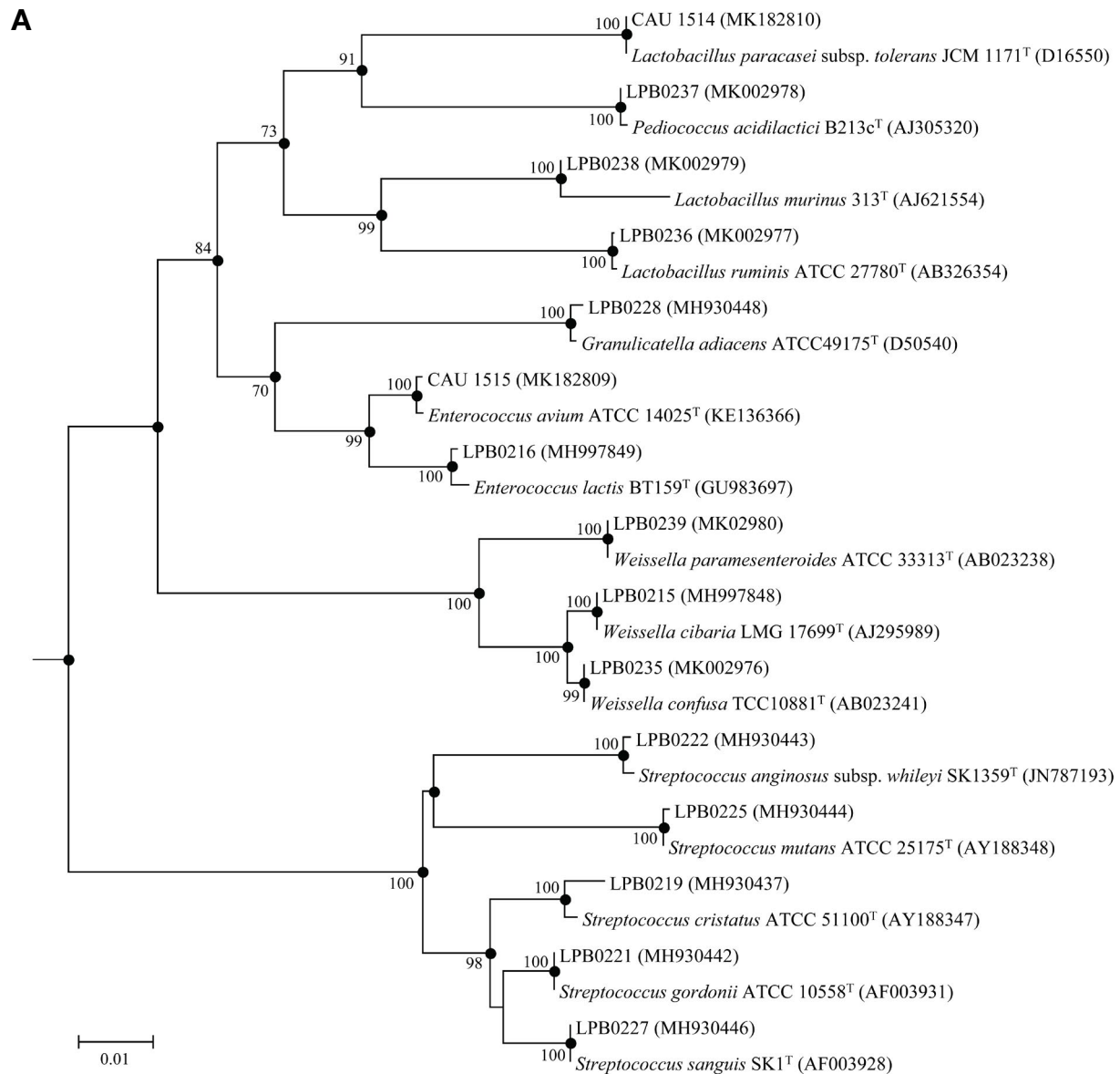


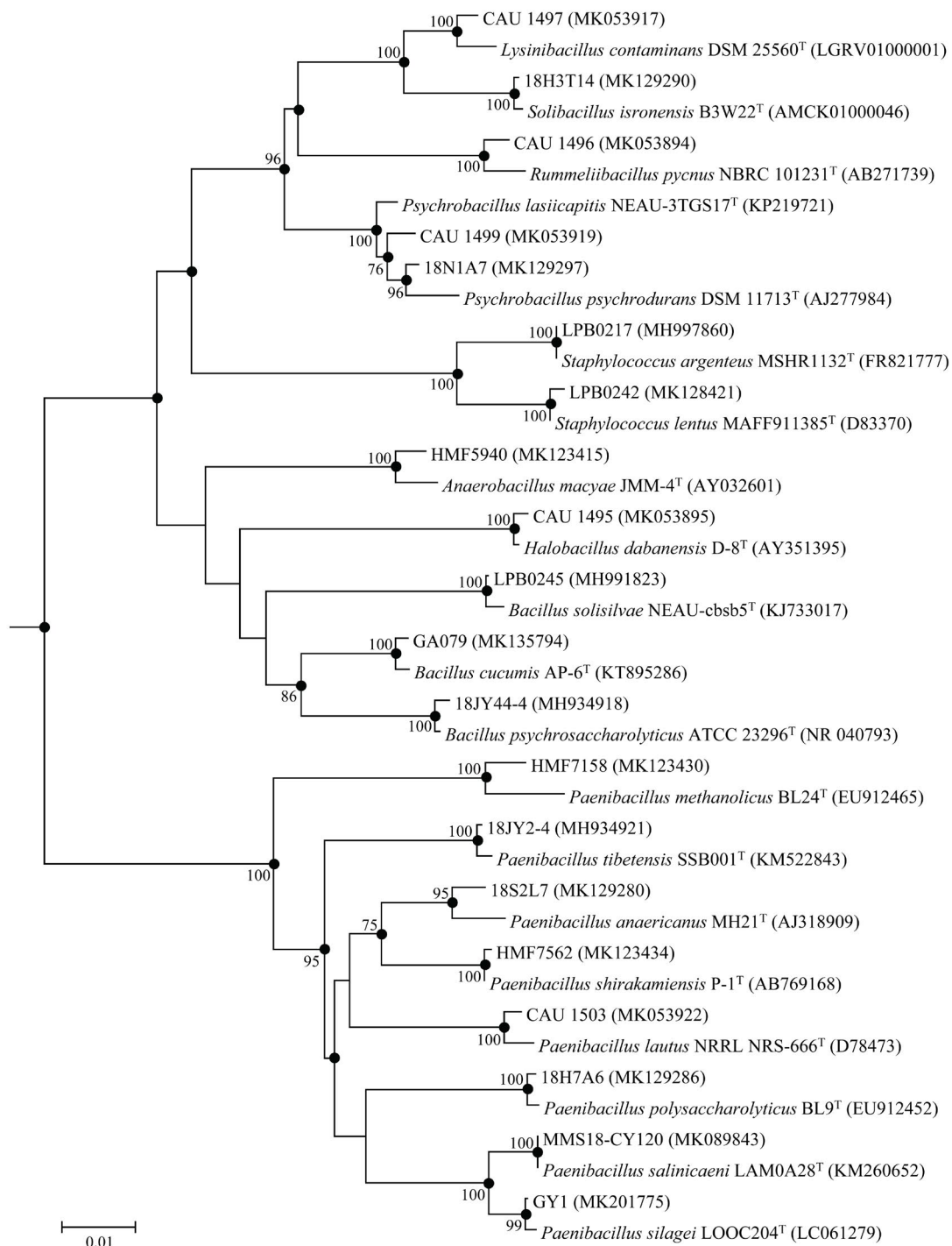
Fig. 1. Neighbor-joining tree based on 16S rRNA gene sequences showing the phylogenetic relationships between the strains isolated in this study and their relatives of (A) the order *Bacillales* and (B) the order *Lactobacillales*. Filled circles indicate that the corresponding nodes (groupings) were also recovered in the maximum likelihood tree. Bootstrap values (>70%) are shown above nodes. *Escherichia coli* ATCC 11775^T (X80725) was used as an outgroup.

lates (LPB0219, LPB0221, LPB0222, LPB0225, and LPB0227) of the family *Streptococcaceae*.

At the genus level, 9 genera were found in the order *Bacillales*; *Anaerobacillus* (1 species), *Bacillus* (3 species), *Halobacillus* (1 species), *Paenibacillus* (8 species), *Lysinibacillus* (1 species), *Psychrobacillus* (2 species), *Rummeliibacillus* (1 species), *Solibacillus* (1 species), and *Staphylococcus* (2 species). Six genera were found in the order *Lactobacillales*; *Enterococcus* (2 species), *Granulicatella* (1 species), *Lactobacillus* (3 species), *Pediococcus* (1 species), *Streptococcus* (5 species), and *Weissella* (3

species).

These unrecorded species showed diverse cellular morphologies including rods, short rods, cocci, or ovoid (Fig. 2). The colony colors were white, cream, yellow, orange, or red. Most species were positive at Gram-staining, but a minor portion was stained Gram-negative. All species reported here grew aerobically, but some species also grew at anaerobic condition indicating their facultative anaerobic respiration. The enzymatic and physiological properties examined by API kits were also variable depending on species. The features of oxidase, catalase, nitrate re-

B**Fig. 1.** Continued.

duction, indole production, glucose fermentation, arginine dihydrolase, urease, β -galactosidase, gelatin hydrolysis, esculin hydrolysis, and utilization or acidification of carbon sources of each species are described in the proto-logues.

From the natural and artificial environments in Korea,

35 species of 15 genera belonging to the phylum *Firmicutes* were found and examined taxonomically. The presence of the above mentioned *Firmicutes* species reported in overseas, but not in Korea. Thus, the following 35 species are reported as unrecorded species in Korea and their phenotypic properties are given.

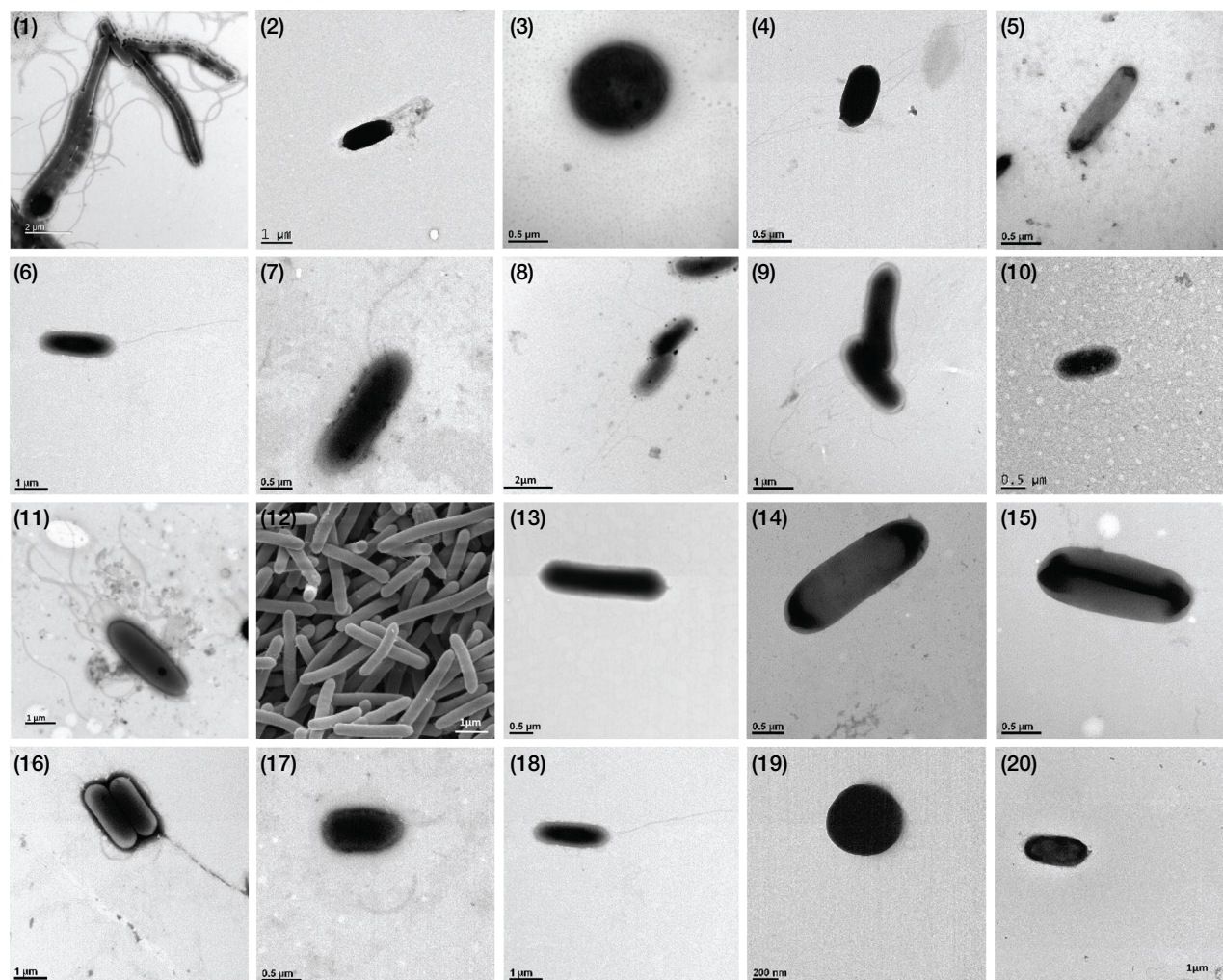
A

Fig. 2. Transmission electron micrographs or scanning electron micrographs of cells of (A) the order *Bacillales* and (B) the order *Lactobacillales*. Strains: 1, HMF5940; 2, GA079; 3, 18JY44-4; 4, LPB0245; 5, CAU 1495; 6, 18S2L7; 7, CAU 1503; 8, HMF7158; 9, 18H7A6; 10, MS18-CY120; 11, HMF7562; 12, GY1; 13, 18JY2-4; 14, CAU 1497; 15, CAU 1499; 16, 18N1A7; 17, CAU1496; 18, 18H3T14; 19, LPB0217; 20, LPB0242; 21, CAU 1515; 22, CAU 1514; 23, LPB0228; 24, LPB0216; 25, LPB0238; 26, LPB0236; 27, LPB0237; 28, LPB0215; 29, LPB0235; 30, LPB0239; 31, LPB0222; 32, LPB0219; 33, LPB0221; 34, LPB0225; 35, LPB0227.

Description of *Anaerobacillus macyae* HMF5940

Cells are Gram-staining-positive, non-flagellated, rod-shaped, and aerobic. Colonies are circular, smooth, convex, and white colored after 3 days on MA at 30°C. Positive for oxidase. Hydrolyses esculin, but not gelatin. Reduces nitrate to nitrite. Cannot produce acid from glucose. Does not produce indole. Possesses activity of β -galactosidase, but not arginine dihydrolase or urease. Does not assimilate glucose, arabinose, mannose, mannitol, maltose, *N*-acetyl-glucosamine, potassium gluconate, capric acid, adipic acid, malic acid, trisodium citrate, or phenylacetic acid. Strain HMF5940 (=NIBRBAC000502507) was isolated from sea salt, Taean, Korea. The GenBank accession number for the 16S rRNA gene sequence is

MK123415.

Description of *Bacillus cucumis* GA079

Cells are Gram-staining-negative, non-flagellated, rod-shaped, and aerobic. Colonies are circular, glistening, convex, and cream colored after 3 days on R2A at 30°C. Positive for oxidase. Hydrolyses esculin, but not gelatin. Reduces nitrate to nitrite. Cannot produce acid from glucose. Does not produce indole. Possesses activity of β -galactosidase, but not arginine dihydrolase or urease. Assimilates glucose, arabinose, mannitol, maltose, *N*-acetyl-glucosamine, potassium gluconate, but not mannose, capric acid, adipic acid, malic acid, trisodium citrate, or phenylacetic acid. Strain GA079 (=NIBRBAC000502569)

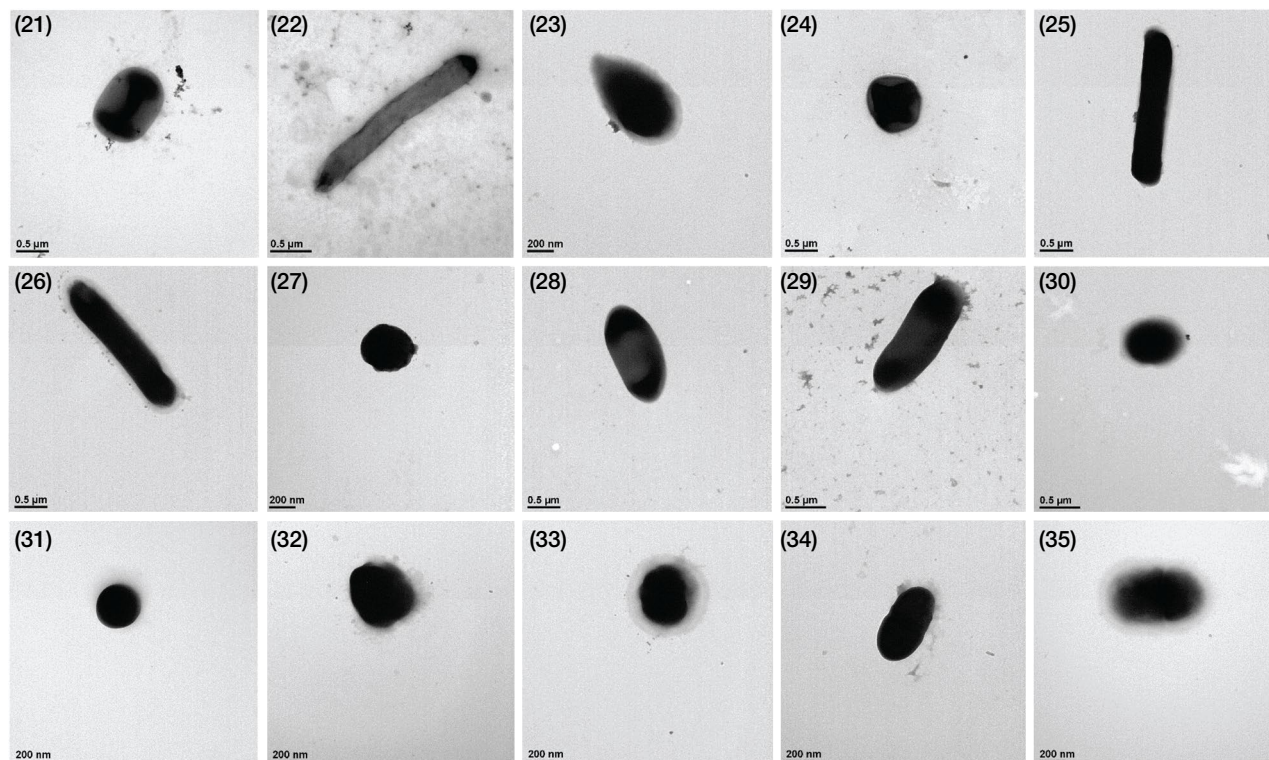
B

Fig. 2. Continued.

was isolated from soil, Daejeon, Korea. The GenBank accession number for the 16S rRNA gene sequence is MK135794.

Description of *Bacillus psychrosaccharolyticus* 18JY44-4

Cells are Gram-staining-positive, non-flagellated, coccoid-shaped, and aerobic. Colonies are circular, convex, smooth, and yellow colored after 4 days on R2A at 25°C. Hydrolyses esculin and gelatin. Reduces nitrate to nitrite. Cannot produce acid from glucose. Does not produce indole. Possesses activity of β -galactosidase, but not arginine dihydrolase or urease. Assimilates glucose, arabinose, mannose, mannitol, maltose, *N*-acetyl-glucosamine, potassium gluconate, adipic acid, but not capric acid, malic acid, trisodium citrate, or phenylacetic acid. Strain 18JY44-4 (= NIBRBAC000502331) was isolated from soil, Ulsan, Korea. The GenBank accession number for the 16S rRNA gene sequence is MH934918.

Description of *Bacillus solisilvae* LPB0245

Cells are Gram-staining-positive, non-flagellated, rod-shaped, and aerobic. Colonies are filamentous, rough, convex, and white colored after 3 days on R2A at 25°C.

Negative for oxidase. Does not hydrolyze esculin and gelatin. Does not reduce nitrate to nitrite. Cannot produce acid from glucose. Does not produce indole. Does not possess activity of β -galactosidase, arginine dihydrolase, or urease. Weakly assimilates glucose, mannose, mannitol, but not arabinose, maltose, *N*-acetyl-glucosamine, potassium gluconate, capric acid, adipic acid, malic acid, trisodium citrate, or phenylacetic acid. Strain LPB0245 (= NIBRBAC000502432) was isolated from soil, Ulsan, Korea. The GenBank accession number for the 16S rRNA gene sequence is MH991823.

Description of *Enterococcus avium* CAU 1515

Cells are Gram-staining-positive, non-flagellated, ovoid-shaped, and aerobic. Colonies are circular, convex, smooth, shiny, opaque, and white colored after 1–2 days on MRS at 37°C. Hydrolyses esculin, but not gelatin. Does not reduce nitrate to nitrite. Can produce acid from glucose. Does not produce indole. Does not possess activity of β -galactosidase, arginine dihydrolase, or urease. Assimilates glucose, mannose, mannitol, *N*-acetyl-glucosamine (weakly), maltose (weakly), and potassium gluconate (weakly), but not arabinose, capric acid, adipic acid, malic acid, trisodium citrate, or phenylacetic acid. Strain

CAU 1515 (=NIBRBAC000502382) was isolated from human feces, Seoul, Korea. The GenBank accession number for the 16S rRNA gene sequence is MK182809.

Description of *Enterococcus lactis* LPB0216

Cells are Gram-staining-positive, non-flagellated, coccoid-shaped, and facultative anaerobic. Colonies are circular, entire, smooth, convex, and white colored after 3 days on AB at 30°C. Negative for oxidase. Does not produce indole. Does not possess activity of urease. Hydrolyses esculin, but not gelatin. Acidifies glucose, mannitol, lactose, saccharose, maltose, salicin, arabinose, cellobiose, raffinose, rhamnose (weakly), and trehalose (weakly), but not xylose, glycerol, mannose, melezitose, or sorbitol. Strain LPB0216 (=NIBRBAC000502413) was isolated from mouse gut, Korea Institute of Oriental Medicine, Korea. The GenBank accession number for the 16S rRNA gene sequence is MH997849.

Description of *Granulicatella adiacens* LPB0228

Cells are Gram-staining-positive, non-flagellated, coccoid-shaped, and aerobic. Colonies are circular, entire, smooth, convex, and cream colored after 3 days on TSY at 37°C. Negative for oxidase. Weakly hydrolyses esculin, but not gelatin. Does not reduce nitrate to nitrite. Cannot produce acid from glucose. Does not produce indole. Does not possess activity of β -galactosidase, arginine dihydrolase, or urease. Does not assimilate glucose, arabinose, mannose, mannitol, maltose, *N*-acetyl-glucosamine, potassium gluconate, capric acid, adipic acid, malic acid, trisodium citrate, or phenylacetic acid. Strain LPB0228 (=NIBRBAC000502439) was isolated from human sputum, Seoul National University Hospital, Korea. The GenBank accession number for the 16S rRNA gene sequence is MH930448.

Description of *Halobacillus dabanensis* CAU 1495

Cells are Gram-staining-positive, non-flagellated, rod-shaped, and aerobic. Colonies are circular, convex, smooth, shiny, opaque, and pale orange colored after 1–2 days on MA at 30°C. Hydrolyses gelatin, but not esculin. Does not reduce nitrate to nitrite. Cannot produce acid from glucose. Does not produce indole. Does not possess activity of β -galactosidase, arginine dihydrolase, or urease. Does not assimilate glucose, arabinose, mannose, mannitol, maltose, *N*-acetyl-glucosamine, potassium gluconate, capric acid, adipic acid, malic acid, trisodium citrate, or phenylacetic acid. Strain CAU 1495 (=NIBRBAC000502365) was isolated from human sputum, Seoul National University Hospital, Korea. The GenBank accession number for the 16S rRNA gene sequence is MK053895.

Description of *Lactobacillus murinus* LPB0238

Cells are Gram-staining-positive, non-flagellated, rod-shaped, and facultative anaerobic. Colonies are irregular, undulate, wrinkled, flat, and creamy white colored after 3 days on MRS at 30°C. Negative for oxidase. Produces indole. Does not possess activity of urease. Hydrolyses esculin, but not gelatin. Weakly acidifies glucose, lactose, saccharose, maltose, xylose, arabinose, cellobiose, mannose, melezitose, raffinose, rhamnose, and trehalose, but not mannitol, salicin, glycerol, or sorbitol. Strain LPB0238 (=NIBRBAC000502429) was isolated from mouse gut, Daejeon University, Korea. The GenBank accession number for the 16S rRNA gene sequence is MK002979.

Description of *Lactobacillus paracasei* subsp. *tolerans* CAU 1514

Cells are Gram-staining-positive, non-flagellated, rod-shaped, and aerobic. Colonies are circular, raised, smooth, shiny, opaque, and white colored after 1–2 days on MRS at 37°C. Hydrolyses esculin, but not gelatin. Does not reduce nitrate to nitrite. Produces acid from glucose. Does not produce indole. Possesses activity of β -galactosidase (weakly), but not arginine dihydrolase or urease. Assimilates glucose, arabinose, mannose, mannitol, maltose, *N*-acetyl-glucosamine (weakly), potassium gluconate, capric acid, adipic acid, malic acid (weakly), trisodium citrate, and phenylacetic acid. Strain CAU 1514 (=NIBRBAC000502381) was isolated from human feces, Seoul, Korea. The GenBank accession number for the 16S rRNA gene sequence is MK182810.

Description of *Lactobacillus ruminis* LPB0236

Cells are Gram-staining-positive, non-flagellated, rod-shaped, and facultative anaerobic. Colonies are circular, entire, smooth, convex, and white colored after 3 days on MRS at 30°C. Negative for oxidase. Does not produce indole. Does not possess activity of urease. Hydrolyses esculin, but not gelatin. Acidifies glucose, mannitol, lactose, saccharose, maltose, salicin, arabinose (weakly), cellobiose, mannose, and raffinose, but not xylose, glycerol, melezitose, sorbitol, rhamnose, or trehalose. Strain LPB0236 (=NIBRBAC000502427) was isolated from mouse gut, Korea Institute of Oriental Medicine, Korea. The GenBank accession number for the 16S rRNA gene sequence is MK002977.

Description of *Lysinibacillus contaminans* CAU 1497

Cells are Gram-staining-positive, non-flagellated, rod-shaped, and aerobic. Colonies are circular, convex, smooth, shiny, opaque, and cream colored after 1–2 days

on MA at 30°C. Does not hydrolyze esculin and gelatin. Does not reduce nitrate to nitrite. Cannot produce acid from glucose. Produce indole. Possesses activity of arginine dihydrolase and urease (weakly), but not β -galactosidase. Assimilates trisodium citrate, but not glucose, arabinose, mannose, mannitol, maltose, *N*-acetyl-glucosamine, potassium gluconate, capric acid, adipic acid, malic acid, or phenylacetic acid. Strain CAU 1497 (=NIBRBAC000502362) was isolated from mouse mine drainage, Gapyeong-gun, Korea. The GenBank accession number for the 16S rRNA gene sequence is MK053917.

Description of *Paenibacillus anaericanus* 18S2L7

Cells are Gram-staining-positive, flagellated, rod-shaped, and facultative anaerobic. Colonies are circular, convex, smooth, and white colored after 2 days on MH at 30°C. Negative for oxidase. Hydrolyses esculin, but not gelatin. Reduces nitrate to nitrite. Cannot produce acid from glucose. Does not produce indole. Possesses activity of β -galactosidase, but not arginine dihydrolase or urease. Does not assimilate glucose, arabinose, mannose, mannitol, maltose, *N*-acetyl-glucosamine, potassium gluconate, capric acid, adipic acid, malic acid, trisodium citrate, or phenylacetic acid. Strain 18S2L7 (=NIBRBAC000502535) was isolated from soil, Chungju, Korea. The GenBank accession number for the 16S rRNA gene sequence is MK129280.

Description of *Paenibacillus lautus* CAU 1503

Cells are Gram-staining-positive, non-flagellated, rod-shaped, and aerobic. Colonies are circular, convex, smooth, shiny, opaque, and cream colored after 2–3 days on MA at 30°C. Does not hydrolyze esculin and gelatin. Does not reduce nitrate to nitrite. Cannot produce acid from glucose. Does not produce indole. Possesses activity of urease (weakly), but not arginine dihydrolase or β -galactosidase. Does not assimilate glucose, arabinose, mannose, mannitol, maltose, *N*-acetyl-glucosamine, potassium gluconate, capric acid, adipic acid, malic acid, trisodium citrate, or phenylacetic acid. Strain CAU 1503 (=NIBRBAC000502370) was isolated from mouse sea water, Seopjikoji, Jeju-do, Korea. The GenBank accession number for the 16S rRNA gene sequence is MK053922.

Description of *Paenibacillus methanolicus* HMF7158

Cells are Gram-staining-positive, non-flagellated, rod-shaped, and aerobic. Colonies are circular, convex, smooth, and white colored after 3 days on R2A at 30°C. Positive for oxidase. Does not hydrolyze esculin and gelatin. Does not reduce nitrate to nitrite. Cannot produce acid from glucose. Does not produce indole. Does not possess activity of arginine dihydrolase, β -galactosidase, or ure-

ase. Does not assimilate glucose, arabinose, mannose, mannitol, maltose, *N*-acetyl-glucosamine, potassium gluconate, capric acid, adipic acid, malic acid, trisodium citrate, or phenylacetic acid. Strain HMF7158 (=NIBRBAC000502511) was isolated from moss, Yongin, Korea. The GenBank accession number for the 16S rRNA gene sequence is MK123430.

Description of *Paenibacillus polysaccharolyticus* 18H7A6

Cells are Gram-staining-positive, non-flagellated, rod-shaped, and facultative anaerobic. Colonies are circular, flat, smooth, undulate, and white colored after 2 days on MH at 30°C. Positive for oxidase. Hydrolyses esculin and gelatin. Reduces nitrate to nitrite. Cannot produce acid from glucose. Does not produce indole. Possesses activity of β -galactosidase, but not arginine dihydrolase or urease. Assimilates glucose, arabinose, mannitol, maltose, but not mannose, *N*-acetyl-glucosamine, potassium gluconate, capric acid, adipic acid, malic acid, trisodium citrate, or phenylacetic acid. Strain 18H7A6 (=NIBRBAC000502542) was isolated from soil, Gimpo, Korea. The GenBank accession number for the 16S rRNA gene sequence is MK129286.

Description of *Paenibacillus salinicaenis* MMS18-CY120

Cells are Gram-staining-variable, non-flagellated, rod-shaped, and aerobic. Colonies are circular, convex, smooth, and cream colored after 3 days on R2A at 30°C. Negative for oxidase. Hydrolyses esculin and gelatin. Does not reduce nitrate to nitrite. Cannot produce acid from glucose. Does not produce indole. Possesses activity of β -galactosidase and urease, but not arginine dihydrolase. Assimilates glucose, arabinose, mannose, mannitol, maltose, *N*-acetyl-glucosamine, potassium gluconate, malic acid, trisodium citrate and phenylacetic acid, but not capric or and adipic acid. Strain MMS18-CY120 (=NIBRBAC000502557) was isolated from soil, Cheongyand-gun, Korea. The GenBank accession number for the 16S rRNA gene sequence is MK089843.

Description of *Paenibacillus salinicaenis* HMF7562

Cells are Gram-staining-positive, flagellated, rod-shaped, and aerobic. Colonies are circular, convex, smooth, and white colored after 3 days on R2A at 30°C. Positive for oxidase. Hydrolyses esculin, but not gelatin. Does not reduce nitrate to nitrite. Cannot produce acid from glucose. Does not produce indole. Possesses activity of β -galactosidase, but not arginine dihydrolase or urease. Assimilates glucose, maltose, *N*-acetyl-glucosamine, but not arabinose, mannose, mannitol, potassium

gluconate, capric acid, adipic acid, malic acid, trisodium citrate, or phenylacetic acid. Strain HMF7562 (=NIBRBAC000502516) was isolated from wood, Daejeon, Korea. The GenBank accession number for the 16S rRNA gene sequence is MK123434.

Description of *Paenibacillus silageis* GY1

Cells are Gram-staining-negative, non-flagellated, rod-shaped, and aerobic. Colonies are circular, convex, smooth, and white colored after 3 days on LB at 25°C. Positive for oxidase. Hydrolyses esculin, but not gelatin. Reduces nitrate to nitrite. Cannot produce acid from glucose. Does not produce indole. Possesses activity of β -galactosidase, but not arginine dihydrolase or urease. Assimilates arabinose, mannose, mannitol and maltose, but not glucose, *N*-acetyl-glucosamine, potassium gluconate, capric acid, adipic acid, malic acid, trisodium citrate, or phenylacetic acid. Strain GY1 (=NIBRBAC000502403) was isolated from lichen, Gimnyeong, Jeju-do, Korea. The GenBank accession number for the 16S rRNA gene sequence is MK201775.

Description of *Paenibacillus tibetensis* 18JY2-4

Cells are Gram-staining-negative, non-flagellated, rod-shaped, and aerobic. Colonies are circular, convex, smooth, and yellow colored after 4 days on R2A at 25°C. Hydrolyses esculin, but not gelatin. Does not reduce nitrate to nitrite. Cannot produce acid from glucose. Does not produce indole. Does not possess activity of β -galactosidase, arginine dihydrolase, or urease. Assimilates glucose, *N*-acetyl-glucosamine, and maltose, but not arabinose, mannose, mannitol, potassium gluconate, capric acid, adipic acid, malic acid, trisodium citrate, or phenylacetic acid. Strain 18JY2-4 (=NIBRBAC000502333) was isolated from soil, Ulleung-gun, Korea. The GenBank accession number for the 16S rRNA gene sequence is MH934921.

Description of *Pediococcus acidilactici* LPB0237

Cells are Gram-staining-positive, non-flagellated, coccoid-shaped, and facultative anaerobic. Colonies are circular, entire, smooth, convex, and creamy white colored after 3 days on MRS at 30°C. Negative for oxidase. Does not produce indole. Does not possess activity of urease. Hydrolyses esculin, but not gelatin. Acidifies glucose, mannitol, lactose, saccharose, maltose, salicin, xylose, arabinose, glycerol (weakly), cellobiose, mannose, raffinose, sorbitol (weakly), rhamnose, and trehalose, but not melezitose. Strain LPB0237 (=NIBRBAC000502428) was isolated from mouse gut, Korea Institute of Oriental Medicine, Korea. The GenBank accession number for the 16S rRNA gene sequence is MK002978.

Description of *Psychrobacillus lasiucapitis* CAU 1499

Cells are Gram-staining-positive, non-flagellated, rod-shaped, and aerobic. Colonies are circular, convex, smooth, shiny, opaque, and yellow colored after 2–3 days on GYE at 30°C. Hydrolyses esculin, but not gelatin. Does not reduce nitrate to nitrite. Cannot produce acid from glucose. Does not produce indole. Does not possess activity of β -galactosidase, arginine dihydrolase, or urease. Assimilates glucose, arabinose (weakly), mannitol (weakly), maltose (weakly), potassium gluconate, capric acid (weakly), adipic acid (weakly), and trisodium citrate, but not mannose, *N*-acetyl-glucosamine, malic acid, or phenylacetic acid. Strain CAU 1499 (=NIBRBAC000502361) was isolated from sea sand, Busan, Korea. The GenBank accession number for the 16S rRNA gene sequence is MK053919.

Description of *Psychrobacillus psychrodurans* 18N1A7

Cells are Gram-staining-positive, flagellated, rod-shaped, and aerobic. Colonies are circular, convex, smooth, and white colored after 2 days on MH at 30°C. Negative for oxidase. Hydrolyses esculin, but not gelatin. Does not reduce nitrate to nitrite. Cannot produce acid from glucose. Does not produce indole. Possesses activity of β -galactosidase, but not arginine dihydrolase or urease. Does not assimilate glucose, arabinose, mannose, mannitol, maltose, *N*-acetyl-glucosamine, potassium gluconate, capric acid, adipic acid, malic acid, trisodium citrate, or phenylacetic acid. Strain 18N1A7 (=NIBRBAC000502551) was isolated from around the river soil, Hwachon-gun, Korea. The GenBank accession number for the 16S rRNA gene sequence is MK129297.

Description of *Rummeliibacillus pycnus* CAU 1496

Cells are Gram-staining-positive, non-flagellated, oval rod-shaped, and aerobic. Colonies are circular, flat, smooth, lobate margin, shiny, and cream colored after 1–2 days on GYE at 30°C. Does not hydrolyze esculin or gelatin. Does not reduce nitrate to nitrite. Cannot produce acid from glucose. Produce indole. Does not possess activity of β -galactosidase, arginine dihydrolase, or urease. Does not assimilate glucose, arabinose, mannose, mannitol, maltose, *N*-acetyl-glucosamine, potassium gluconate, capric acid, adipic acid, malic acid, trisodium citrate, or phenylacetic acid. Strain CAU 1496 (=NIBRBAC000502369) was isolated from mine drainage, Gapyeong-gun, Korea. The GenBank accession number for the 16S rRNA gene sequence is MK053894.

Description of *Solibacillus isronensis* 18H3T14

Cells are Gram-staining-positive, non-flagellated, rod-shaped, and aerobic. Colonies are circular, raised, smooth,

and white colored after 2 days on MH at 30°C. Negative for oxidase. Does not hydrolyze esculin or gelatin. Does not reduce nitrate to nitrite. Cannot produce acid from glucose. Does not produce indole. Does not possess activity of β -galactosidase, arginine dihydrolase, or urease. Assimilates potassium gluconate, but not glucose, arabinose, mannose, mannitol, maltose, *N*-acetyl-glucosamine, capric acid, adipic acid, malic acid, trisodium citrate, or phenylacetic acid. Strain 18H3T14 (=NIBRBAC000502547) was isolated from around the river soil, Seoul, Korea. The GenBank accession number for the 16S rRNA gene sequence is MK129290.

Description of *Staphylococcus argenteus* LPB0217

Cells are Gram-staining-positive, non-flagellated, coccoid-shaped, and facultative anaerobic. Colonies are circular, entire, smooth, convex, and creamy white colored after 3 days on ABat 30°C. Negative for oxidase. Does not produce indole. Possesses activity of urease. Does not hydrolyze esculin or gelatin. Acidifies glucose, lactose, saccharose, maltose, mannose, and trehalose, but not mannitol, salicin, xylose, arabinose, glycerol, cellobiose, melezitose, raffinose, sorbitol, or rhamnose. Strain LPB0217 (=NIBRBAC000502414) was isolated from mouse gut, Korea Institute of Oriental Medicine, Korea. The GenBank accession number for the 16S rRNA gene sequence is MH997860.

Description of *Staphylococcus lentus* LPB0242

Cells are Gram-staining-positive, non-flagellated, coccoid-shaped, and facultative anaerobic. Colonies are circular, entire, smooth, convex, and cream colored after 3 days on TSY at 37°C. Positive for oxidase. Hydrolyses esculin, but not gelatin. Reduces nitrate to nitrite. Cannot produce acid from glucose. Does not produce indole. Possesses activity of β -galactosidase, but not arginine dihydrolase or urease. Does not assimilate glucose, arabinose, mannose, mannitol, maltose, *N*-acetyl-glucosamine, potassium gluconate, capric acid, adipic acid, malic acid, trisodium citrate, or phenylacetic acid. Strain LPB0242 (=NIBRBAC000502440) was isolated from human sputum, Seoul National University Hospital, Korea. The GenBank accession number for the 16S rRNA gene sequence is MK128421.

Description of *Streptococcus anginosus* subsp. *whileyi* LPB0222

Cells are Gram-staining-positive, non-flagellated, coccoid-shaped, and facultative anaerobic. Colonies are circular, entire, smooth, convex, and cream colored after 3 days on TSY at 37°C. Negative for oxidase. Hydrolyses esculin and gelatin. Does not reduce nitrate to nitrite. Cannot

produce acid from glucose. Does not produce indole. Possesses activity of β -galactosidase (weakly) and arginine dihydrolase, but not urease. Does not assimilate glucose, arabinose, mannose, mannitol, maltose, *N*-acetyl-glucosamine, potassium gluconate, capric acid, adipic acid, malic acid, trisodium citrate, or phenylacetic acid. Strain LPB0222 (=NIBRBAC000502419) was isolated from human sputum, Seoul National University Hospital, Korea. The GenBank accession number for the 16S rRNA gene sequence is MH930443.

Description of *Streptococcus cristatus* LPB0219

Cells are Gram-staining-positive, non-flagellated, coccoid-shaped, and facultative anaerobic. Colonies are circular, entire, smooth, convex, and cream colored after 3 days on TSY at 37°C. Negative for oxidase. Hydrolyses esculin (weakly), but not gelatin. Does not reduce nitrate to nitrite. Cannot produce acid from glucose. Does not produce indole. Does not possess activity of β -galactosidase, arginine dihydrolase, or urease. Does not assimilate glucose, arabinose, mannose, mannitol, maltose, *N*-acetyl-glucosamine, potassium gluconate, capric acid, adipic acid, malic acid, trisodium citrate, or phenylacetic acid. Strain LPB0219 (=NIBRBAC000502416) was isolated from human sputum, Seoul National University Hospital, Korea. The GenBank accession number for the 16S rRNA gene sequence is MH930437.

Description of *Streptococcus gordonii* LPB0221

Cells are Gram-staining-positive, non-flagellated, coccoid-shaped, and facultative anaerobic. Colonies are circular, entire, smooth, convex, and cream colored after 3 days on TSY at 37°C. Negative for oxidase. Hydrolyses esculin (weakly), but not gelatin. Does not reduce nitrate to nitrite. Can produce acid from glucose. Does not produce indole. Does not possess activity of β -galactosidase, arginine dihydrolase, or urease. Does not assimilate glucose, arabinose, mannose, mannitol, maltose, *N*-acetyl-glucosamine, potassium gluconate, capric acid, adipic acid, malic acid, trisodium citrate, or phenylacetic acid. Strain LPB0221 (=NIBRBAC000502418) was isolated from human sputum, Seoul National University Hospital, Korea. The GenBank accession number for the 16S rRNA gene sequence is MH930442.

Description of *Streptococcus mutans* LPB0225

Cells are Gram-staining-positive, non-flagellated, coccoid-shaped, and facultative anaerobic. Colonies are circular, entire, smooth, convex, and cream colored after 3 days on TSY at 37°C. Negative for oxidase. Hydrolyses esculin, but not gelatin. Does not reduce nitrate to nitrite. Cannot produce acid from glucose. Does not produce indole.

Does not possess activity of β -galactosidase, arginine dihydrolase, or urease. Does not assimilate glucose, arabinose, mannose, mannitol, maltose, *N*-acetyl-glucosamine, potassium gluconate, capric acid, adipic acid, malic acid, trisodium citrate, or phenylacetic acid. Strain LPB0225 (=NIBRBAC000502421) was isolated from human sputum, Seoul National University Hospital, Korea. The GenBank accession number for the 16S rRNA gene sequence is MF930444.

Description of *Streptococcus sanguinis* LPB0227

Cells are Gram-staining-positive, non-flagellated, cocci-shaped, and facultative anaerobic. Colonies are circular, entire, smooth, convex, and cream colored after 3 days on TSY at 37°C. Negative for oxidase. Hydrolyses esculin (weakly), but not gelatin. Does not reduce nitrate to nitrite. Can produce acid from glucose. Does not produce indole. Does not possess activity of β -galactosidase, arginine dihydrolase, or urease. Does not assimilate glucose, arabinose, mannose, mannitol, maltose, *N*-acetyl-glucosamine, potassium gluconate, capric acid, adipic acid, malic acid, trisodium citrate, or phenylacetic acid. Strain LPB0227 (=NIBRBAC000502422) was isolated from human sputum, Seoul National University Hospital, Korea. The GenBank accession number for the 16S rRNA gene sequence is MH930446.

Description of *Weissella cibaria* LPB0215

Cells are Gram-staining-positive, non-flagellated, short rod-shaped, and facultative anaerobic. Colonies are circular, entire, smooth, convex, and white colored after 3 days on MRS at 30°C. Negative for oxidase. Does not produce indole. Does not possess activity of urease. Hydrolyses esculin, but not gelatin. Acidifies glucose, mannitol, lactose, saccharose, maltose, salicin, xylose, arabinose, cellobiose, mannose, raffinose, rhamnose, and trehalose, but not glycerol, melezitose, or sorbitol. Strain LPB0215 (=NIBRBAC000502437) was isolated from mouse gut, Korea Institute of Oriental Medicine, Korea. The GenBank accession number for the 16S rRNA gene sequence is MH997848.

Description of *Weissella confusa* LPB0235

Cells are Gram-staining-positive, non-flagellated, short rod-shaped, and facultative anaerobic. Colonies are circular, entire, smooth, convex, and white colored after 3 days on MRS at 30°C. Negative for oxidase. Produces indole. Does not possess activity of urease. Hydrolyses esculin, but not gelatin. Acidifies glucose, saccharose, maltose, salicin, xylose, cellobiose, mannose, raffinose (weakly), and trehalose (weakly), but not mannitol, lactose, arabinose, glycerol, melezitose, sorbitol, or rhamnose. Strain

LPB0235 (=NIBRBAC000502426) was isolated from mouse gut, Korea Institute of Oriental Medicine, Korea. The GenBank accession number for the 16S rRNA gene sequence is MK002976.

Description of *Weissella paramesenteroides* LPB0239

Cells are Gram-staining-positive, non-flagellated, coccoid-shaped, and facultative anaerobic. Colonies are circular, entire, smooth, convex, and creamy white colored after 3 days on MRS at 30°C. Negative for oxidase. Does not produce indole. Does not possess activity of urease. Does not hydrolyze esculin and gelatin. Acidifies glucose, lactose (weakly), saccharose, maltose, salicin (weakly), xylose (weakly), arabinose, raffinose and trehalose, but not mannitol, glycerol, cellobiose, mannose, melezitose, sorbitol, or rhamnose. Strain LPB0239 (=NIBRBAC000502430) was isolated from mouse gut, Daejeon University, Korea. The GenBank accession number for the 16S rRNA gene sequence is MK002980.

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