

Report of 20 unrecorded bacterial species in Korea belonging to the phylum *Firmicutes* during surveys in 2020

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During a project aiming to comprehensively investigate indigenous prokaryotic species in Korea, a total of 20 bacterial strains phylogenetically belonging to the the class *Bacilli* of the phylum *Firmicutes* were isolated from various environmental sources such as soil, air, tidal flat, sea water, grain, wetland, breast milk and healthy human urine. Phylogenetic analysis based on 16S rRNA gene sequences revealed that 20 bacterial strains showed the high sequence similarities ($\geq 98.7\%$) to the closest type strains and formed robust phylogenetic clades with closely related species of validly published names in the class *Bacilli* of the phylum *Firmicutes*. In the present study, we report 20 species of 13 genera of seven families of two orders of one class in the phylum *Firmicutes*, which have not been previously reported in Korea. Morphological, biochemical, and physiological characteristics, isolation sources, and NIBR deposit numbers of these unrecorded bacterial species are described in the species descriptions.

Keywords: 16S rRNA, *Bacilli*, bacterial diversity, *Firmicutes*, indigenous prokaryotic species in Korea, unrecorded species

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INTRODUCTION

The phylum *Firmicutes* is one of the major phyla in the domain *Bacteria*. *Firmicutes* species are generally Gram-positive bacteria with a low G+C content and known to make up a high proportion of human gut microbiota that plays an important role in health maintenance (Qin *et al.*, 2010). At the time of writing, based on 16S rRNA gene sequences, the phylum *Firmicutes* comprises seven classes with validly published names (<https://lpsn.dsmz.de/phylum/firmicutes>): *Bacilli*, *Clostridia*, *Culicoidibacteria*, *Erysipelotrichia*, *Limnochordia*, *Negativicutes* and *Thermolithobacteria*. The class *Bacilli* is divided into two orders *Bacillales* and *Lactobacillales*. The formation of endospores is one of the characteristics of the order *Bacillales*, whereas the order *Lactobacillales* does not form endospores and produces lactic acid (Ludwig *et al.*,

2009).

In this study, bacterial strains belonging to the phylum *Firmicutes* were isolated from various environments during the investigation of indigenous bacterial species in Korea. As a result, 20 strains representing unrecorded bacterial species in Korea are described.

MATERIALS AND METHODS

Twenty bacterial strains belonging to *Firmicutes* were isolated from various environmental niches such as soil, air, tidal flat, seawater, grain, wetland, breast milk and healthy human urine (Table 1). Treatment of environmental samples and bacterial isolation were handled independently in several laboratories. Pure cultures of isolated bacteria were grown on various culture media including

Table 1. Summary of isolated strains belonged to the *Firmicutes* and their taxonomic affiliations.

Phylum	Class	Order	Family	Genus	Strain ID	NIBR ID	Most closely related species	Similarity (%)	Isolation source	Medium	Incubation conditions
<i>Firmicutes</i>	<i>Bacilli</i>		<i>Bacillaceae</i>	<i>Bacillus</i>	KYW2075	NIBRBAC000506234	<i>Bacillus horikoshii</i>	99.18	Sea water	MA	25°C, 3d
				<i>Bacillus</i>	MMS20-LR456	NIBRBAC000506166	<i>Bacillus fordii</i>	99.65	Soil	LA	45°C, 2d
				<i>Cytobacillus</i>	JHSTF-M24	NIBRBAC000506397	<i>Cytobacillus firmus</i>	99.52	Tidal flat	MA	25°C, 2d
				<i>Gracilibacillus</i>	KYW2175	NIBRBAC000506236	<i>Gracilibacillus diposauri</i>	99.31	Sea water	MA	25°C, 3d
				<i>Gracilibacillus</i>	CAU 1626	NIBRBAC000506283	<i>Gracilibacillus massiliensis</i>	98.72	Soil	MA	30°C, 3-5d
				<i>Halobacillus</i>	BDTF-R2	NIBRBAC000506392	<i>Halobacillus trueperi</i>	99.13	Tidal flat	R2A	25°C, 5d
				<i>Neobacillus</i>	HMF5486	NIBRBAC000506257	<i>Neobacillus mesonae</i>	99.73	Air	R2A	25°C, 3d
				<i>Neobacillus</i>	HMG1274	NIBRBAC000506260	<i>Neobacillus drentensis</i>	99.57	Soil	R2A	25°C, 3d
				<i>Brevibacillus</i>	MMS20-HD16	NIBRBAC000506175	<i>Brevibacillus nitrificans</i>	100	Soil	NA	30°C, 2d
				<i>Brevibacillus</i>	MMS20-A12-23T	NIBRBAC000506173	<i>Brevibacillus invocatus</i>	99.86	Soil	TSA	30°C, 1d
				<i>Paenibacillaceae</i>	JHSTF-R20	NIBRBAC000506396	<i>Paenibacillus xylanivorans</i>	99.39	Tidal flat	R2A	25°C, 2d
				<i>Paenibacillus</i>	JHPTF-R3	NIBRBAC000506399	<i>Paenibacillus tritici</i>	98.85	Tidal flat	R2A	25°C, 3d
				<i>Paenibacillus</i>	HMF5593	NIBRBAC000506258	<i>Paenibacillus taihuensis</i>	99.29	Soil	R2A	25°C, 3d
				<i>Planococcaceae</i>	MMS20-LR3019	NIBRBAC000506167	<i>Psychrobacillus glaciei</i>	99.79	Soil	LA	30°C, 1d
<i>Staphylococcaceae</i>	<i>Staphylococcus</i>	CAU 1627	NIBRBAC000506284	<i>Staphylococcus lugdunensis</i>	100	Human milk	NA	30°C, 2-3d			
	<i>Staphylococcus</i>	I2-44	NIBRBAC000506147	<i>Staphylococcus edaphicus</i>	100	Wetland	R2A	30°C, 3d			
<i>Aerococcaceae</i>	<i>Aerococcus</i>	CAU 1608	NIBRBAC000506269	<i>Aerococcus urinae</i>	99.86	Healthy human urine	BHI	37°C, 2-3d			
<i>Lactobacillales</i>	<i>Lactobacillus</i>	BM-39	NIBRBAC000506400	<i>Lactobacillus paragasseri</i>	100	Breast milk	MRS	37°C, 3d			
	<i>Limosilactobacillus</i>	BM-27	NIBRBAC000506401	<i>Limosilactobacillus fermentum</i>	99.87	Breast milk	MRS	30°C, 3d			
<i>Streptococcaceae</i>	<i>Lactococcus</i>	B5	NIBRBAC000506402	<i>Lactococcus taiwanensis</i>	99.93	Grain	MRS	30°C, 3d			

R2A, marine agar (MA), tryptic soy agar (TSA), nutrient agar (NA), Luria-Bertani agar (LA), brain heart infusion agar (BHI) and de Man, Rogosa and Sharpe agar (MRS) at 25–45°C for 1–5 days, depending on each strain. The designated strain IDs, isolation sources, culture media and incubation conditions are summarized in Table 1. All pure cultures were maintained as 10–20% glycerol suspension at –80°C and lyophilized ampoules.

Bacterial genomic DNA was extracted using the DNeasy Blood and Tissue kit (Qiagen) following the manufacturer's protocols. The 16S rRNA gene was amplified by PCR using primers pBact 27F and pUniv 1492R (Lane, 1991). The PCR amplicons were purified using a gel extraction kit (Qiagen). The 16S rRNA gene sequences were determined at Biofact (Daejeon, Republic of Korea) using an automated DNA analyzer (ABI 3730XL; Applied Biosystems) and the sequencing primers 27F, 1492R, 518F and 805R to generate a consensus sequence (Baker *et al.*, 2003). The 16S rRNA gene sequences of the isolated strains were aligned with those of the related type strains of validly published bacterial species collected from the EzBioCloud (<https://www.ezbiocloud.net/>) (Yoon *et al.*, 2017). Multiple alignments of the 16S rRNA gene sequences of the isolates and those of the related type strains were carried out using the CLUSTAL_X program (Larkin *et al.*, 2007) for the phylogenetic analysis. Phylogenetic trees were reconstructed by neighbour-joining (NJ) method (Saitou and Nei, 1987) using the MEGA X software (Kumar *et al.*, 2018). The NJ trees were reconstructed using the Jukes-Cantor model (Jukes and Cantor, 1969). Tree topologies were evaluated on the basis of bootstrap analysis of 1000 replicates (Felsenstein, 1985).

Cellular morphology and size were examined by either transmission electron microscopy or scanning electron microscopy. Gram staining was performed using standard procedures. Biochemical characteristics were tested using API 20NE kits (bioMérieux) according to the manufacturer's protocol.

RESULTS AND DISCUSSION

Twenty strains belonging to *Firmicutes* were classified into two orders in the class *Bacilli*: 16 strains in the order *Bacillales* and four strains in the order *Lactobacillales* (Table 1). These strains were Gram-staining-positive and rod-shaped except for the strains CAU 1627, I2-44, and CAU 1608, which were coccus-shaped (Fig. 1). Among 16 strains belonging to the order *Bacillales*, eight strains were assigned to the family *Bacillaceae* that include the genera *Bacillus*, *Cytobacillus*, *Gracilibacillus*, *Halobacillus* and *Neobacillus*, five strains belonged to the family *Paenibacillaceae* that include the genera *Brevibacillus* and *Paenibacillus*, one strain belonged to the genus

Psychrobacillus in the family *Planococcaceae*, and two strains belonged to the genus *Staphylococcus* in the family *Staphylococcaceae*. For four strains belonging to the order *Lactobacillales*, one strain belonged to the genus *Aerococcus* in the family *Aerococcaceae*, two strains were assigned to the family *Lactobacillaceae* that include the genera *Lactobacillus* and *Limosilactobacillus*, and one strain belonged to the genus *Lactococcus* within the family *Streptococcaceae*. Phylogenetic relationship between the isolates and those of the related type strains are presented in Fig. 2 and Fig. 3. The detailed descriptions of each species including morphological and physiological characteristics are presented in the following section.

Description of *Bacillus horikoshii* KYW2075

Cells are Gram-staining-positive, flagellated and rod-shaped. Colonies are circular, entire, convex, smooth, opaque and pale yellow-colored after 3 days of incubation on MA at 25°C. Esculin hydrolysis is weakly positive and negative reactions are obtained for nitrate reduction, indole production, glucose fermentation, arginine dihydrolase, urease and gelatinase in API 20NE. Does not utilize L-arabinose, D-mannose, capric acid, adipic acid and phenylacetic acid. Weakly utilizes D-glucose, D-mannitol, N-acetyl-glucosamine, D-maltose, potassium gluconate and trisodium citrate. Strain KYW2075 (=NIBR BAC000506234) was isolated from a seawater sample, Gwangyang, Jeollanam-do, Republic of Korea (34°54'04.30"N, 127°41'41.02"E).

Description of *Bacillus fordii* MMS20-LR456

Cells are Gram-staining-positive, non-flagellated and rod-shaped. Colonies are circular, convex, entire and yellow-colored after 2 days of incubation on LA at 45°C. Esculin hydrolysis is weakly positive and negative reactions are obtained for nitrate reduction, indole production, glucose fermentation, arginine dihydrolase, urease and β -galactosidase in API 20NE. Does not utilize D-glucose, L-arabinose, D-mannose, D-mannitol, N-acetyl-glucosamine, D-maltose, capric acid and adipic acid. Weakly utilizes potassium gluconate, trisodium citrate and phenylacetic acid. Strain MMS20-LR456 (=NIBRBAC000506166) was isolated from a soil sample, Suncheon, Jeollanam-do, Republic of Korea (34°54'18.8"N, 127°30'49.7"E).

Description of *Brevibacillus nitrificans* MMS20-HD16

Cells are Gram-staining-positive, non-flagellated and rod-shaped. Colonies are circular, convex, entire and white-colored after 2 days of incubation on NA at 30°C. Gelatinase is weakly positive and negative reactions are obtained for indole production, glucose fermentation, argi-

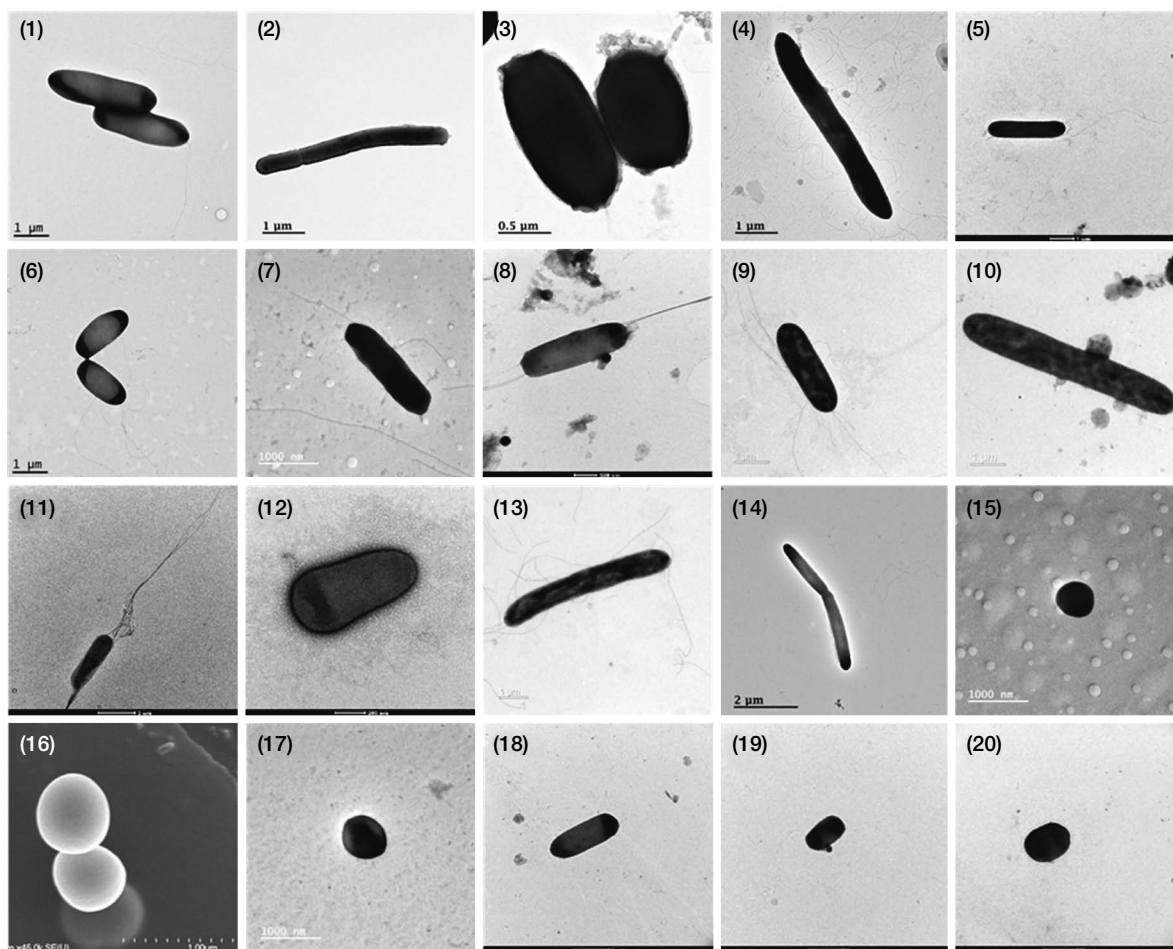


Fig. 1. Transmission electron micrographs and scanning electron micrographs of cells of the species belonging to *Firmicutes* isolated in this study. Strains: 1. KYW2075; 2. MMS20-LR456; 3. MMS20-HD16; 4. MMS20-AI2-23T; 5. JHSTF-M24; 6. KYW2175; 7. CAU 1626; 8. BDTF-R2; 9. HMF5486; 10. HMG1274; 11. JHSTF-R20; 12. JHPTF-R3; 13. HMF5593; 14. MMS20-LR3019; 15. CAU 1627; 16. I2-44; 17. CAU 1608; 18. BM-39; 19. B5; 20. BM-27.

nine dihydrolase, urease, esculin hydrolysis and β -galactosidase in API 20NE. Does not utilize D-glucose, L-arabinose, D-mannose, potassium gluconate, capric acid, adipic acid, malic acid, trisodium citrate and phenylacetic acid. Strain MMS20-HD16 (= NIBRBAC000506175) was isolated from a soil sample, Suncheon, Jeollanam-do, Republic of Korea (34°54'18.8"N, 127°30'49.7"E).

Description of *Brevibacillus invocatus* MMS20-AI2-23T

Cells are Gram-staining-positive, flagellated and rod-shaped. Colonies are circular, convex, entire and white-colored after 1 day of incubation on TSA at 30°C. Negative reactions are obtained for nitrate reduction, indole production, glucose fermentation, arginine dihydrolase, urease, esculin hydrolysis, gelatinase and β -galactosidase in API 20NE. Does not utilize D-glucose, L-arabinose, D-mannose, N-acetyl-glucosamine, D-maltose, potassium

gluconate, capric acid, adipic acid, malic acid, trisodium citrate and phenylacetic acid. Strain MMS20-AI2-23T (= NIBRBAC000506173) was isolated from a soil sample, Okcheon, Chungcheongbuk-do, Republic of Korea (36°19'24.3"N, 127°43'42.4"E).

Description of *Cytobacillus firmus* JHSTF-M24

Cells are Gram-staining-positive, flagellated and rod-shaped. Colonies are circular, glistening and yellowish white-colored after 2 days of incubation on MA at 25°C. Negative reactions are obtained for indole production, glucose fermentation and β -galactosidase in API 20NE. Does not utilize L-arabinose, D-mannose, capric acid, adipic acid and phenylacetic acid. Strain JHSTF-M24 (= NIBRBAC000506397) was isolated from a tidal flat sample, Seocheon, Chungcheongnam-do, Republic of Korea (36°01'44.6"N, 126°39'56.8"E).

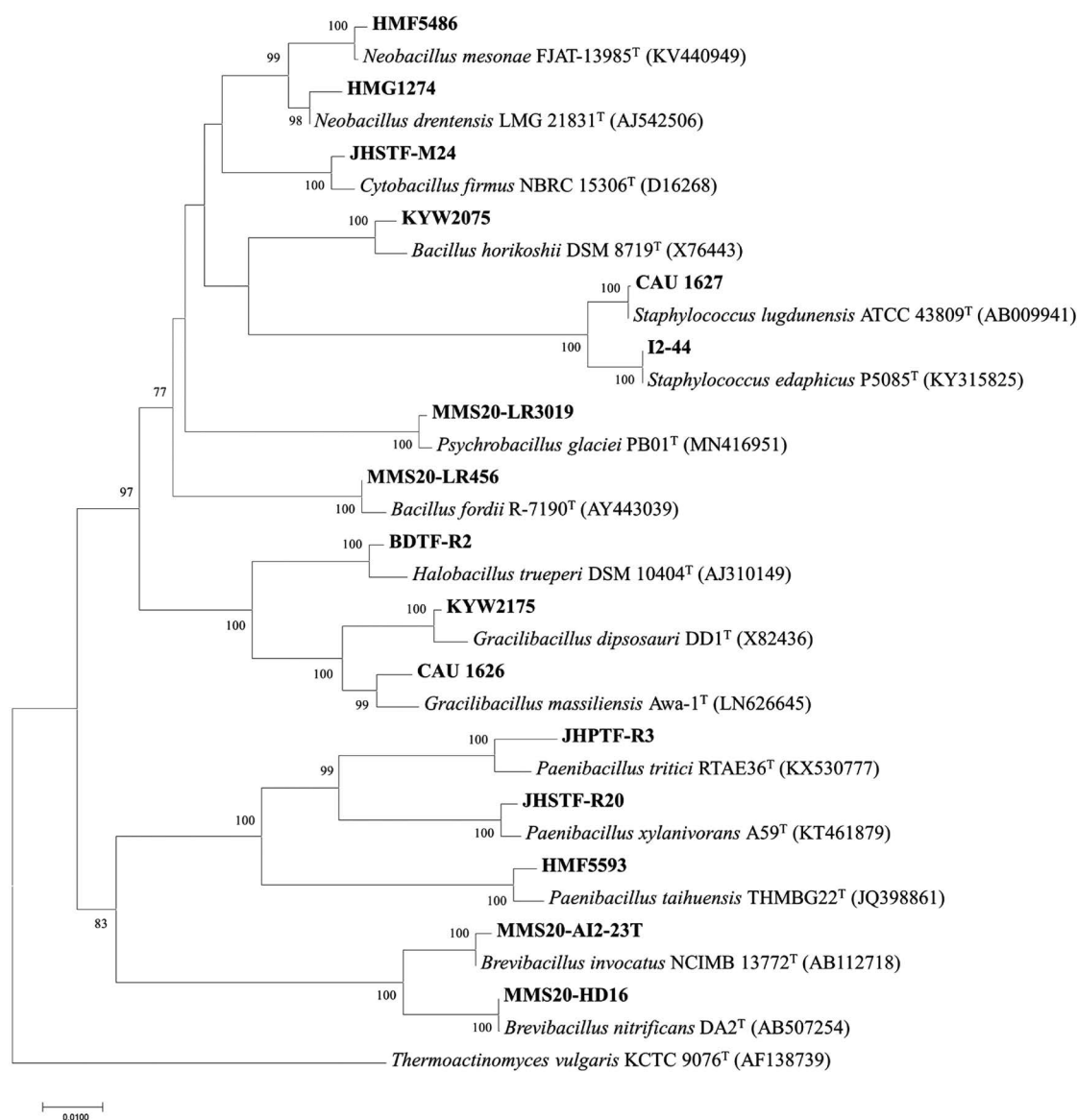


Fig. 2. Neighbor-joining phylogenetic tree, based on 16S rRNA gene sequences, showing the relationship between the strains isolated in this study and their relatives of the order Bacillales. Bootstrap values are shown at branch points based on 1000 replicated datasets; only values above 70% are shown. *Thermoactinomyces vulgaris* KCTC 9076^T (AF138739) was used as an outgroup. Bar, 0.01 substitutions per nucleotide position.

Description of *Gracilibacillus dipsosauri* KYW2175

Cells are Gram-staining-positive, flagellated and rod-shaped. Colonies are circular, entire, convex, opaque and white-colored after 3 days of incubation on MA at 25°C. Negative reactions are obtained for indole production, arginine dihydrolase, urease and gelatinase in API 20NE. Does not utilize D-glucose, L-arabinose, D-mannose, D-mannitol, N-acetyl-glucosamine, D-maltose, potassium gluconate, capric acid, adipic acid, malic acid, trisodium citrate and phenylacetic acid. Strain KYW2175 (=NIBRBAC000506236) was isolated from a seawater

sample, Gwangyang, Jeollanam-do, Republic of Korea (34°54'24.83"N, 127°44'01.47"E).

Description of *Gracilibacillus massiliensis* CAU 1626

Cells are Gram-staining-positive, flagellated and rod-shaped. Colonies are circular, convex, smooth, transparent and white-colored after 3–5 days of incubation on MA at 30°C. Negative reactions are obtained for nitrate reduction, indole production, glucose fermentation, arginine dihydrolase, urease and gelatinase in API 20NE. Does not utilize L-arabinose, D-mannose, D-mannitol, capric acid,

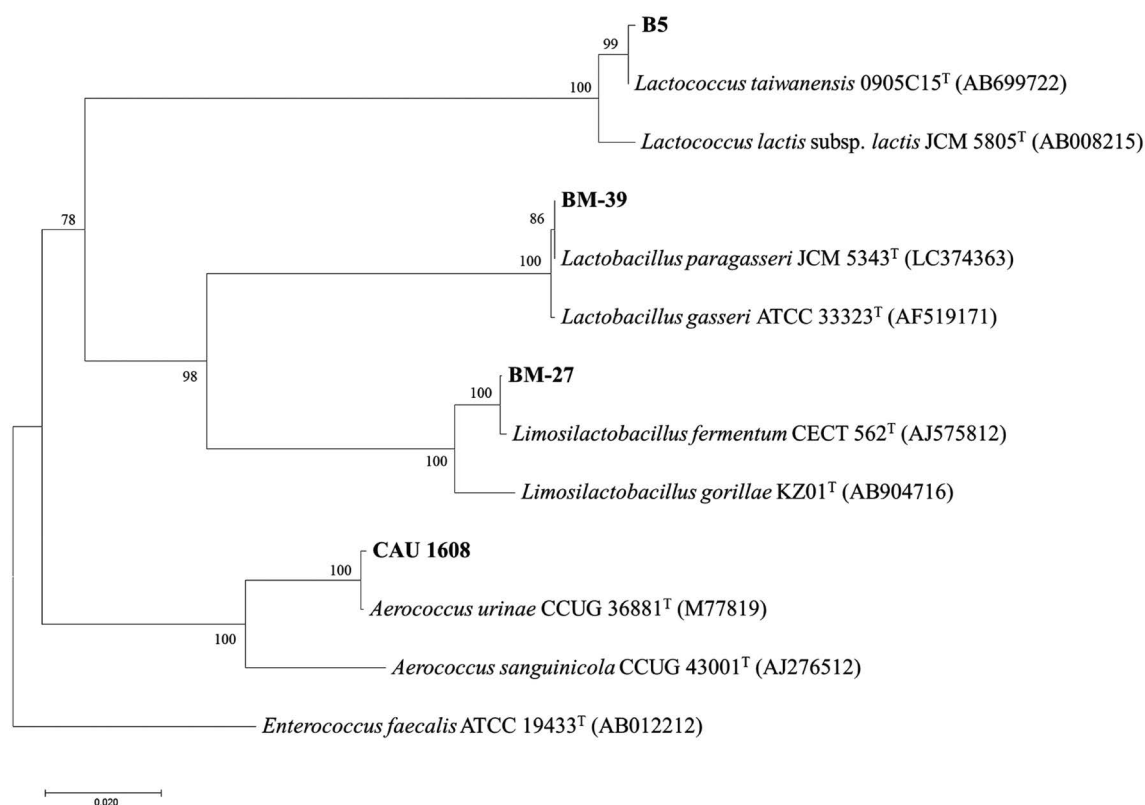


Fig. 3. Neighbor-joining phylogenetic tree, based on 16S rRNA gene sequences, showing the relationship between the strains isolated in this study and their relatives of the order *Lactobacillales*. Bootstrap values are shown at branch points based on 1000 replicated datasets; only values above 70% are shown. *Enterococcus faecalis* ATCC 19433^T (AB012212) was used as an outgroup. Bar, 0.02 substitutions per nucleotide position.

adipic acid, malic acid, trisodium citrate and phenylacetic acid. Strain CAU 1626 (= NIBRBAC000506283) was isolated from a soil sample, Incheon, Republic of Korea (37°31'20.6"N, 126°25'30.0"E).

Description of *Halobacillus trueperi* BDTF-R2

Cells are Gram-staining-positive, non-flagellated and rod-shaped. Colonies are circular, glistening and pale yellow-colored after 5 days of incubation on R2A at 25°C. Negative reactions are obtained for indole production, glucose fermentation, arginine dihydrolase, urease and gelatinase in API 20NE. Does not utilize D-glucose, L-arabinose, D-mannose, D-mannitol, N-acetyl-glucosamine, D-maltose, potassium gluconate, capric acid, adipic acid, malic acid, trisodium citrate and phenylacetic acid. Strain BDTF-R2 (= NIBRBAC000506392) was isolated from a tidal flat sample, Ansan, Gyeonggi-do, Republic of Korea (37°12'23.8"N, 126°38'39.0"E).

Description of *Neobacillus mesonae* HMF5486

Cells are Gram-staining-positive, flagellated and rod-shaped. Colonies are circular, convex, smooth and white-

colored after 3 days of incubation on R2A at 25°C. Negative reactions are obtained for indole production, glucose fermentation and arginine dihydrolase in API 20NE. Does not utilize D-glucose, L-arabinose, D-mannitol, N-acetyl-glucosamine, D-maltose, potassium gluconate, capric acid, adipic acid and trisodium citrate. Strain HMF5486 (= NIBRBAC000506257) was isolated from an air sample, Yongin, Gyeonggi-do, Republic of Korea (37°20'20"N, 127°15'59"E).

Description of *Neobacillus drentensis* HMG1274

Cells are Gram-staining-positive, non-flagellated and rod-shaped. Colonies are circular, convex, smooth and white-colored after 3 days of incubation on R2A at 25°C. Negative reactions are obtained for indole production, glucose fermentation, arginine dihydrolase, urease and gelatinase in API 20NE. Does not utilize potassium gluconate, capric acid, adipic acid and phenylacetic acid. Strain HMG1274 (= NIBRBAC000506260) was isolated from a soil sample, Seoul, Republic of Korea (37°30'17"N, 126°53'46"E).

Description of *Paenibacillus xylanivorans* JHSTF-R20

Cells are Gram-staining-positive, flagellated and rod-shaped. Colonies are circular, glistening and yellowish white-colored after 2 days of incubation on R2A at 25°C. Negative reactions are obtained for nitrate reduction, indole production, glucose fermentation, arginine dihydrolase and urease in API 20NE. Does not utilize D-mannitol, *N*-acetyl-glucosamine, capric acid, adipic acid, malic acid, trisodium citrate and phenylacetic acid. Strain JHSTF-R20 (= NIBRBAC000506396) was isolated from a tidal flat sample, Seocheon, Chungcheongnam-do, Republic of Korea (36°01'44.6"N, 126°39'56.8"E).

Description of *Paenibacillus tritici* JHPTF-R3

Cells are Gram-staining-positive, non-flagellated and rod-shaped. Colonies are circular, slightly convex, glistening and white-colored after 3 days of incubation on R2A at 25°C. Negative reactions are obtained for nitrate reduction, indole production, glucose fermentation, arginine dihydrolase, urease and gelatinase in API 20NE. Does not utilize D-glucose, L-arabinose, D-mannose, D-mannitol, *N*-acetyl-glucosamine, potassium gluconate, capric acid, adipic acid, malic acid, trisodium citrate and phenylacetic acid. Strain JHPTF-R3 (= NIBRBAC000506399) was isolated from a tidal flat sample, Seocheon, Chungcheongnam-do, Republic of Korea (36°01'44.6"N, 126°39'56.8"E).

Description of *Paenibacillus taihuensis* HMF5593

Cells are Gram-staining-positive, flagellated and rod-shaped. Colonies are circular, convex, smooth and white-colored after 3 days of incubation on R2A at 25°C. Negative reactions are obtained for nitrate reduction, indole production, glucose fermentation, arginine dihydrolase, urease and gelatinase in API 20NE. Does not utilize D-mannitol, potassium gluconate, capric acid, adipic acid, malic acid, trisodium citrate and phenylacetic acid. Strain HMF5593 (= NIBRBAC000506258) was isolated from a soil sample, Yongin, Gyeonggi-do, Republic of Korea (37°20'18"N, 127°15'55"E).

Description of *Psychrobacillus glaciei* MMS20-LR3019

Cells are Gram-staining-positive, non-flagellated and rod-shaped. Colonies are circular, convex, entire and yellow-colored after 1 day of incubation on LA at 30°C. Esculin hydrolysis is weakly positive and negative reactions are obtained for nitrate reduction, indole production, glucose fermentation, arginine dihydrolase, urease and β -galactosidase in API 20NE. Does not utilize D-glucose, L-arabinose, D-mannose, D-mannitol, *N*-acetyl-glucosamine, D-maltose, capric acid and adipic acid.

Weakly utilizes potassium gluconate, trisodium citrate and phenylacetic acid. Strain MMS20-LR3019 (= NIBRBAC000506167) was isolated from a soil sample, Suncheon, Jeollanam-do, Republic of Korea (34°54'18.8"N, 127°30'49.7"E).

Description of *Staphylococcus lugdunensis* CAU 1627

Cells are Gram-staining-positive, non-flagellated and coccus-shaped. Colonies are circular, convex, smooth and light orange-colored after 2–3 days of incubation on NA at 30°C. Esculin hydrolysis is weakly positive and negative reactions are obtained for indole production, glucose fermentation, arginine dihydrolase, gelatinase and β -galactosidase in API 20NE. Does not utilize D-glucose, L-arabinose, D-mannitol, *N*-acetyl-glucosamine, D-maltose, potassium gluconate, capric acid, adipic acid, malic acid, tri-sodium citrate and phenylacetic acid. Weakly utilizes D-mannose. Strain CAU 1627 (= NIBRBAC000506284) was isolated from a human milk sample, Seoul, Republic of Korea (37°30'20.0"N, 126°57'32.1"E).

Description of *Staphylococcus edaphicus* I2-44

Cells are Gram-staining-positive, non-flagellated and coccus-shaped. Colonies are round, entire, raised and pale yellow-colored after 3 days of incubation on R2A at 30°C. Negative reactions are obtained for nitrate reduction, indole production, arginine dihydrolase, esculin hydrolysis and gelatinase in API 20NE. Does not utilize L-arabinose, D-mannose, capric acid, adipic acid, malic acid, trisodium citrate and phenylacetic acid. Weakly utilizes *N*-acetyl-glucosamine. Strain I2-44 (= NIBRBAC000506147) was isolated from a wetland sample, Seoul, Republic of Korea (37°34'02.2"N, 127°08'59.1"E).

Description of *Aerococcus urinae* CAU 1608

Cells are Gram-staining-positive, non-flagellated and coccus-shaped. Colonies are punctiform, convex, smooth and white-colored after 2–3 days of incubation on BHI at 37°C. Esculin hydrolysis is weakly positive and negative reactions are obtained for nitrate reduction, indole production, arginine dihydrolase, urease, gelatinase and β -galactosidase in API 20NE. Does not utilize D-glucose, L-arabinose, D-mannose, D-mannitol, *N*-acetyl-glucosamine, D-maltose, potassium gluconate, capric acid, adipic acid, malic acid, trisodium citrate and phenylacetic acid. Strain CAU 1608 (= NIBRBAC000506269) was isolated from a healthy human urine sample, Seoul, Republic of Korea (37°30'20.0"N, 126°57'32.1"E).

Description of *Lactobacillus paragasseri* BM-39

Cells are Gram-staining-positive, non-flagellated and ovoid and rod-shaped. Colonies are circular, slightly con-

vex, glistening and yellowish gray-colored after 3 days of incubation on MRS at 37°C. Negative reactions are obtained for nitrate reduction, indole production, glucose fermentation, arginine dihydrolase, urease, esculin hydrolysis, gelatinase and β -galactosidase in API 20NE. Does not utilize D-glucose, L-arabinose, D-mannose, D-mannitol, N-acetyl-glucosamine, D-maltose, potassium gluconate, capric acid, adipic acid, malic acid, trisodium citrate and phenylacetic acid. Strain BM-39 (=NIBRBAC000506400) was isolated from a breast milk sample, Siheung, Gyeonggi-do, Republic of Korea (37°23'02"N, 126°51'32"E).

Description of *Lactococcus taiwanensis* B5

Cells are Gram-staining-positive, non-flagellated and ovoid and rod-shaped. Colonies are circular, convex, glistening and yellowish white-colored after 3 days of incubation on MRS at 30°C. Negative reactions are obtained for nitrate reduction, indole production and gelatinase in API 20NE. Does not utilize D-glucose, L-arabinose, D-mannose, D-mannitol, N-acetyl-glucosamine, D-maltose, potassium gluconate, capric acid, adipic acid, malic acid, trisodium citrate and phenylacetic acid. Strain B5 (=NIBRBAC000506402) was isolated from a grain sample, Suwon, Gyeonggi-do, Republic of Korea (37°17'25"N, 127°00'03"E).

Description of *Limosilactobacillus fermentum* BM-27

Cells are Gram-staining-positive, non-flagellated and ovoid and rod-shaped. Colonies are circular, convex, glistening and yellowish white-colored after 3 days of incubation on MRS at 30°C. Negative reactions are obtained for nitrate reduction, indole production, glucose fermentation, urease and gelatinase in API 20NE. Does not utilize D-glucose, L-arabinose, D-mannose, D-mannitol, N-acetyl-glucosamine, D-maltose, potassium gluconate, capric acid, adipic acid, malic acid, trisodium citrate and phenylacetic acid. Strain BM-27 (=NIBRBAC000506401) was isolated from a breast milk sample, Siheung, Gyeonggi-do, Republic of Korea (37°23'02"N, 126°51'32"E).

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