

A report of 30 unrecorded bacterial species in Korea, isolated from marine ecosystems in 2021

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To obtain unrecorded bacterial species in Korea, various marine samples were collected from Jeollanam-do Province, Korea in 2021. After plating the samples on marine agar and marine R2A agar, and incubating aerobically and anaerobically, approximately 1200 bacterial strains were isolated and identified using 16S rRNA gene sequences. A total of 30 strains showed $\geq 98.7\%$ 16S rRNA gene sequence similarity with validly published bacterial species but not reported in Korea, indicating that they are unrecorded bacterial species in Korea. The unrecorded bacterial strains belonged to 4 phyla, 7 classes, 13 orders, 19 families, and 22 genera, which were assigned to *Azospirillum*, *Loktanella*, and *Pseudovibrio* of the class *Alphaproteobacteria*; *Grimontia*, *Halomonas*, *Marinobacter*, *Microbulbifer*, *Photobacterium*, *Pseudoalteromonas*, *Pseudidiomarina*, *Ferrimonas*, *Shewanella*, *Simidua*, *Thalassotalea*, and *Vibrio* of the class *Gammaproteobacteria*; *Priestia* and *Enterococcus* of the class *Bacilli*; *Persicobacter* of the class *Cytophagia*; *Aureivirga* of the class *Flavobacteriia*; *Propionigenium* and *Psychrilyobacter* of the class *Fusobacteriia*; and *Tepidibacter* of the class *Clostridia*. The details of the unreported species including Gram reaction, colony and cell morphology, biochemical characteristics, and phylogenetic position are also provided in the description of the strains.

Keywords: 16S rRNA, anaerobic bacteria, islands, seawater, tidal-flat sediment, unrecorded bacterial species

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INTRODUCTION

Marine ecosystems cover more than 70% of the Earth's surface and contain most of aquatic prokaryotes accounting for 1.2×10^{29} cells (Whitman *et al.*, 1998). It is well known that marine prokaryotes play crucial roles in many biogeochemical processes that sustain the marine ecosystem (Beja *et al.*, 2001; Tripp, 2013; Sanchez-Baracaldo, 2015). Therefore, numerous studies on the marine prokaryotes have been conducted in the past decades to obtain valuable and novel species from the ecosystems and elucidate their physiology and metabolisms (Cho and Giovannoni, 2004; Cho *et al.*, 2004; Carini *et al.*, 2015). As one of the studies in Korea, the research program of 'The Survey of Korean Indigenous Species,' has been conducted by the National Institute of Biological Resources (NIBR) since 2006. Owing to the research program, many unrecorded or new prokaryotic species have

been discovered from diverse marine ecosystems on the Korean Peninsula (Cho *et al.*, 2017; Joung *et al.*, 2018; Jung *et al.*, 2021). In 2021, the Honam National Institute of Biological Resources (HNIBR) was newly established for biological survey from islands and coastal areas, and launched research programs focusing on unrecorded species discovered therein.

This study is a part of the research programs supported by the HNIBR in 2021. We tried to isolate previously unrecorded bacterial species in seawater, tidal flat, and aquaculture water in shrimp farms collected from islands and coastal areas in Jeollanam-do Province. In particular, bacterial cultivation under anaerobic conditions was conducted for the tidal flat samples. Based on the 16S rRNA gene-based phylogenetic analyses herein, 30 bacterial strains assigned to the classes *Alphaproteobacteria*, *Gammaproteobacteria*, *Cytophagia*, *Fusobacteriia*, *Bacilli*, *Clostridia*, and *Flavobacteriia* were identified

as new records for bacterial species in Korea, for which taxonomic information and phenotypic characteristics are reported.

MATERIALS AND METHODS

Various marine samples were collected from seawater, tidal flat, and shrimp farms in 2021 and more explained in Island Bioresource total Information System (IBIS; <https://ibis.hnibr.re.kr>). Using a spread plating technique on agar media, an aliquot (100 μ L) of the seawater samples was directly spread onto Marine agar (BD Diagnostics) and aerobically incubated at 20°C for 7 days. Bacterial strains were purified as single colonies and the pure cultures were preserved at -80°C in 20% (v/v) glycerol suspension, as well as lyophilized ampoules. On the other hand, the tidal flat samples were collected using a 30 cm long and 2 cm diameter stainless-steel soil sampler and sub-samples taken at 15 cm depth were immediately transported to the laboratory in an anaerobic jar (Mitsubishi Gas Chemical). After transporting to the laboratory, the samples were placed in a vinyl anaerobic chamber (Coy Laboratory Products) filled with N₂ : H₂ : CO₂ (90 : 5 : 5). Using a homogenizer (IKA), 1 g of the sub-sample was thoroughly mixed with 100 mL of sterile seawater. An aliquot (100 μ L) of the homogenized sample was spread onto R2A agar in aged seawater (marine R2A) and anaerobically incubated in an anaerobic jar at 20°C for 7 days. Details on the strains are shown in Table 1.

For the determination of colony morphology, bacterial colonies were observed after reaching the stationary phase on agar plates. Cellular morphology, including cell shape, presence of flagella, and cell size, was examined by transmission electron microscopy (CM200; Philips) after staining with 2% (w/v) uranyl acetate, and a scanning electron microscope (S-4800; Hitachi). Gram staining was performed using a Gram-staining kit (bioMérieux). Catalase and oxidase activities were examined using 3% hydrogen peroxide and oxidase reagent (bioMérieux), respectively. API 20NE galleries (bioMérieux) were employed for additional biochemical characterization. In API 20NE, the strains were tested according to the manufacturer's instructions except 2% NaCl API AUX medium and 1% L-cysteine added therein for anaerobic cultivation.

For the determination of phylogenetic position of the strains isolated herein, bacterial DNA extraction, PCR amplification, and 16S rRNA gene sequencing were performed using the standard procedures as previously described (Yang and Cho, 2008). The 16S rRNA gene sequences were obtained using the primers 518F and 800R. The resultant 16S rRNA gene sequences were initially compared with those of other bacterial strains with validly published names using the EzBioCloud (Yoon *et al.*,

2017) and the NIBR database. A sequence similarity of 98.7% was used as the cut-off value for bacterial species demarcation (Chun *et al.*, 2018). Therefore, the bacterial strains exhibiting $\geq 98.7\%$ 16S rRNA gene sequence similarities with validly published species, but never reported in Korea were determined as unreported bacterial species. For determining phylogenetic position, multiple sequence alignments between the 16S rRNA gene sequences of 30 strains and those of the unreported species were performed using ClustalW, which was implemented in MEGA X (Kumar *et al.*, 2018). Using the unambiguously aligned 16S rRNA gene sequences, phylogenetic trees based on the neighbor-joining method were reconstructed. The robustness of the inferred phylogenetic trees was evaluated by bootstrap analyses based on 1000 random re-samplings (Felsenstein, 1985).

RESULTS AND DISCUSSION

The 16S rRNA gene sequence analyses using approximately 1200 bacterial strains obtained herein revealed that many strains belonged to novel species or previously unreported species in Korea. Of these, a total of 30 strains showed $\geq 98.7\%$ 16S rRNA gene sequence similarities with unrecorded bacterial species in Korea. The strain information, identification, taxonomic assignment from species to classes, isolation source and sequence accession numbers including the HNIBR and GenBank are listed in Table 1. Phylogenetic assignment of the strains to established bacterial species based on 16S rRNA gene sequence similarity was confirmed by the phylogenetic tree analysis (Figs. 1 and 2). All strains identified as unrecorded species formed a robust clade with the type strains of each corresponding species with high bootstrap values.

The 30 unrecorded bacterial species were phylogenetically diverse, belonging to 4 phyla, 7 classes, 13 orders, 19 families, and 22 genera (Table 1). At the generic level, the unreported species were assigned to *Azospirillum*, *Loktanella*, and *Pseudovibrio* of the class *Alphaproteobacteria*; *Grimontia*, *Halomonas*, *Marinobacter*, *Microbulbifer*, *Photobacterium*, *Pseudoalteromonas*, *Pseudidiomarina*, *Ferrimonas*, *Shewanella*, *Simiduia*, *Thalassotalea*, and *Vibrio* of the class *Gammaproteobacteria*; *Priestia* and *Enterococcus* of the class *Bacilli*; *Peesicobacter* of the class *Cytophagia*; *Aureivirga* of the class *Flavobacteriia*; *Propionigenium* and *Psychrilyobacter* of the class *Fusobacteriia*; and *Tepidibacter* of the class *Clostridia*.

The 30 unrecorded bacterial species identified in this study were Gram-staining-negative or positive, flagellated or non-flagellated, short- or straight-rod or coccoid-shaped bacteria (Fig. 3). Detailed morphological, physiological, and biochemical characteristics of the un-

Table 1. Summary of strains isolated and their taxonomic affiliations.

Class	Order	Family	Strain ID	Accession number	Closest species	Similarity (%)	Source
Alphaproteobacteria	Rhodospirillales	Azospirillaceae	HNIBRBA272	OL742670	<i>Azospirillum massiliensis</i>	98.8	Shrimp farm
		Rhodobacteraceae	HNIBRBA683	OL742681	<i>Loktanelia agnita</i>	99.5	Seawater
	Rhizobiales	Stappiaceae	HNIBRBA773	OL742685	<i>Pseudovibrio stylochi</i>	98.7	Seawater
		Pseudalteromonadaceae	HNIBRBA54	OL742662	<i>Pseudalteromonas amylolytica</i>	98.7	Seawater
	Alteromonadales	Idiomarinaceae	HNIBRBA270	OL742669	<i>Pseudidiomarina aquimaris</i>	98.9	Shrimp farm
		Colwelliaceae	HNIBRBA350	OL742672	<i>Thalassotalea marina</i>	99.8	Shrimp farm
		Shewanellaceae	HNIBRBA489	OL742676	<i>Shewanella submarina</i>	99.6	Seawater
			HNIBRBA684	OL742682	<i>Shewanella kaireitica</i>	99.9	Seawater
			HNIBRBA601	OL742678	<i>Ferrimonas futsuensis</i>	99.2	Seawater
			HNIBRBA151	OL742664	<i>Vibrio furnissii</i>	98.7	Tidal flat
Gammaproteobacteria	Vibrionales		HNIBRBA220	OL742666	<i>Vibrio vulnificus</i>	98.7	Seawater
			HNIBRBA244	OL742667	<i>Vibrio mexicanus</i>	99.3	Seawater
			HNIBRBA255	OL742668	<i>Vibrio nigripulchritudo</i>	99.2	Shrimp farm
			HNIBRBA406	OL742675	<i>Vibrio caribbeanicus</i>	99.7	Seawater
			HNIBRBA666	OL742679	<i>Vibrio halioticoli</i>	99.6	Seawater
			HNIBRBA682	OL742680	<i>Vibrio anguillarum</i>	99.5	Seawater
			HNIBRBA688	OL742684	<i>Vibrio ezurae</i>	99.6	Seawater
			HNIBRBA685	OL742683	<i>Photobacterium sanguinanceri</i>	100	Seawater
			HNIBRBA506	OL742677	<i>Grimontia sedimenti</i>	99.3	Seawater
			HNIBRBA189	OL742665	<i>Microbulbifer marinus</i>	99.7	Tidal flat
Bacilli	Cellvibrionales	Microbulbiferaceae	HNIBRBA189	OL742665	<i>Microbulbifer marinus</i>	99.7	Tidal flat
		Cellvibrionaceae	HNIBRBA370	OL742673	<i>Simidiuia agarivornas</i>	99.2	Seawater
	Pseudomonadales	Marinobacteraceae	HNIBRBA833	OL742686	<i>Marinobacter xesospongiae</i>	99.8	Seawater
		Halomonadaceae	HNIBRBA1036	OL742690	<i>Halomonas lactosivorans</i>	99.4	Tidal flat
	Bacillales	Bacillales	HNIBRBA38	OL742661	<i>Priestia taiwanensis</i>	98.9	Seawater
		Lactobacillales	HNIBRBA274	OL742671	<i>Enterococcus sulfureus</i>	100	Shrimp farm
	Cytophagia	Cytophagales	HNIBRBA100	OL742663	<i>Persicobacter psychrovividus</i>	100	Seawater
		Flavobacteriales	HNIBRBA397	OL742674	<i>Aureivirga marina</i>	99.6	Seawater
	Fusobacteriia	Fusobacteriales	HNIBRBA853	OL742687	<i>Psychrilyobacter atlanticus</i>	99.4	Tidal flat
			HNIBRBA855	OL742688	<i>Propionigenium maris</i>	99.4	Tidal flat
Clostridia	Clostridiales	HNIBRBA866	OL742689	<i>Tepidibacter mesophilus</i>	99.1	Tidal flat	

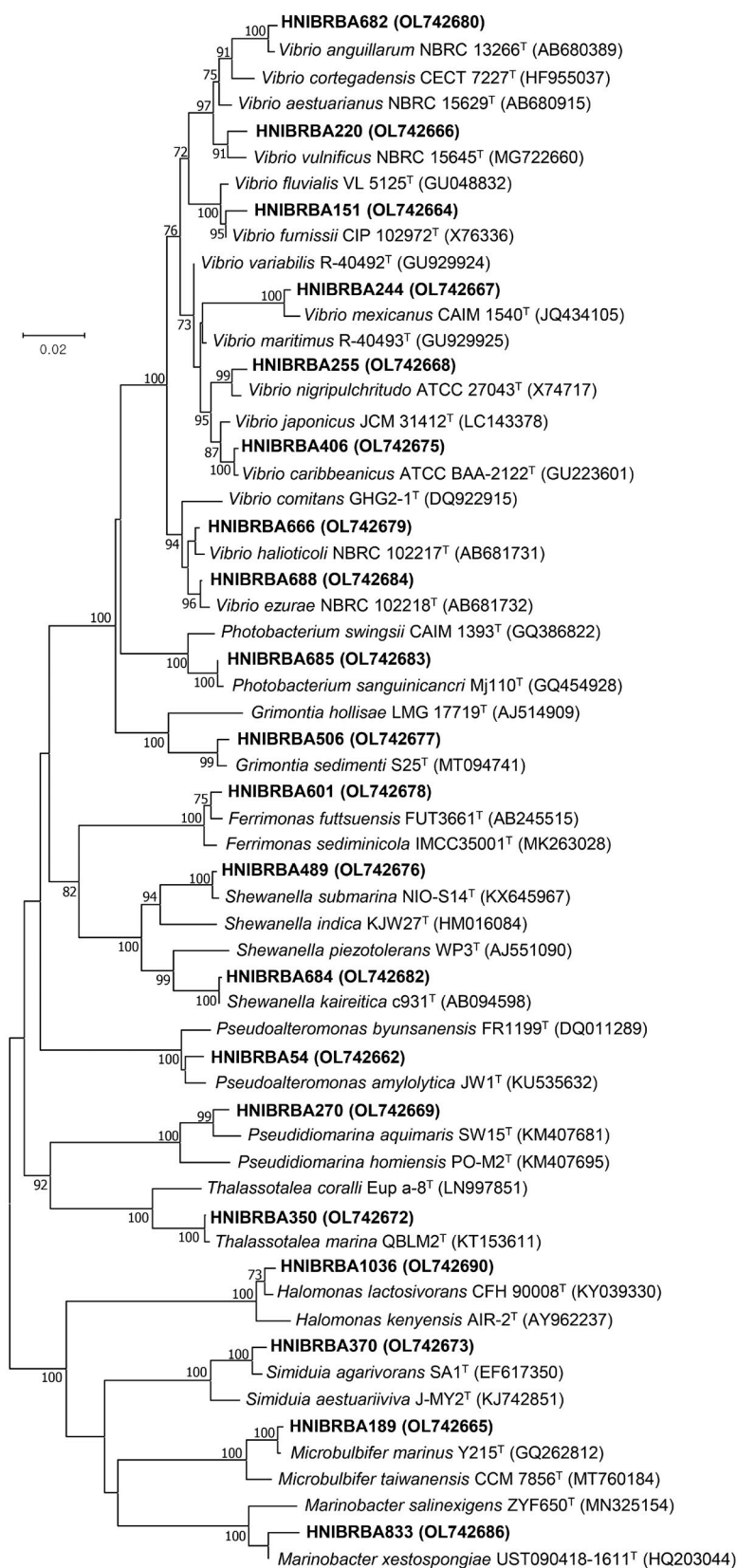


Fig. 1. Neighbor-joining phylogenetic tree based on 16S rRNA gene sequences showing the relationship between the strains isolated in this study and their closest bacterial species in the class *Gammaproteobacteria*. Bootstrap values over 70% are shown.

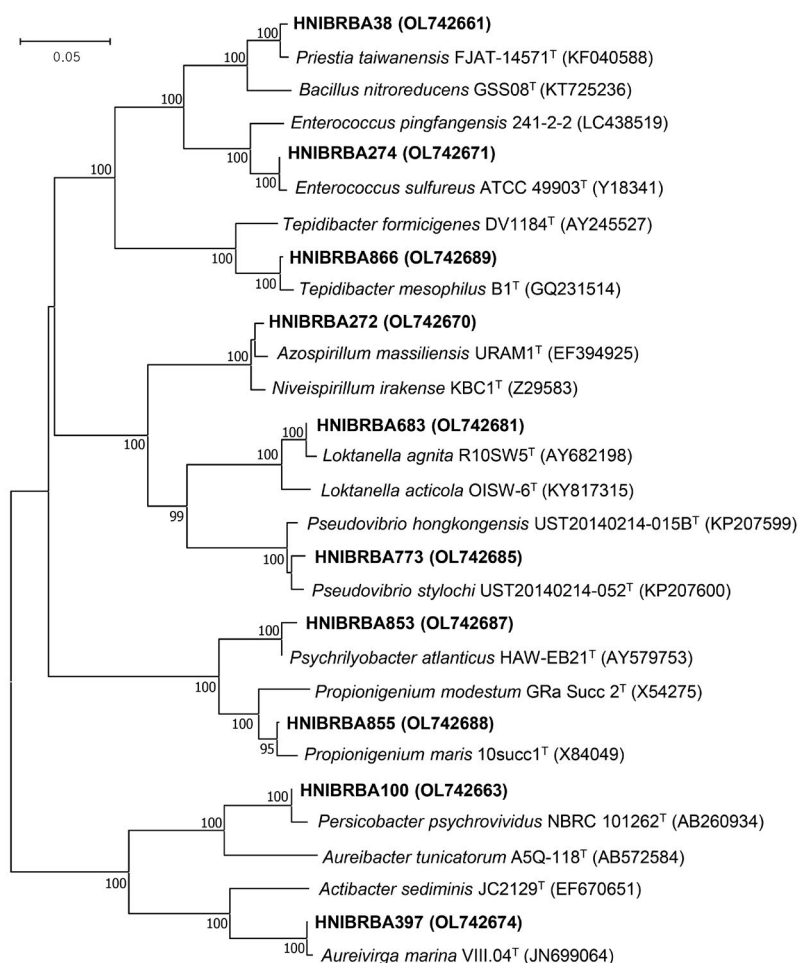


Fig. 2. Neighbor-joining phylogenetic tree based on 16S rRNA gene sequences showing the relationship between the strains isolated in this study and their closest bacterial species in classes *Alphaproteobacteria*, *Bacilli*, *Cytophagia*, *Flavobacteriia*, *Fusobacteriia*, and *Clostridia*. Bootstrap values over 70% are shown.

recorded bacterial species are elucidated in the following strain descriptions.

Description of *Priestia taiwanensis* GHH55

Cells are Gram-stain-positive, flagellated, and rod-shaped. Colonies are circular, flat, smooth, and cream yellow-colored after incubation for 3 days on MA at 20°C. Positive for esculin hydrolysis and cytochrome oxidase; but negative for nitrates reduction, indole production, glucose fermentation, arginine hydrolysis, urease, gelatin hydrolysis, and β -galactosidase in API 20NE. L-Arabinose, D-mannose, D-mannitol, D-maltose, potassium gluconate, capric acid, adipic acid, malic acid, trisodium citrate and phenylacetic acid are not utilized as a sole carbon source. Strain GHH55 (=HNIBRBA38) was isolated from seawater collected off Goha-do (34°45'55"N, 126°22'23"E), Mokpo, Jeollanam-do, Korea. The GenBank accession number of the 16S rRNA gene sequence of strain GHH55

is OL742661.

Description of *Pseudoalteromonas amylolytica* GHH101

Cells are Gram-stain-negative, flagellated, and rod-shaped. Colonies are circular, convex, smooth, and brown-colored after incubation for 3 days on MA at 20°C. Positive for urease, gelatin hydrolysis, and cytochrome oxidase; but negative for nitrates reduction, indole production, glucose fermentation, arginine dihydrolase, esculin hydrolysis, and β -galactosidase in API 20NE. 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 are not utilized as a sole carbon source. Strain GHH101 (=HNIBRBA54) was isolated from seawater collected off Goha-do (34°45'55"N, 126°22'23"E), Mokpo, Jeollanam-do, Korea. The GenBank accession num-

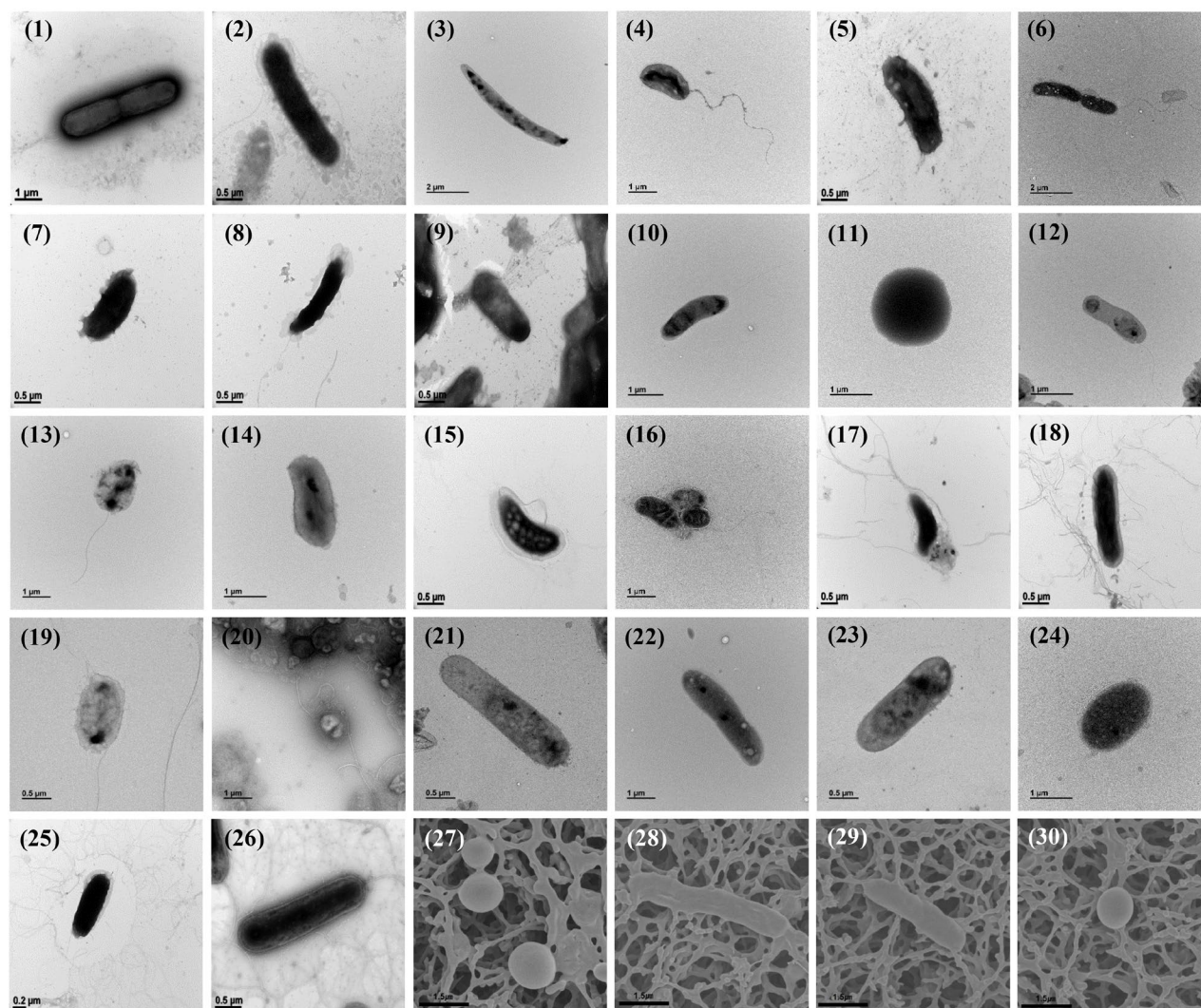


Fig. 3. Transmission electron micrographs of scanning electron micrographs of cells of the strain isolated in this study. Strains: 1, HNI-BRBA38; 2, HNIBRBA54; 3, HNIBRBA100; 4, HNIBRBA151; 5, HNIBRBA189; 6, HNIBRBA220; 7, HNIBRBA244; 8, HNIBRBA255; 9, HNIBRBA270; 10, HNIBRBA272; 11, HNIBRBA274; 12, HNIBRBA350; 13, HNIBRBA370; 14, HNIBRBA397; 15, HNIBRBA406; 16, HNIBRBA489; 17, HNIBRBA506; 18, HNIBRBA601; 19, HNIBRBA666; 20, HNIBRBA682; 21, HNIBRBA683; 22, HNIBRBA684; 23, HNIBRBA685; 24, HNIBRBA688; 25, HNIBRBA773; 26, HNIBRBA833; 27, HNIBRBA853; 28, HNIBRBA855; 29, HNIBRBA866; 30, HNIBRBA1036. Scale bars are indicated in parenthesis after strain ID.

ber of the 16S rRNA gene sequence of strain GHH101 is OL742662.

Description of *Persicobacter psychrovidus* HN9

Cells are Gram-stain-negative, non-flagellated, and rod-shaped. Colonies are circular, convex, entire, and orange-colored after incubation for 3 days on MA at 20°C. Positive for urease, esculin hydrolysis, β -galactosidase, and cytochrome oxidase; but negative for nitrates reduction, indole production, glucose fermentation, arginine dihydrolase, and gelatin hydrolysis in API 20NE. 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 are not utilized as a sole carbon source. Strain HN9 (= HNIBRBA100) was isolated from seawater collected off Songho (34°18'55"N, 126°31'07"E), Haenam, Jeollanam-do, Korea. The GenBank accession number of the 16S rRNA gene sequence of strain HN9 is OL742663.

Description of *Vibrio furnissii* GHR53

Cells are Gram-stain-negative, flagellated, and rod-shaped. Colonies are circular, convex, entire, and yellow-colored after incubation for 3 days on MA at 20°C. Positive for nitrates reduction, indole production, glucose fermentation, arginine dihydrolase, gelatin hydrolysis,

β -galactosidase, and cytochrome oxidase; but negative for urease and esculin hydrolysis in API 20NE. D-Glucose, L-arabinose, D-mannose, D-mannitol, *N*-acetyl-glucosamine, D-maltose, potassium gluconate, capric acid, malic acid, and trisodium citrate are utilized as a sole carbon source; but not adipic acid and phenylacetic acid. Strain GHR53 (=HNIBRBA151) was isolated from tidal flat in Goha-do (34°46'10"N, 126°21'38"E), Mokpo, Jeollanam-do, Korea. The GenBank accession number of the 16S rRNA gene sequence of strain GHR53 is OL742664.

Description of *Microbulbifer marinus* AT30

Cells are Gram-stain-negative, non-flagellated, and rod-shaped. Colonies are irregular, convex, entire, and yellow-colored after incubation for 3 days on MA at 20°C. Positive for nitrates reduction, gelatin hydrolysis, β -galactosidase, and cytochrome oxidase; but negative for indole production, glucose fermentation, arginine dihydrolase, urease, and gelatin hydrolysis in API 20NE. 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 are not utilized as a sole carbon source. Strain AT30 (=HNIBRBA189) was isolated from tidal flat in Am-tae-do (34°51'44"N, 126°08'06"E), Shinan, Jeollanam-do, Korea. The GenBank accession number of the 16S rRNA gene sequence of strain AT30 is OL742665.

Description of *Vibrio vulnificus* ND14

Cells are Gram-stain-negative, flagellated, and rod-shaped. Colonies are irregular, convex, entire, and yellow-colored after incubation for 3 days on MA at 20°C. Positive for nitrates reduction, indole production, glucose fermentation, arginine dihydrolase, urease, esculin hydrolysis, gelatin hydrolysis, β -galactosidase, and cytochrome oxidase in API 20NE. D-Glucose, D-mannose, D-mannitol, *N*-acetyl-glucosamine, malic acid, and trisodium citrate are utilized as a sole carbon source; but not L-arabinose, capric acid, adipic acid, and phenylacetic acid. Strain ND14 (=HNIBRBA220) was isolated from seawater collected off Nokdong (34°31'56"N, 127°07'18"E), Goheung, Jeollanam-do, Korea. The GenBank accession number of the 16S rRNA gene sequence of strain ND14 is OL742666.

Description of *Vibrio mexicanus* ND47

Cells are Gram-stain-negative, flagellated, and rod-shaped. Colonies are circular, convex, entire, and yellow-colored after incubation for 3 days on MA at 20°C. Positive for nitrates reduction, indole production, glucose fermentation, esculin hydrolysis, β -galactosidase, and cytochrome oxidase; but negative for arginine dihydro-

lase, urease, and gelatin hydrolysis in API 20NE. D-Mannitol, *N*-acetyl-glucosamine, D-maltose and malic acid are utilized as a sole carbon source; but not D-glucose, L-arabinose, D-mannose, potassium gluconate, capric acid, adipic acid, trisodium citrate, and phenylacetic acid. Strain ND47 (=HNIBRBA244) was isolated from seawater collected off Nokdong (34°31'56"N, 127°07'18"E), Goheung, Jeollanam-do, Korea. The GenBank accession number of the 16S rRNA gene sequence of strain ND47 is OL742667.

Description of *Vibrio nigripulchritudo* JNT22

Cells are Gram-stain-negative, flagellated, and rod-shaped. Colonies are circular, convex, entire, and black-colored after incubation for 3 days on MA at 20°C. Positive for glucose fermentation, gelatin hydrolysis, β -galactosidase, and cytochrome oxidase; but negative for nitrates reduction, indole production, arginine dihydrolase, and esculin hydrolysis in API 20NE. D-Mannose, D-maltose and malic acid are utilized as a sole carbon source; but not D-glucose, L-arabinose, D-mannitol, *N*-acetyl-glucosamine, potassium gluconate, capric acid, adipic acid, trisodium citrate and phenylacetic acid. Strain JNT22 (=HNIBRBA255) was isolated from aquaculture water collected from a shrimp farm (34°32'11"N, 126°20'41"E), Jindo, Jeollanam-do, Korea. The GenBank accession number of the 16S rRNA gene sequence of strain JNT22 is OL742668.

Description of *Pseudidiomarina aquimaris* JNT52

Cells are Gram-stain-negative, non-flagellated, and rod-shaped. Colonies are circular, convex, entire, and yellow-colored after incubation for 3 days on MA at 20°C. Positive for nitrates reduction, arginine dihydrolase, urease, esculin hydrolysis, and cytochrome oxidase; but negative for indole production glucose fermentation, gelatin hydrolysis, and β -galactosidase in API 20NE. 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 are not utilized as a sole carbon source. Strain JNT52 (=HNIBRBA270) was isolated from aquaculture water collected from a shrimp farm (34°32'11"N, 126°20'41"E), Jindo, Jeollanam-do, Korea. The GenBank accession number of the 16S rRNA gene sequence of strain JNT52 is OL742669.

Description of *Azospirillum massiliensis* GHA1

Cells are Gram-stain-negative, non-flagellated, and rod-shaped. Colonies are circular, convex, entire, and yellow-colored after incubation for 3 days on R2A at 20°C. Positive for nitrates reduction, arginine dihydrolase, ure-

ase, esculin hydrolysis, gelatin hydrolysis, β -galactosidase, and cytochrome oxidase; but negative for indole production and glucose fermentation in API 20NE. D-Glucose, D-mannose, *N*-acetyl-glucosamine, D-maltose, and malic acid are utilized as a sole carbon source; but not L-arabinose, potassium gluconate, capric acid, adipic acid, trisodium citrate, and phenylacetic acid. Strain GHA1 (= HNIBRBA272) was isolated from aquaculture water collected from a shrimp farm (34°41'29"N, 126°22'21"E), Haenam, Jeollanam-do, Korea. The GenBank accession number of the 16S rRNA gene sequence of strain GHA1 is OL742670.

Description of *Enterococcus sulfuresus* GHA5

Cells are Gram-stain-positive, non-flagellated, and coccoid-shaped. Colonies are circular, convex, entire, and yellow-colored after incubation for 3 days on R2A at 20°C. Positive for glucose fermentation, esculin hydrolysis, gelatin hydrolysis, and β -galactosidase; but negative for nitrates reduction, indole production, arginine dihydrolase, urease, and cytochrome oxidase in API 20NE. D-Glucose are utilized as a sole carbon source; but not 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 GHA5 (= HNIBRBA274) was isolated from aquaculture water collected from a shrimp farm (34°41'29"N, 126°22'21"E), Haenam, Jeollanam-do, Korea. The GenBank accession number of the 16S rRNA gene sequence of strain GHA5 is OL742671.

Description of *Thalassotalea marina* JDF44

Cells are Gram-stain-negative, non-flagellated, and rod-shaped. Colonies are circular, convex, smooth, and yellow-colored after incubation for 3 days on MA at 20°C. Positive for esculin hydrolysis, β -galactosidase, and cytochrome oxidase; but negative for nitrates reduction, indole production, glucose fermentation, arginine dihydrolase, urease, and gelatin hydrolysis in API 20NE. 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 are not utilized as a sole carbon source. Strain JDF44 (= HNIBRBA350) was isolated from aquaculture water collected from a shrimp farm (34°32'08"N, 126°20'43"E), Jindo, Jeollanam-do, Korea. The GenBank accession number of the 16S rRNA gene sequence of strain JDF44 is OL742672.

Description of *Simiduia agrarivorans* BK30

Cells are Gram-stain-negative, non-flagellated, and coccoid-shaped. Colonies are circular, convex, entire, and yellow-colored after incubation for 3 days on MA at 20°C.

Positive for nitrates reduction, gelatin hydrolysis, and cytochrome oxidase; but negative for indole production, glucose fermentation, arginine, urease, esculin hydrolysis, and β -galactosidase in API 20NE. D-Maltose are utilized as a sole carbon source; but not 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 BK30 (= HNIBRBA370) was isolated from the seawater collected off Jaeun-do (34°51'00"N, 126°02'18"E), Shinan, Jeollanam-do, Korea. The GenBank accession number of the 16S rRNA gene sequence of strain BK30 is OL742673.

Description of *Aureivirga marina* PJ35

Cells are Gram-stain-negative, non-flagellated, and rod-shaped. Colonies are circular, convex, entire, and brown-colored after incubation for 3 days on MA at 20°C. Positive for nitrates reduction, urease, and cytochrome oxidase; but negative for indole production, glucose fermentation, arginine dihydrolase, esculin hydrolysis, gelatin hydrolysis, and β -galactosidase in API 20NE. 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 are not utilized as a sole carbon source. Strain PJ35 (= HNIBRBA397) was isolated from seawater collected off Jaeun-do (34°55'8.59"N, 126°03'37.26"E), Shinan, Jeollanam-do, Korea. The GenBank accession number of the 16S rRNA gene sequence of strain PJ35 is OL742674.

Description of *Vibrio caribbeanicus* PJ47

Cells are Gram-stain-negative, flagellated, and rod-shaped. Colonies are translucent, convex, smooth, and cream-colored after incubation for 3 days on MA at 20°C. Positive for arginine dihydrolase, urease, gelatin hydrolysis, and cytochrome oxidase; but negative for nitrates reduction, indole production, glucose fermentation, esculin hydrolysis, and β -galactosidase in API 20NE. D-Glucose, *N*-acetyl-glucosamine and malic acid are utilized as a sole carbon source; but not L-arabinose, D-mannose, D-mannitol, D-maltose, potassium gluconate, capric acid, adipic acid, trisodium citrate, and phenylacetic acid. Strain PJ47 (= HNIBRBA406) was isolated from seawater collected off Jaeun-do (34°55'8.59"N, 126°03'37.26"E), Shinan, Jeollanam-do, Korea. The GenBank accession number of the 16S rRNA gene sequence of strain PJ47 is OL742675.

Description of *Shewanella submarina* KY46

Cells are Gram-stain-negative, flagellated, and rod-shaped. Colonies are translucent, convex, entire, and cream-colored after incubation for 3 days on MA at 20°C. Positive for nitrates reduction, arginine dihydrolase, urease, esculin hydrolysis, gelatin hydrolysis, and cy-

tochrome oxidase; but negative for indole production, glucose fermentation, and β -galactosidase in API 20NE. D-Mannose, *N*-acetyl-glucosamine and D-maltose are utilized as a sole carbon source; but not D-glucose, L-arabinose, D-mannitol, potassium gluconate, capric acid, adipic acid, malic acid, trisodium citrate, and phenylacetic acid. Strain KY46 (=HNIBRBA489) was isolated from the seawater collected off Myo-do (34°53'00"N, 127°44'52"E), Kwangyang, Jeollanam-do, Korea. The GenBank accession number of the 16S rRNA gene sequence of strain KY46 is OL742676.

Description of *Grimontia sedimenti* KG16

Cells are Gram-stain-negative, flagellated, and rod-shaped. Colonies are circular, entire, opaque, and cream-colored after incubation for 3 days on MA at 20°C. Positive for nitrates reduction, indole production, glucose fermentation, β -galactosidase, and cytochrome oxidase; but negative for arginine dihydrolase, urease, esculin hydrolysis, and gelatin hydrolysis in API 20NE. D-Glucose, D-mannose, *N*-acetyl-glucosamine, and malic acid are utilized as a sole carbon source; but not L-arabinose, D-mannitol, D-maltose, potassium gluconate, capric acid, adipic acid, trisodium citrate, and phenylacetic acid. Strain KG16 (=HNIBRBA506) was isolated from seawater collected off Geumgap (34°23'46"N, 126°16'32"E), Jindo, Jeollanam-do, Korea. The GenBank accession number of the 16S rRNA gene sequence of strain KG16 is OL742677.

Description of *Ferrimonas futtsuensis* KJ05

Cells are Gram-stain-negative, flagellated, and rod-shaped. Colonies are circular, entire, opaque, and brown-colored after incubation for 3 days on MA at 20°C. Positive for nitrates reduction, arginine dihydrolase, urease, and cytochrome oxidase; but negative for indole production, glucose fermentation, esculin hydrolysis, gelatin hydrolysis, and β -galactosidase in API 20NE. 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 are not utilized as a sole carbon source. Strain KJ05 (=HNIBRBA601) was isolated from seawater collected off Daegu (34°31'32"N, 126°47'23"E), Gagnjin, Jeollanam-do, Korea. The GenBank accession number of the 16S rRNA gene sequence of strain KJ05 is OL742678.

Description of *Vibrio haliotocoli* SK37

Cells are Gram-stain-negative, non-flagellated, and rod-shaped. Colonies are circular, entire, and yellow-colored after incubation for 3 days on MA at 20°C. Positive for nitrates reduction, indole production, glucose fermentation, gelatin hydrolysis, and cytochrome oxidase; but neg-

ative for arginine dihydrolase, urease, esculin hydrolysis, and β -galactosidase in API 20NE. 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 are not utilized as a sole carbon source. Strain SK37 (=HNIBRBA666) was isolated from seawater collected off the Wando Port (34°19'33"N, 126°45'01"E), Wando, Jeollanam-do, Korea. The GenBank accession number of the 16S rRNA gene sequence of strain SK37 is OL742679.

Description of *Vibrio anguillarum* SK53

Cells are Gram-stain-negative, non-flagellated, and rod-shaped. Colonies are circular, convex, entire, and yellow-colored after incubation for 3 days on MA at 20°C. Positive for nitrates reduction, indole production, glucose fermentation, arginine dihydrolase, urease, esculin hydrolysis, gelatin hydrolysis, β -galactosidase, and cytochrome oxidase in API 20NE. *N*-Acetyl-glucosamine, D-maltose, and potassium gluconate are utilized as a sole carbon source; but not D-glucose, L-arabinose, D-mannose, D-mannitol, capric acid, adipic acid, malic acid, trisodium citrate, and phenylacetic acid. Strain SK53 (=HNIBRBA682) was isolated from seawater collected off the Wando Port (34°19'33"N, 126°45'01"E), Wando, Jeollanam-do, Korea. The GenBank accession number of the 16S rRNA gene sequence of strain SK53 is OL742680.

Description of *Loktanella agnita* SK54

Cells are Gram-stain-negative, non-flagellated, and rod-shaped. Colonies are irregular, convex, smooth, and cream-colored after incubation for 3 days on MA at 20°C. Positive for nitrates reduction, arginine dihydrolase, urease, β -galactosidase, and cytochrome oxidase; but negative for indole production, glucose fermentation, esculin hydrolysis, and gelatin hydrolysis in API 20NE. 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 are not utilized as a sole carbon source. Strain SK54 (=HNIBRBA683) was isolated from seawater collected off the Wando Port (34°19'33"N, 126°45'01"E), Wando, Jeollanam-do, Korea. The GenBank accession number of the 16S rRNA gene sequence of strain SK54 is OL742681.

Description of *Shewanella kaireitica* SK55

Cells are Gram-stain-negative, non-flagellated, and rod-shaped. Colonies are entire, semitranslucent, smooth, and pink-colored after incubation for 3 days on MA at 20°C. Positive for nitrates reduction, arginine dihydrolase, urease, esculin hydrolysis, gelatin hydrolysis, β -galactosidase, and cytochrome oxidase; but negative for

indole production and glucose fermentation in API 20NE. 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 are not utilized as a sole carbon source. Strain SK55 (=HNIBRBA684) was isolated from seawater collected off the Wando Port (34°19'33"N, 126°45'01"E), Wando, Jeollanam-do, Korea. The GenBank accession number of the 16S rRNA gene sequence of strain SK55 is OL742682.

Description of *Photobacterium sanguinancrri* SK56

Cells are Gram-stain-negative, non-flagellated, and rod-shaped. Colonies are circular, convex, entire, and cream-colored after incubation for 3 days on MA at 20°C. Positive for nitrates reduction, glucose fermentation, arginine dihydrolase, urease, β -galactosidase, and cytochrome oxidase; but negative for indole production, esculin hydrolysis, and gelatin hydrolysis in API 20NE. 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, are not utilized as a sole carbon source. Strain SK56 (=HNIBRBA685) was isolated from seawater collected off the Wando Port (34°19'33"N, 126°45'01"E), Wando, Jeollanam-do, Korea. The GenBank accession number of the 16S rRNA gene sequence of strain SK56 is OL742683.

Description of *Vibrio ezuræ* SK59

Cells are Gram-stain-negative, non-flagellated, and rod-shaped. Colonies are circular, convex, entire, and yellow-colored after incubation for 3 days on MA at 20°C. Positive for nitrates reduction, indole production, glucose fermentation, and cytochrome oxidase; but negative for arginine dihydrolase, urease, esculin hydrolysis, gelatin hydrolysis, and β -galactosidase in API 20NE. 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 are not utilized as a sole carbon source. Strain SK59 (=HNIBRBA688) was isolated from seawater collected off the Wando Port (34°19'33"N, 126°45'01"E), Wando, Jeollanam-do, Korea. The GenBank accession number of the 16S rRNA gene sequence of strain SK59 is OL742684.

Description of *Pseudovibrio stylochi* KHSW6

Cells are Gram-stain-negative, flagellated, and rod-shaped. Colonies are circular, convex, smooth, and yellow-colored after incubation for 3 days on MA at 20°C. Positive for nitrates reduction and cytochrome oxidase;

but negative for indole production, glucose fermentation, arginine dihydrolase, urease, esculin hydrolysis, gelatin hydrolysis, and β -galactosidase in API 20NE. D-Glucose, and D-mannose are utilized as a sole carbon source; but not L-arabinose, D-mannitol, N-acetyl-glucosamine, D-maltose, potassium gluconate, capric acid, adipic acid, malic acid, trisodium citrate, and phenylacetic acid. Strain KHSW6 (=HNIBRBA773) was isolated from seawater collected off Jungsan (34°43'50.03"N, 127°19'33.95"E), Goheung, Jeollanam-do, Korea. The GenBank accession number of the 16S rRNA gene sequence of strain KHSW6 is OL742685.

Description of *Marinobacter xestospongiae* SSB41

Cells are Gram-stain-negative, flagellated, and rod-shaped. Colonies are circular, convex, smooth, and cream-colored after incubation for 3 days on MA at 20°C. Positive for nitrates reduction and cytochrome oxidase; but negative for indole production, glucose fermentation, arginine dihydrolase, urease, esculin hydrolysis, gelatin hydrolysis, and β -galactosidase in API 20NE. Malic acid and trisodium citrate are utilized as a sole carbon source; but not D-glucose, D-arabinose, D-mannose, D-mannitol, N-acetyl-glucosamine, D-maltose, potassium gluconate, capric acid, adipic acid, and phenylacetic acid. Strain SSB41 (=HNIBRBA833) was isolated from seawater collected off Jaeun-do (34°51'51"N, 125°59'59"E), Shian, Jeollanam-do, Korea. The GenBank accession number of the 16S rRNA gene sequence of strain SSB41 is OL742686.

Description of *Psychrilyobacter atlanticus* GHS7

Cells are Gram-stain-negative, non-flagellated, and cocci-shaped. Colonies are circular, convex, entire, and white-colored after anaerobic incubation for 7 days on marine R2A at 20°C. Positive for esculin hydrolysis; but negative for nitrate reduction, indole production, glucose fermentation, arginine dihydrolase, urease, gelatin hydrolysis, β -galactosidase, and cytochrome oxidase in API 20NE. 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 are not utilized as a sole carbon source. Strain GHS7 (=HNIBRBA853) was isolated from tidal flat collected off Goha-do (34°45'34.91"N, 126°21'53.67"E), Mokpo, Jeollanam-do, Korea. The GenBank accession number of the 16S rRNA gene sequence of strain GHS7 is OL742687.

Description of *Propionigenium maris* GHS9

Cells are Gram-stain-negative, non-flagellated, and rod-shaped. Colonies are circular, convex, entire, and white-

colored after anaerobic incubation for 7 days on marine R2A at 20°C. Positive for indole production, glucose fermentation, arginine dihydrolase, and cytochrome oxidase; but negative for nitrate reduction, urease, esculin hydrolysis, gelatin hydrolysis, and β -galactosidase in API 20NE. 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 are not utilized as a sole carbon source. Strain GHS9 (=HNIBRBA855) was isolated from tidal flat collected off Goha-do (34°45'34.91"N, 126°21'53.67"E), Mokpo, Jeollanam-do, Korea. The GenBank accession number of the 16S rRNA gene sequence of strain GHS9 is OL742688.

Description of *Tepidibacter mesophilus* GHS20

Cells are Gram-stain-positive, non-flagellated, and rod-shaped. Colonies are circular, convex, entire, and white-colored after anaerobic incubation for 7 days on marine R2A at 20°C. Negative for nitrates reduction, indole production, glucose fermentation, arginine dihydrolase, urease, esculin hydrolysis, gelatin hydrolysis, β -galactosidase, and cytochrome oxidase in API 20NE. 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 are not utilized as a sole carbon source. Strain GHS20 (=HNIBRBA866) was isolated from tidal flat collected off Goha-do (34°45'34.91"N, 126°21'53.67"E), Mokpo, Jeollanam-do, Korea. The GenBank accession number of the 16S rRNA gene sequence of strain GHS20 is OL742689.

Description of *Halomonas lactosivorans* KEMB43-101

Cells are Gram-stain-negative, non-flagellated, and cocc-shaped. Colonies are circular, convex, entire, and yellow-colored after incubation for 3 days on MA at 20°C. Positive for arginine dihydrolase, urease, and cytochrome oxidase; but negative for nitrate reduction, indole production, glucose fermentation, esculin hydrolysis, gelatin hydrolysis, and β -galactosidase in API 20NE. Malic acid and trisodium citrate are utilized as a sole carbon source but not D-glucose, L-arabinose, D-mannose, D-mannitol, *N*-acetyl-glucosamine, D-maltose, potassium gluconate, capric acid, adipic acid, and phenylacetic acid are not utilized as a sole carbon source. Strain KEMB43-101 (=HNIBRBA1036) was isolated from tidal flat collected off Goha-do (34°45'34.91"N, 126°21'53.67"E), Mokpo, Jeollanam-do, Korea. The GenBank accession number of the 16S rRNA gene sequence of strain KEMB43-101 is OL742690.

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