

Discovery of 18 previously unrecorded bacterial species in the coastal areas surrounding Korean islands in 2023

Yeonjung Lim, Hyeonuk Sa, Minjeong Kim, Minseok Kim, Jisoo Han, Hyerim Cho and Jang-Cheon Cho*

Department of Biological Sciences and Bioengineering, Inha University, Incheon 22212, Republic of Korea

*Correspondent: chojc@inha.ac.kr

Bacterial communities residing on islands have a significant impact on the functioning and establishment of a unique isolated ecosystem. Notwithstanding, systematic research on the indigenous microbial resources of domestic islands has been lacking. In order to understand the biodiversity and potential bioresources, we conducted sampling in 2023 from coastal waters from various islands off the west coast of the Korean Peninsula, including Baengnyeongdo, Daebudo, Deokjeokdo, Jangbongdo, Yeonpyeongdo Islands, along with Somaemuldo Island along the south coast. The coastal seawater samples were used to unearth microbial resources through the standard dilution plating. In total, approximately 1,600 bacterial strains were isolated from the samples as single colonies and identified using 16S rRNA gene sequence analyses. Eighteen strains, exhibiting $\geq 98.7\%$ 16S rRNA gene sequence similarity to bacterial species with validly published names but not previously reported in Korea, were categorized as unrecorded bacterial species in Korea. These unrecorded bacterial strains displayed phylogenetic diversity, representing three phyla, four classes, 9 orders, 13 families, and 18 genera. The unrecorded species were assigned to the classes *Alphaproteobacteria* (*Aliiroseovarius*, *Kiloniella*, *Maritalea*, *Palleronia*, and *Roseobacter*), *Gammaproteobacteria* (*Aliamphritea*, *Aliivibrio*, *Enterovibrio*, *Francisella*, *Leucothrix*, *Pseudoalteromonas*, *Psychrobium*, *Shewanella*, and *Vibrio*), *Flavobacteriia* (*Aquimarina*, *Pseudofulvibacter*, and *Tenacibaculum*), and *Verrucomicrobiae* (*Roseibacillus*). This study presents comprehensive descriptions of the taxonomic attributes of these unrecorded species, covering morphology, biochemistry, and phylogenetic position.

Keywords: 16S rRNA, coast, island, prokaryote, seawater, unrecorded bacterial species

© 2024 National Institute of Biological Resources
DOI:10.12651/JSR.2024.13.3.318

INTRODUCTION

The Korean Peninsula possesses a complex coastline with approximately 3,400 islands scattered throughout. Each island is composed of marine, coastal, and soil ecosystems that contribute to its unique characteristics, and these ecological systems are known to function as bridges, resulting in islands having high levels of biodiversity (Li *et al.*, 2020). Furthermore, due to their geographic isolation, islands provide excellent research opportunities for independent evolutionary adaptations (Losos and Ricklefs, 2009). Despite ranking as the 12th most island-rich country globally (Armstrong, 2023), research focusing on the diversity of indigenous bacterial species centered around Korean islands has been relatively scarce.

To address this research gap, the Honam National Institute of Biological Resources (HNIBR) initiated the research program entitled 'Survey of Island-Coastal Indigenous Organisms (Prokaryotes) (2023)' after 2022 (Cho *et*

al., 2023). Building on prior studies focused on freshwater ecosystems (Joung *et al.*, 2018; 2019; Kim *et al.*, 2021), this research program has aimed to explore previously undiscovered bacterial species native to the islands of Korea. The ocean, enveloping about 70% of Earth's surface and holding 97% of the planet's water, hosts a diverse ecosystem, play a crucial role in Earth's system by contributing to nearly half of the net primary production, circulating carbon through the atmosphere and ocean, and forming the foundation of marine food webs (Field *et al.*, 1998; Sunagawa *et al.*, 2020). Considering that islands are surrounded by the sea and harbor distinctive terrestrial habitats, the coastal microbial community is anticipated to be a key determinant in shaping the distinctive ecosystem of each island.

As part of a research initiative supported by HNIBR, this study explores the isolation of previously unrecorded bacterial species from various island and coastal habitats. In 2023, these species were isolated from coastal seawater

samples obtained from six islands: Baengnyeongdo, Daebudo, Deokjeokdo, Jangbongdo, Somaemuldo, and Yeonpyeongdo Islands. Through 16S rRNA gene-based phylogenetic analyses, 18 bacterial strains spanning *Alphaproteobacteria*, *Gammaproteobacteria*, *Flavobacteriia*, and *Verrucomicrobiae* were identified as novel additions to bacterial repositories in Korea, accompanied by comprehensive taxonomic and phenotypic descriptions.

MATERIALS AND METHODS

In 2023, coastal seawater samples were collected from six Korea islands, encompassing Baengnyeongdo, Daebudo, Deokjeokdo, Jangbong, and Yeonpyeongdo Islands off the northwest coast, as well as Somaemuldo Island off the south coast. The collected samples were promptly refrigerated and transported to the laboratory within six hours for subsequent analysis.

The seawater samples were serially diluted, with 200 μ L from each dilution plated on three different types of media: marine agar 2216 (MA; Difco), R2A agar (Difco) prepared from aged seawater (M-R2A), and 1/10-diluted M-R2A agar. Following aerobic incubation for 3–7 days at 20–25°C, colonies were isolated and maintained on MA media. A total of 1,594 strains were isolated as single colonies, subsequently purified, and stored at –80°C in a 20% (v/v) glycerol suspension and lyophilized ampoules. The designation and isolation sources of each strain are provided in Table 1.

The examination of colony morphology was carried out using a magnifying glass on agar plates containing bacterial colonies at the stationary phase. Cellular characteristics, including size, shape, and flagella presence, were examined via transmission electron microscopy (CM200; Philips) after staining with 1% (w/v) uranyl acetate. Gram staining procedures were performed using a Gram-staining kit (bioMérieux) and the KOH method. Biochemical traits were assessed using the API 20NE kit (bioMérieux) in accordance with the manufacturer's instructions.

The strains were subjected to standard bacterial DNA extraction, PCR amplification, and 16S rRNA gene sequencing procedures to determine their phylogenetic position. Universal primers 27F and 1492R (Weisburg *et al.*, 1991) were used for 16S rRNA gene amplification, and Sanger sequencing was conducted with primers 518F and 805R. The resulting sequences were initially compared to those of other bacterial strains with validly published names using the EzTaxon-e server (Yoon *et al.*, 2017), with a 16S rRNA gene sequence similarity threshold of 98.7% for species demarcation (Chun *et al.*, 2018). Bacterial strains exhibiting a 16S rRNA gene sequence similarity of $\geq 98.7\%$ with species of validly published names, yet unreported in Korea, were categorized as unrecorded

bacterial species.

For phylogenetic analysis, the 16S rRNA gene sequences were aligned using the SILVA Incremental Aligner integrated into the ARB software (Pruesse *et al.*, 2007). Phylogenetic trees were then constructed using neighbor-joining, maximum-likelihood, and minimum-evolution algorithms, supported by bootstrap analyses consisting of 1,000 replicates. These analyses were conducted using MEGA 7 software (Kumar *et al.*, 2016) with pre-aligned sequences exported from ARB.

RESULTS AND DISCUSSION

Analysis of the 16S rRNA gene sequences of 1,594 bacterial strains revealed that a total of 18 strains meet the criteria for being classified as an unrecorded bacterial species in Korea. Comprehensive strain information, including isolation source and method, identification, taxonomic assignment from species to classes, and sequence accession numbers including HNIBR and GenBank data, is presented in detail in Table 1. The validation of the phylogenetic assignment of strains was achieved through the construction of phylogenetic trees, as illustrated in Fig. 1. All strains identified as unrecorded species formed a robust clade with the type strains of each corresponding species, supported by significant bootstrap values (Fig. 1).

The 18 unrecorded bacterial species exhibited phylogenetic variation across three phyla, four classes, nine orders, 13 families, and 18 genera, as detailed in Table 1. At the genera level, the unrecorded species were assigned to *Aliiroseovarius*, *Kiloniella*, *Maritalea*, *Palleronia*, and *Roseobacter* of the class *Alphaproteobacteria*; *Aliamphritea*, *Aliivibrio*, *Enterovibrio*, *Francisella*, *Leucothrix*, *Pseudoalteromonas*, *Psychrobium*, *Shewanella*, and *Vibrio* of the class *Gammaproteobacteria*; *Aquimarina*, *Pseudofulvibacter*, and *Tenacibaculum* of the class *Flavobacteriia*; and *Roseibacillus* of the class *Verrucomicrobiae*.

The 18 unrecorded bacterial species identified in this study are Gram-staining-negative, flagellated or non-flagellated, rod- or bacilli- or streptococci- or coccoid-shaped bacteria (Fig. 2). Further details regarding the morphological, physiological, and biochemical characteristics of these unrecorded bacterial species are provided in the following strain description sections.

Description of *Kiloniella laminariae* IMCC43645

Cells are Gram-stain-negative, aerobic, non-flagellated, and rod-shaped. Colonies are circular, convex, entire, and cream-colored after incubation for 3 days on MA at 25°C. Positive for nitrate reduction, indole production, esculin hydrolysis, and cytochrome oxidase; but negative for glucose fermentation, arginine dihydrolase, urease, gelatin hydrolysis, and β -galactosidase in API 20NE. D-glucose,

Table 1. Taxonomic affiliation of 18 bacterial strains isolated from seawater samples collected off the coast of Korean islands in 2023.

Class	Order	Family	Genus	Strain ID	HNIBR ID	Accession number	Closest species	16S rRNA Similarity	Isolation source/ Location	Isolation medium	Incubation conditions
Alphaproteobacteria	Kiloniellales	Kiloniellaceae	<i>Kiloniella</i>	IMCC43645	HNIBRBA6982	OR717403	<i>Kiloniella laminariae</i>	99.44	Seawater/Daebudo	M-R2A	25°C, 3 days
				IMCC44605	HNIBRBA6994	OR717416	<i>Maritalea porphyrae</i>	100	Seawater/Deokjeokdo	MA	20°C, 3 days
Rhodobacteriales	Rhodobacteraceae	Rhodobacteraceae	<i>Aliiroseovarius</i>	IMCC45350	HNIBRBA6979	OR717400	<i>Aliiroseovarius marinus</i>	99.13	Seawater/Tongyeong port	M-1/10R2A	25°C, 3 days
				IMCC44117	HNIBRBA6988	OR717409	<i>Roseobacter denitrificans</i>	100	Seawater/Yeonpyeongdo	M-R2A	20°C, 3 days
				IMCC43693	HNIBRBA6983	OR717404	<i>Palleronia raja</i>	99.78	Seawater/Daebudo	MA	25°C, 3 days
				IMCC44007	HNIBRBA6978	OR717399	<i>Pseudalteromonas aurantia</i>	99.86	Seawater/Yeonpyeongdo	MA	25°C, 3 days
Alteromonadales			<i>Psychrobium</i>	IMCC33692	HNIBRBA6987	OR717408	<i>Psychrobium conchae</i>	99.17	Seawater/Baengryeongdo	M-1/10R2A	20°C, 3 days
				IMCC44053	HNIBRBA6985	OR717406	<i>Shewanella chilikensis</i>	99.86	Seawater/Yeonpyeongdo	MA	20°C, 3 days
				IMCC44634	HNIBRBA6995	OR717417	<i>Aliamphritea spongicola</i>	99.79	Seawater/Jangbongdo	M-R2A	20°C, 3 days
Gammaproteobacteria	Thiotrichales	Francisellaceae	<i>Francisella</i>	IMCC44360	HNIBRBA6992	OR717414	<i>Francisella salimarina</i>	98.97	Seawater/Jangbongdo	MA	20°C, 3 days
				IMCC44228	HNIBRBA6991	OR717413	<i>Leucothrix mucor</i>	98.97	Seawater/Yeonpyeongdo	M-R2A	20°C, 3 days
Vibrionales	Vibrionaceae	Vibrionaceae	<i>Aliivibrio</i>	IMCC44015	HNIBRBA6984	OR717405	<i>Aliivibrio finisterrensis</i>	99.73	Seawater/Yeonpyeongdo	MA	20°C, 3 days
				IMCC44014	HNIBRBA6981	OR717402	<i>Enterovibrio calviensis</i>	98.97	Seawater/Yeonpyeongdo	MA	25°C, 3 days
				IMCC44158	HNIBRBA6990	OR717412	<i>Vibrio cortegadensis</i>	99.84	Seawater/Yeonpyeongdo	MA	20°C, 3 days
Flavobacteriia	Flavobacteriales	Flavobacteriaceae	<i>Aquimarina</i>	IMCC45041	HNIBRBA6980	OR717401	<i>Aquimarina megaterium</i>	99.58	Seawater/Somaemuldo	MA	25°C, 3 days
				IMCC45000	HNIBRBA6986	OR717407	<i>Pseudofulvibacter marinus</i>	100	Seawater/Tongyeong port	MA	25°C, 3 days
Verrucomicrobiae	Verrucomicrobiales	Verrucomicrobiaceae	<i>Roseobacillus</i>	IMCC44476	HNIBRBA6993	OR717415	<i>Tenacibaculum singaporensis</i>	98.99	Seawater/Jangbongdo	MA	20°C, 3 days
				IMCC44129	HNIBRBA6989	OR717411	<i>Roseobacillus persicus</i>	99.86	Seawater/Yeonpyeongdo	MA	20°C, 3 days

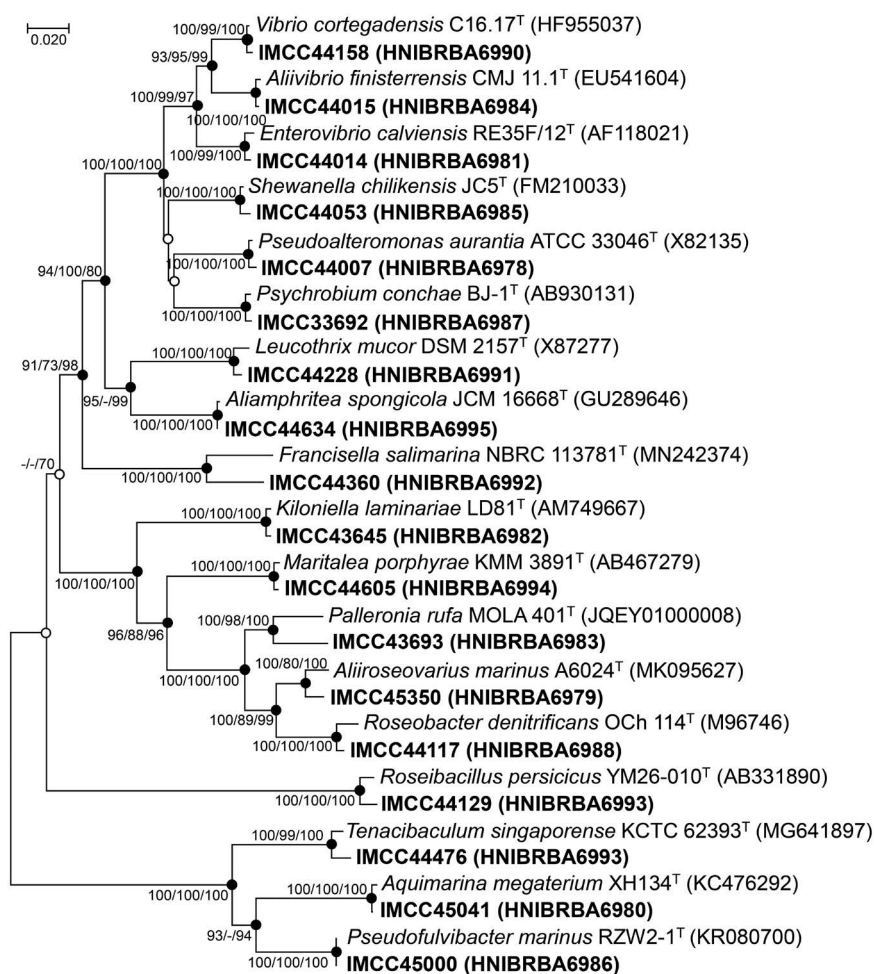


Fig. 1. The neighbor-joining (NJ) phylogenetic tree based on 16S rRNA gene sequences showing the relationship between the 18 strains isolated in this study and their closest bacterial species. Bootstrap values over 70% are shown at nodes for NJ, maximum-likelihood (ML), and minimum evolution (ME) methods, respectively (NJ/ML/ME; -, less than 70%). Filled circles indicate that the corresponding node was also recovered in the trees reconstructed with both the NJ and ME algorithms, while open circles indicate that the corresponding node was recovered in the tree generated with only one of these algorithms. Scale bar = 0.02 substitutions per nucleotide position.

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 sole carbon sources. Strain IMCC43645 (= HNIBRBA6982) was isolated from a coastal seawater sample, Nogari Beach in Daebudo Island, Ongjin-gun, Incheon, Korea.

Description of *Maritalea porphyrae* IMCC44605

Cells are Gram-stain-negative, aerobic, flagellated, and rod-shaped. Colonies are circular, convex, entire, and beige-colored after incubation for 3 days on MA at 20°C. Positive for nitrate reduction and β -galactosidase; but negative for indole production, glucose fermentation, arginine dihydrolase, urease, esculin hydrolysis, gelatin hydrolysis, and cytochrome oxidase in API 20NE. D-glucose, L-arab-

inose, 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 sole carbon sources. Strain IMCC44605 (= HNIBRBA6994) was isolated from a coastal seawater sample, Seopori Beach in Deokjeokdo Island, Ongjin-gun, Incheon, Korea.

Description of *Aliiroseovarius marinus* IMCC45350

Cells are Gram-stain-negative, aerobic, non-flagellated, and rod-shaped. Colonies are circular, raised, entire, and cream-colored after incubation for 3 days on MA at 25°C. Positive for esculin hydrolysis, and cytochrome oxidase; but negative for nitrate reduction, indole production, glucose fermentation, arginine dihydrolase, urease, gelatin hydrolysis and β -galactosidase in API 20NE. *N*-Acetyl-glucos-

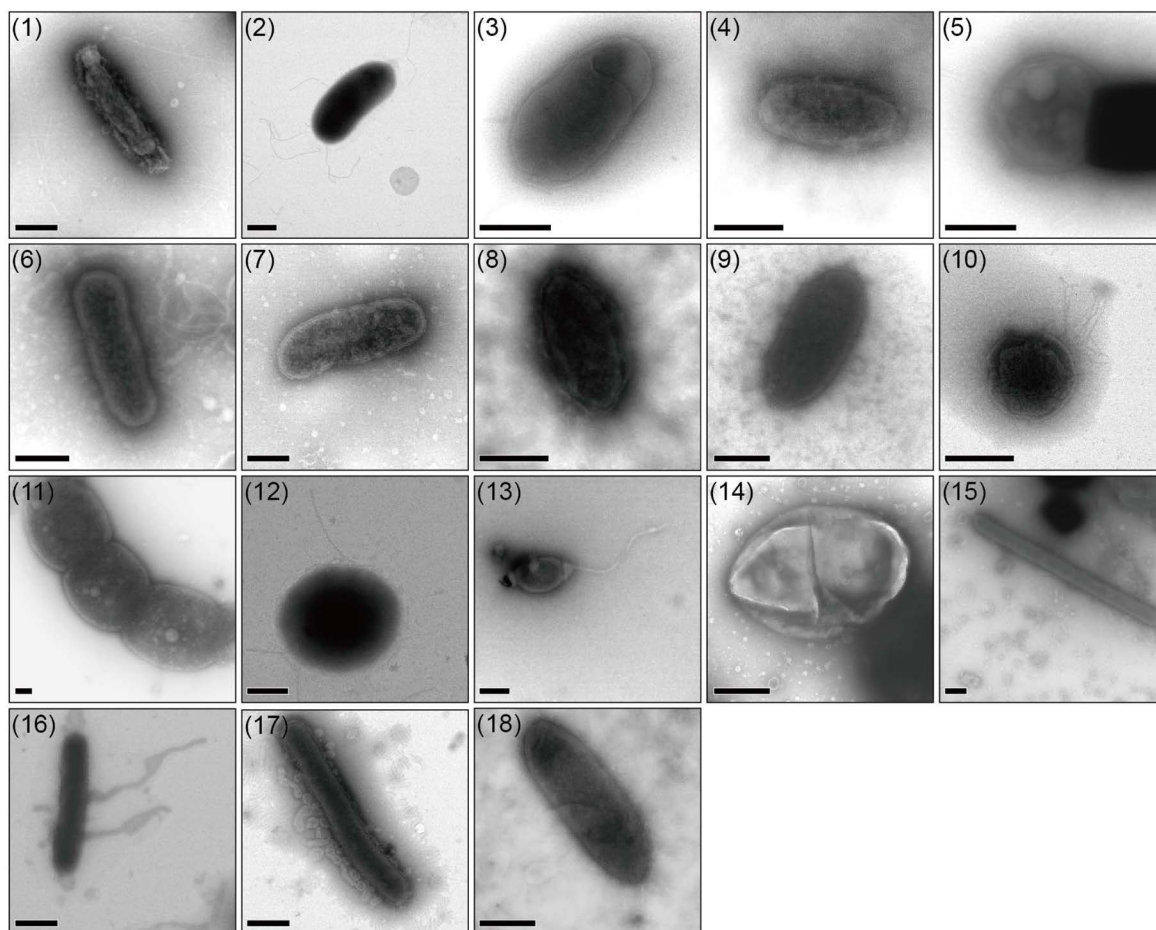


Fig. 2. Transmission electron micrographs of cells of the strains isolated in this study. Strains: 1, IMCC43645; 2, IMCC44605; 3, IMCC45350; 4, IMCC44117; 5, IMCC43693; 6, IMCC44007; 7, IMCC33692; 8, IMCC44053; 9, IMCC44634; 10, IMCC44360; 11, IMCC44228; 12, IMCC44015; 13, IMCC44014; 14, IMCC44158; 15, IMCC45041; 16, IMCC45000; 17, IMCC44476; 18, IMCC44129. Scale bars indicate 0.5 μm .

amine serves as the sole carbon source; whereas D-glucose, L-arabinose, D-mannose, D-mannitol, D-maltose, potassium gluconate, capric acid, adipic acid, malic acid, trisodium citrate, and phenylacetic acid are not utilized. Strain IMCC45350 (= HNIBRBA6979) was isolated from a coastal seawater sample, Tongyeong Port, Tongyeong, Gyeongsangnam-do, Korea.

Description of *Roseobacter denitrificans* IMCC44117

Cells are Gram-stain-negative, aerobic, non-flagellated, and rod-shaped. Colonies are circular, flat, entire, and pale pink-colored after incubation for 3 days on MA at 20°C. Positive for nitrate reduction, gelatin hydrolysis, and cytochrome oxidase; but negative for indole production, glucose fermentation, 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 sole carbon sources. Strain IMCC

acid, malic acid, trisodium citrate, and phenylacetic acid are not utilized as sole carbon sources. Strain IMCC44117 (= HNIBRBA6988) was isolated from a coastal seawater sample, Garaechilgi Beach in Yeonpyeongdo Island, Ongjin-gun, Incheon, Korea.

Description of *Palleronia rufa* IMCC43693

Cells are Gram-stain-negative, aerobic, non-flagellated, and coccoid-shaped. Colonies are circular, flat, entire, and caramel-colored after incubation for 3 days on MA at 25°C. Positive for urease, esculin hydrolysis, β -galactosidase, and cytochrome oxidase; but negative for nitrate 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 sole carbon sources. Strain IMCC

43693 (= HNIBRBA6983) was isolated from a coastal seawater sample, Bangahmeori Beach in Daebudo Island, Ongjin-gun, Incheon, Korea.

Description of *Pseudoalteromonas aurantia* IMCC44007

Cells are Gram-stain-negative, aerobic, non-flagellated, and rod-shaped. Colonies are circular, raised, undulate, translucent, and orange-colored after incubation for 3 days on MA at 25°C. Positive for glucose fermentation, esculin hydrolysis, gelatin hydrolysis, β -galactosidase, and cytochrome oxidase; but negative for nitrate reduction, indole production, arginine dihydrolase, and urease 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 sole carbon sources. Strain IMCC44007 (= HNIBRBA6978) was isolated from a coastal seawater sample, Guridong Beach in Yeonpyeongdo Island, Ongjin-gun, Incheon, Korea.

Description of *Psychrobium conchae* IMCC33692

Cells are Gram-stain-negative, aerobic, non-flagellated, and rod-shaped. Colonies are circular, convex, entire, glistening, opaque, and cream-colored after incubation for 3 days on MA at 20°C. Positive for glucose fermentation, esculin hydrolysis, β -galactosidase, and cytochrome oxidase; but negative for nitrate reduction, indole production, 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 sole carbon sources. Strain IMCC33692 (= HNIBRBA6987) was isolated from a coastal seawater sample, Gobong Port in Baengnyeongdo Island, Ongjin-gun, Incheon, Korea.

Description of *Shewanella chilikensis* IMCC44053

Cells are Gram-stain-negative, aerobic, flagellated, and rod-shaped. Colonies are circular, convex, entire, mucoid, opaque, and pale orange-colored after incubation for 3 days on MA at 20°C. Positive for nitrate reduction, gelatin hydrolysis, and cytochrome oxidase; but negative for indole production, glucose fermentation, arginine dihydrolase, urease, esculin hydrolysis, and β -galactosidase in API 20NE. *N*-Acetyl-glucosamine, capric acid, and malic acid serve as the sole carbon source, whereas D-glucose, L-arabinose, D-mannose, D-mannitol, D-maltose, potassium gluconate, adipic acid, trisodium citrate, and phenylacetic acid are not utilized. Strain IMCC44053 (= HNIBRBA6985) was isolated from a tidal flats sample in Yeonpyeongdo Island, Ongjin-gun, Incheon, Korea.

Description of *Aliamphritea spongicola* IMCC44634

Cells are Gram-stain-negative, aerobic, non-flagellated, and rod-shaped. Colonies are circular, raised, entire, brittle, and white-colored after incubation for 3 days on MA at 20°C. Positive for esculin hydrolysis, and β -galactosidase; but negative for nitrate reduction, indole production, glucose fermentation, arginine dihydrolase, urease, gelatin hydrolysis, 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 sole carbon sources. Strain IMCC 44634 (= HNIBRBA6995) was isolated from a coastal seawater sample, Geoneojang Beach in Jangbongdo Island, Ongjin-gun, Incheon, Korea.

Description of *Francisella salimarina* IMCC44360

Cells are Gram-stain-negative, aerobic, flagellated, and coccoid-shaped. Colonies are circular, flat, entire, translucent, and pale yellow-colored after incubation for 3 days on MA at 20°C. Negative for nitrate 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 sole carbon sources. Strain IMCC44360 (= HNIBRBA6992) was isolated from a coastal seawater sample, Jinchon Beach in Jangbongdo Island, Ongjin-gun, Incheon, Korea.

Description of *Leucothrix mucor* IMCC44228

Cells are Gram-stain-negative, aerobic, non-flagellated, and filamentous shaped. Colonies are circular, flat, entire, and white-colored after incubation for 3 days on MA at 20°C. Positive for nitrate reduction; but negative for 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 sole carbon sources. Strain IMCC44228 (= HNIBRBA6991) was isolated from a coastal seawater sample, Dangseom Port in Yeonpyeongdo Island, Ongjin-gun, Incheon, Korea.

Description of *Aliivibrio finisterrensis* IMCC44015

Cells are Gram-stain-negative, aerobic, flagellated, and coccoid-shaped. Colonies are circular, convex, entire, opaque, and pale yellow-colored after incubation for 3

days on MA at 20°C. Positive for nitrate reduction, glucose fermentation, and β -galactosidase; but negative for indole production, arginine dihydrolase, urease, esculin hydrolysis, gelatin hydrolysis, 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 sole carbon sources. Strain IMCC44015 (= HNIBRBA6984) was isolated from a coastal seawater sample, Guridong Beach in Yeonpyeongdo Island, Ongjin-gun, Incheon, Korea.

Description of *Enterovibrio calviensis* IMCC44014

Cells are Gram-stain-negative, aerobic, flagellated, and bacilli-shaped. Colonies are circular, flat, entire, opaque, and cream-colored after incubation for 3 days on MA at 25°C. Positive for nitrate reduction, indole production, arginine dihydrolase, esculin hydrolysis, β -galactosidase, and cytochrome oxidase; but negative for glucose fermentation, 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 sole carbon sources. Strain IMCC 44014 (= HNIBRBA6981) was isolated from a coastal seawater sample, Guridong Beach in Yeonpyeongdo Island, Ongjin-gun, Incheon, Korea.

Description of *Vibrio cortegadensis* IMCC44158

Cells are Gram-stain-negative, aerobic, non-flagellated, and rod-shaped. Colonies are circular, flat, entire, opaque, and cream-colored after incubation for 3 days on MA at 20°C. Positive for nitrate reduction, indole production, glucose fermentation, arginine dihydrolase, and cytochrome oxidase; but negative for 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 sole carbon sources. Strain IMCC 44158 (= HNIBRBA6990) was isolated from a coastal seawater sample, Dangseom Port in Yeonpyeongdo Island, Ongjin-gun, Incheon, Korea.

Description of *Aquimarina megaterium* IMCC45041

Cells are Gram-stain-negative, aerobic, non-flagellated, and rod-shaped. Colonies are circular, convex, entire, and orange-colored after incubation for 3 days on MA at 25°C. Positive for esculin hydrolysis, and cytochrome oxidase; but negative for nitrate reduction, indole production, glucose fermentation, arginine dihydrolase, urease, 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 sole carbon sources. Strain IMCC45041 (= HNIBRBA6980) was isolated from a coastal seawater sample, Somaemuldo Port, Tongyeong, Gyeongsangnam-do, Korea.

Description of *Pseudofulvibacter marinus* IMCC45000

Cells are Gram-stain-negative, aerobic, non-flagellated, and rod-shaped. Colonies are circular, convex, entire, opaque, and yellow-colored after incubation for 3 days on MA at 25°C. Positive for glucose fermentation, esculin hydrolysis, and cytochrome oxidase; but negative for nitrate reduction, indole production, arginine dihydrolase, urease, 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 sole carbon sources. Strain IMCC 45000 (= HNIBRBA6986) was isolated from a coastal seawater sample, Tongyeong Port, Tongyeong, Gyeongsangnam-do, Korea.

Description of *Tenacibaculum singaporense* IMCC44476

Cells are Gram-stain-negative, aerobic, non-flagellated, and rod-shaped. Colonies are circular, umbonate, entire, and yellow-colored after incubation for 3 days on MA at 20°C. Positive for gelatin hydrolysis, and cytochrome oxidase; but negative for nitrate reduction, indole production, glucose fermentation, 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 sole carbon sources. Strain IMCC44476 (= HNIBRBA6993) was isolated from a coastal seawater sample, Handle Beach in Jangbongdo Island, Ongjin-gun, Incheon, Korea.

Description of *Roseibacillus persicus* IMCC44129

Cells are Gram-stain-negative, aerobic, non-flagellated, and rod-shaped. Colonies are circular, convex, entire, and pink-colored after incubation for 3 days on MA at 20°C. Positive for esculin hydrolysis, β -galactosidase, and cytochrome oxidase; but negative for nitrate 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 sole carbon sources. Strain IMCC44129 (= HNIBRBA6989) was isolated from a coastal seawater sample, Garaechilgi Beach in Yeonpyeongdo Island, Ongjin-gun, Incheon, Korea.

CONFLICTS OF INTEREST

The author of this paper has no affiliation with any interests and is solely responsible for the paper.

ACKNOWLEDGEMENTS

This study was supported by the research grant “Survey of island-coastal indigenous organisms (Prokaryotes)” (HNIBR202301211) from Honam National Institute of Biological Resources (HNIBR) in Korea.

REFERENCES

- Armstrong, M. 2023. The Countries with the most islands. Statista Inc. [Available from <https://www.statista.com/chart/15364/the-estimated-number-of-islands-by-country/>, accessed 21 February 2024].
- Cho, H., Y. Lim, S. Kim, H. Jo, M. Kim and J.-C. Cho. 2023. A report of 20 unrecorded bacterial species isolated from the coastal area of Korean islands in 2022. *J. Species Res.* 12:165-173. <https://doi.org/10.12651/JSR.2023.12.2.165>
- Chun, J., A. Oren, A. Ventosa, H. Christensen, D.R. Arahal, M.S. da Costa, A.P. Rooney, H. Yi, X.-W. Xu, S. De Meyer and M.E. Trujillo. 2018. Proposed minimal standards for the use of genome data for the taxonomy of prokaryotes. *Int. J. Syst. Evol. Microbiol.* 68:461-466. <https://doi.org/10.1099/ijsem.0.002516>
- Field, C.B., M.J. Behrenfeld, J.T. Randerson and P. Falkowski. 1998. Primary production of the biosphere: integrating terrestrial and oceanic components. *Science* 281:237-240. <https://doi.org/10.1126/science.281.5374.237>
- Joung, Y., H.-J. Jang, M.W. Jung, J. Hwang, J. Song and J.-C. Cho. 2019. A report of 26 unrecorded bacterial species in Korea, isolated from urban streams of the Han River watershed in 2018. *J. Species Res.* 8:249-258. <https://doi.org/10.12651/JSR.2019.8.3.249>
- Joung, Y., M. Park, H.-J. Jang, I. Jung and J.-C. Cho. 2018. A report of 22 unrecorded bacterial species in Korea, isolated from the North Han River basin in 2017. *J. Species Res.* 7:193-201. <https://doi.org/10.12651/JSR.2018.7.3.193>
- Kim, M., J. Song, D. Yu, Y. Kim, S.H. Bae, M.S. Park, Y. Lim and J.-C. Cho. 2021. A report of 28 unrecorded bacterial species in Korea, isolated from freshwater and sediment of the Han River watershed in 2020. *J. Species Res.* 10:227-236. <https://doi.org/10.12651/JSR.2021.10.3.227>
- Kumar, S., G. Stecher and K. Tamura. 2016. MEGA7: molecular evolutionary genetics analysis version 7.0 for bigger datasets. *Mol. Biol. Evol.* 33:1870-1874. <https://doi.org/10.1093/molbev/msw054>
- Li, S.-P., P. Wang, Y. Chen, M.C. Wilson, X. Yang, C. Ma, J. Lu, X.-Y. Chen, J. Wu, W.-S. Shu and L. Jiang. 2020. Island biogeography of soil bacteria and fungi: similar patterns, but different mechanisms. *ISME J* 14:1886-1896. <https://doi.org/10.1038/s41396-020-0657-8>
- Losos, J.B. and R.E. Ricklefs. 2009. Adaptation and diversification on islands. *Nature* 457:830-836. <https://doi.org/10.1038/nature07893>
- Pruesse, E., C. Quast, K. Knittel, B.M. Fuchs, W. Ludwig, J. Peplies and F.O. Glöckner. 2007. SILVA: a comprehensive online resource for quality checked and aligned ribosomal RNA sequence data compatible with ARB. *Nucleic Acids Res.* 35:7188-7196. <https://doi.org/10.1093/nar/gkm864>
- Sunagawa, S., S.G. Acinas, P. Bork, C. Bowler, Tara Oceans Coordinators, D. Eveillard, G. Gorsky, L. Guidi, D. Iudicone, E. Karasenti, F. Lombard, H. Ogata, S. Pesant, M.B. Sullivan, P. Wincker and C. Vargas. 2020. Tara Oceans: towards global ocean ecosystems biology. *Nat. Rev. Microbiol.* 18:428-445. <https://doi.org/10.1038/s41579-020-0364-5>
- Weisburg, W.G., S.M. Barns, D.A. Pelletier and D.J. Lane. 1991. 16S ribosomal DNA amplification for phylogenetic study. *J. Bacteriol.* 173:697-703. <https://doi.org/10.1128/jb.173.2.697-703.1991>
- Yoon, S.H., S.M. Ha, S. Kwon, J. Lim, Y. Kim, H. Seo and J. Chun. 2017. Introducing EzBioCloud: a taxonomically united database of 16S rRNA gene sequences and whole-genome assemblies. *Int. J. Syst. Evol. Microbiol.* 67:1613-1617. <https://doi.org/10.1099/ijsem.0.001755>

Submitted: March 21, 2024

Revised: May 24, 2024

Accepted: June 11, 2024