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Acinetobacter marinus sp. nov. and *Acinetobacter seohaensis* sp. nov., Isolated from Sea Water of the Yellow Sea in Korea

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Abstract Two Gram-negative, nonmotile, coccobacilli, SW-3^T and SW-100^T, were isolated from sea water of the Yellow Sea in Korea. Strains SW-3^T and SW-100^T contained ubiquinone-9 (Q-9) as the predominant respiratory lipoquinone and $C_{18:1}$ ω9c and $C_{16:0}$ as the major fatty acids. The DNA G+C contents of strains SW-3^T and SW-100^T were 44.1 mol% and 41.9 mol%, respectively. A neighbor-joining tree based on 16S rRNA gene sequences showed that the two isolates fell within the evolutionary radiation enclosed by the genus *Acinetobacter*. Strains SW-3^T and SW-100^T exhibited a 16S rRNA gene similarity value of 95.7% and a mean DNA-DNA relatedness level of 9.2%. Strain SW-3^T exhibited 16S rRNA gene sequence similarity levels of 93.5–96.9% to the validly described *Acinetobacter* species and fifteen *Acinetobacter* genomic species. Strain SW-100^T exhibited 16S rRNA gene sequence similarity levels of less than 97.0% to the other *Acinetobacter* species except *Acinetobacter towneri* DSM 14962^T (98.0% similarity). Strains SW-3^T and SW-100^T exhibited mean levels of DNA-DNA relatedness of 7.3–16.7% to the type strains of some phylogenetically related *Acinetobacter* species. On the basis of phenotypic, phylogenetic, and genetic data, strains SW-3^T and SW-100^T were classified in the genus *Acinetobacter* as two distinct novel species, for which the names *Acinetobacter marinus* sp. nov. (type strain SW-3^T=KCTC 12259^T=DSM 16312^T) and *Acinetobacter seohaensis* sp. nov. (type strain SW-100^T=KCTC 12260^T=DSM 16313^T) are proposed, respectively.

Keywords: Marine bacteria, polyphasic taxonomy, *Acinetobacter marinus* sp. nov., *Acinetobacter seohaensis* sp. nov.

The first species of the genus *Acinetobacter* was proposed by Baumann *et al.* [3], and at present, the genus comprises seventeen species with validly published names, *Acinetobacter calcoaceticus* [3], *Acinetobacter baumannii*, *Acinetobacter haemolyticus*, *Acinetobacter johnsonii*, *Acinetobacter junii* [4], *Acinetobacter lwoffii* [4, 6], *Acinetobacter radioresistens* [18], *Acinetobacter schindleri*, *Acinetobacter ursingii* [16], *Acinetobacter baylyi*, *Acinetobacter bouvetii*, *Acinetobacter gerneri*, *Acinetobacter grimontii*, *Acinetobacter tandoii*, *Acinetobacter tjernbergiae*, *Acinetobacter towneri* [8], and *Acinetobacter parvus* [17]. In addition to species with the recognized names, there are many genomospecies within the genus *Acinetobacter* [4, 5, 12, 24]. *Acinetobacter* species are Gram-negative, nonmotile, strictly aerobic, and oxidase-negative. The genus *Acinetobacter* is phylogenetically related to the family *Moraxellaceae* of the *γ-Proteobacteria* [1, 19]. *Acinetobacter* species distribute widely in nature, including soil and water [3], sewage [25], human clinical specimens [16, 17], and activated sludge [8]. In this study, we describe two Gram-negative, slightly halophilic bacterial strains, SW-3^T and SW-100^T, isolated from a marine environment in Korea. The two isolates were considered to be *Acinetobacter*-like strains from 16S rRNA gene sequence comparisons. The aim of the present study was to determine the exact taxonomic positions of strains SW-3^T and SW-100^T by a polyphasic characterization that included phenotypic properties, detailed phylogenetic analysis based on 16S rRNA gene sequences, and genetic relatedness.

Materials and Methods

Bacterial Strains and Cultural Conditions

Sea water collected from the Yellow Sea, Korea, was used as the source for isolation of bacterial strains. Strains SW-3^T and SW-100^T were isolated by the usual dilution plating technique on marine agar 2216 (MA; Difco) at 30°C. *A. calcoaceticus* KCTC 2357^T was obtained from the Korean Collection for Type Cultures (KCTC), Daejeon, Korea. *A. baumannii* KCCM 40203^T, *A. junii* KCCM 40207^T, *A. radioresistens* KCCM 40171^T, and *A. lwoffii* KCCM 40172^T were obtained from the Korean Culture Center of Microorganisms (KCCM), Seoul, Korea. *A. johnsonii*

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LMG 999^T and *Acinetobacter* genomospecies TU13 (LMG 993) were obtained from the Laboratorium voor Microbiologie Universiteit Gent (LMG), Gent, Belgium. *Acinetobacter* genomospecies 10 (NCIMB 9019) and *Acinetobacter* genomospecies 11 (NCIMB 8250) were obtained from the National Collections of Industrial Food and Marine Bacteria (NCIMB), Aberdeen, United Kingdom. *A. towneri* DSM 14962^T was obtained from the Deutsche Sammlung von Mikroorganismen und Zellkulturen (DSMZ), Braunschweig, Germany. To investigate their morphological, physiological, and biochemical characteristics, strains SW-3^T and SW-100^T were routinely cultivated at 30°C on MA. The cell biomass of strains SW-3^T and SW-100^T for respiratory lipoquinone analysis and for DNA extraction was obtained from cultures grown in MB at 30°C. The cell mass of reference strains for DNA extraction was obtained from cultivation in nutrient broth (Difco) at 30°C. For fatty acid methyl ester analysis, the cell mass of strains SW-3^T and SW-100^T was harvested from agar plates after cultivation at 30°C for 2 days on nutrient agar (NA; Difco) and for 5 days on MA, and the cell mass of *A. calcoaceticus* KCTC 2357^T, *A. baumannii* KCCM 40203^T, *A. junii* KCCM

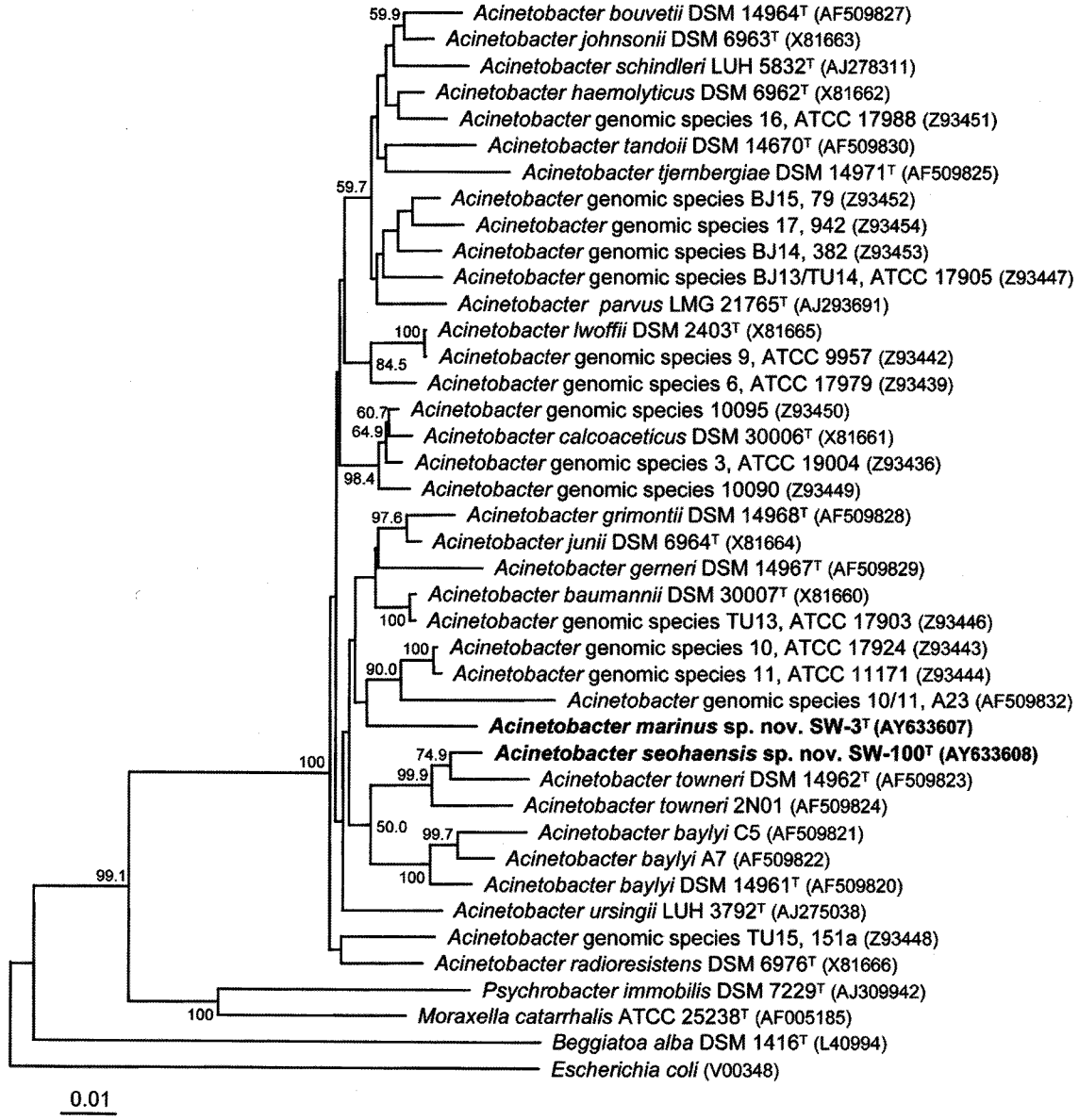

Fig. 1. Neighbourjoining tree based on 16S rRNA gene sequences showing the phylogenetic positions of strains SW-3^T and SW-100^T and the representatives of some other related taxa.

Bootstrap values (expressed as percentages of 1,000 replications) greater than 50% are shown at the branch points. Scale bar, 0.01 substitutions per nucleotide position.

40207^T, *A. radioresistens* KCCM 40171^T, and *A. lwoffii* KCCM 40172^T was harvested from agar plates after cultivation for 2 days on NA at 30°C.

Morphological and Physiological Characterization

The cell morphology was examined by light microscopy (Nikon E600) and transmission electron microscopy (TEM). The presence of flagella was examined by TEM using cells from exponentially growing cultures. The Gram reaction was determined by using the bioMérieux Gram Stain kit according to the manufacturer's instructions. The pH range for growth was determined after incubation for 3 days in marine broth 2216 (MB; Difco) that was adjusted to various pH values (initial pH 4.5–9.5 at intervals of 0.5 pH units). Growth at various NaCl concentrations was investigated after incubation for 3 days in MB or trypticase soy broth (Difco). Growth in the absence of NaCl was investigated after incubation for 7 days in trypticase soy broth lacking NaCl. Growth at various temperatures (4–45°C) was measured after incubation for at least 10 days on MB. Growth under anaerobic conditions was determined after incubation in an anaerobic chamber on MA that had been prepared anaerobically. Catalase and oxidase activities and hydrolysis of casein, starch, and Tweens 20, 40, 60, and 80 were determined as previously described [10]. Hydrolysis of hypoxanthine, tyrosine, and xanthine was performed on MA using the substrate concentrations described previously [10]. Hydrolysis of gelatin and aesculin and nitrate reduction were determined as previously described [14] with a modification that artificial seawater was used. The artificial seawater contained (in l^{-1} distilled water) 23.6 g NaCl, 0.64 g KCl, 4.53 g $MgCl_2 \cdot 6H_2O$, 5.94 g $MgSO_4 \cdot 7H_2O$, and 1.3 g $CaCl_2 \cdot 2H_2O$ [7]. Acid production from carbohydrates was determined as previously described [15]. Utilization of substrates as sole carbon and energy sources was tested according to the method of Baumann and Baumann [2], using supplementation with 2% (v/v) Hutner's mineral base [9] and 1% (v/v) of vitamin solution [22].

Chemosystematic Characterization

Isoprenoid quinones were extracted and analyzed as previously described [13] using reverse-phase HPLC. The fatty acid methyl esters were extracted and prepared according to the standard protocol of the MIDI/Hewlett Packard Microbial Identification System [20].

Molecular Systematics

Chromosomal DNA was isolated and purified according to the method described previously [27], except that ribonuclease T1 was used together with ribonuclease A. The DNA G+C content was determined by the method of Tamaoka and Komagata [23]. DNA was hydrolyzed and the resultant nucleotides were analyzed by reverse-phase HPLC. DNA-DNA hybridization was performed fluorometrically by the method of Ezaki *et al.* [11] using photobiotin-labeled DNA probes and microdilution wells. Hybridization was performed with five replications for each sample. The highest and lowest values obtained for each sample were excluded; the remaining three measurements were used to calculate the mean relatedness value. The 16S rRNA gene was amplified by PCR using two universal primers as previously described [28]. Sequencing of the amplified 16S rRNA gene and phylogenetic analysis were performed as previously described [29].

Nucleotide Sequence Accession Numbers

The GenBank/EMBL/DDBJ accession numbers for the 16S rRNA gene sequences of strains SW-3^T and SW-100^T are AY633607 and AY633608, respectively. The designations and 16S rRNA gene sequence accession numbers of the reference strains used in the phylogenetic analysis are shown in Fig. 1.

RESULTS

Phenotypic Characteristics

Cells of strains SW-3^T and SW-100^T were Gram-negative, coccobacilli, nonspore-forming, and nonmotile. Colonies of strain SW-3^T on MA are circular, smooth, glistening, slightly convex, milky-white in color, and 0.8–1.0 mm in diameter after 3 days of incubation at 30°C. Colonies of strain SW-3^T on NA are circular to slightly irregular, smooth, glistening, raised, cream-colored, and 2.0–4.0 mm in diameter after 3 days of incubation at 30°C. Colonies of strain SW-100^T are circular to slightly irregular, smooth, slightly raised, milky-white in color, and 0.8–1.0 mm in diameter after 3 days of incubation at 30°C on MA. Colonies of strain SW-100^T are circular to slightly irregular, smooth, raised to umbonate, cream-colored, and 2.0–3.0 mm in diameter after 3 days of incubation at 30°C on NA. The two strains grew at a temperature range of 10.0–40.0°C with an optimum temperature at 30–37°C. Optimal pH for growth is 6.0–8.0. Optimal growth occurs in the presence of 0–2% (w/v) NaCl. Strains SW-3^T and SW-100^T were similar in most phenotypic characteristics. Minor differential characteristics of the two strains were as follows; strain SW-3^T grew at pH 5.0, but strain SW-100^T did not at pH 5.0. Strain SW-100^T did not grow in the presence of more than 4% (w/v) NaCl, but strain SW-3^T did not in the presence of more than 8% (w/v) NaCl. Strain SW-100^T utilized 2,3-butanediol as carbon and energy sources, but strain SW-3^T did not. Morphological, cultural, physiological, and biochemical characteristics of strains SW-3^T and SW-100^T are shown in Table 1 or are given in the species description (see below).

Table 1. Phenotypic characteristics of *A. marinus* sp. nov, *A. seohaensis* sp. nov., and some phylogenetically related *Acinetobacter* species.

Characteristic	1	2	3	4	5	6	7	8	9	10	11	12
Growth at 41°C	–	–	+	+	+	v(+)	+	+	–	–	ND	–
Growth at 44°C	–	–	–	–	–	–	+	–	–	–	–	–
Utilization of*												
Citrate	–	–	–	+	+	v(+)	+	+	+	+	+	+
DL-Lactate	+	+	+	ND	ND	+	+	ND	+	+	ND	+
L-Phenylalanine	–	–	–	–	–	–	v(+)	+	–	–	–	+
Phenylacetate	–	–	–	+	–	–	v(+)	+	v	v	–	+
Malonate	–	–	v(+)	+	–	–	v(+)	–	–	–	–	+
L-Histidine	–	–	–	v(–)	+	+	v(+)	–	+	+	+	+
Azelate	–	–	–	+	–	–	v(+)	+	v	v	+	+
D-Malate	–	–	–	ND	ND	+	v(+)	ND	+	+	ND	–
L-Aspartate	–	–	–	+	–	v(+)	+	–	+	v	+	+
L-Leucine	–	–	–	–	–	v(+)	v(+)	–	–	–	–	v(–)
Histamine	–	–	ND	ND	ND	–	–	ND	v	+	ND	–
L-Tyrosine	–	–	ND	ND	ND	v(+)	+	ND	+	v	ND	+
β-Alanine	–	–	–	–	–	–	v(–)	+	+	+	+	+
Ethanol	+	+	ND	ND	ND	+	+	ND	+	+	ND	+
2,3-Butanediol	-	+	ND	ND	ND	–	+	ND	+	+	ND	+
trans-Aconitate	–	–	–	+	–	–	v(+)	–	–	–	+	+
L-Arginine	–	–	–	+	–	v(+)	v(+)	–	–	–	–	+
L-Ornithine	–	–	ND	ND	ND	–	v(–)	ND	–	–	ND	+
DL-4-Aminobutyrate	–	–	–	+	+	v(+)	+	+	+	+	+	+
Isolation source	Sea water	Sea water	Activated sludge	Activated sludge	Activated sludge	Human clinical specimen	Human clinical specimen or natural environment	Activated sludge	Human clinical specimen	Human and animal specimens	Activated sludge	Soil

Species: 1, *A. marinus* sp. nov.; 2, *A. seohaensis* sp. nov.; 3, *A. towneri*, data from Carr *et al.* [8]; 4, *A. baylyi*, data from Carr *et al.* [8]; 5, *A. grimontii*, data from Carr *et al.* [8]; 6, *A. junii*, data from Bouvet and Grimont [4]; 7, *A. baumannii*, data from Bouvet and Grimont [4]; 8, *A. gerneri*, data from from Carr *et al.* [8]; 9, Genomic species 10, data from Bouvet and Grimont [4]; 10, Genomic species 11, data from Bouvet and Grimont [4]; 11, Genomic species A23 (DSM 14960); 12, *A. calcoaceticus*, data from Bouvet and Grimont [4]. +, positive; –, negative; v, variable; ND, not determined; Data in parentheses are for the type strain. All species are Gram-negative and strictly aerobic. Tests positive for all species: catalase, utilization of L-malate and pyruvate. Tests negative for all species: oxidase, hydrolysis of gelatin, and utilization of D-galactose, sucrose, and D-trehalose.

*Utilization of L-malate, pyruvate, and D-galactose was not determined for *A. junii*, *A. baumannii*, and *A. calcoaceticus*.

Table 2. Percentage cellular fatty acid composition of *A. marinus* sp. nov. SW-3^T and *A. seohaensis* sp. nov. SW-100^T on NA and on MA and the type strains of some *Acinetobacter* species on NA.

Fatty acid[a]	1	2	3	4	5	6	7	8	9
Straight-chain fatty acid									
$C_{10:0}$	1.7	1.6	0.3	0.3	0.1	0.1	2.1	0.4	0.5
$C_{12:0}$	7.9	8.5	9.2	9.5	4.0	7.1	4.0	12.1	8.6
$C_{14:0}$	1.0	1.1	1.0	1.0	0.7	0.6	0.9	0.6	1.3
$C_{15:0}$	-	-	-	-	0.9	0.6	1.1	0.2	-
$C_{16:0}$	21.5	13.2	23.6	21.1	19.4	20.6	12.5	18.9	16.9
$C_{16:0}$ N alcohol	-	-	-	0.3	1.3	-	-	-	-
$C_{17:0}$	-	0.5	-	-	2.2	1.6	3.6	1.0	-
$C_{18:0}$	2.2	1.6	1.0	0.6	2.8	1.4	1.9	1.0	1.0
Unsaturated fatty acid									
$C_{16:1}$ ω9c	0.8	1.7	-	0.9	-	1.1	0.8	1.2	-
$C_{16:1}$ ω7c alcohol	-	-	-	-	1.6	-	-	-	-
$C_{17:1}$ ω8c	-	0.6	-	-	3.5	1.6	2.5	1.5	-
$C_{18:1}$ ω7c	6.0	0.7	6.0	0.9	5.6	0.6	0.6	2.0	4.2
$C_{18:1}$ ω9c	38.3	52.8	35.9	42.7	29.3	44.6	33.2	39.5	23.4
Branched fatty acid									
iso-$C_{17:0}$	-	-	-	-	1.6	0.1	-	0.3	-
Hydroxy fatty acid									
$C_{12:0}$ 2-OH	-	-	0.2	0.2	1.3	2.5	5.8	0.2	0.2
$C_{12:0}$ 3-OH	4.9	6.0	4.5	5.4	2.6	3.9	7.6	6.0	6.2
Summed feature[b]									
2	-	0.6	0.2	0.5	1.7	3.7	0.4	1.4	0.8
3	15.1	10.5	17.6	15.2	18.7	8.4	20.8	13.3	36.4

Strain: 1, strain SW-3^T (NA); 2, strain SW-3^T (MA); 3, strain SW-100^T (NA); 4, strain SW-100^T (MA); 5, *A. calcoaceticus* KCTC 2357^T; 6, *A. baumannii* KCCM 40203^T; 7, *A. junii* KCCM 40207^T; 8, *A. radioresistens* KCCM 40171^T; 9, *A. lwoffii* KCCM 40172^T.

[a]Fatty acids representing less than 1.0% in all rows were omitted.

[b]Summed features represent groups of two or three fatty acids that could not be separated by GLC with the MIDI system. Summed feature 2 contained one or more of iso-$C_{16:1}$ I and/or $C_{14:0}$ 3-OH. Summed feature 3 contained one or more of $C_{16:1}$ w7c and/or iso-$C_{15:0}$ 2-OH.

Chemosystematic Characteristics

The predominant isoprenoid quinone found in strains SW-3^T and SW-100^T was ubiquinone-9 (Q-9) and minor amounts of ubiquinone-8 and ubiquinone-10 were present. The major respiratory lipoquinone of the type strain of *A. calcoaceticus* (KCTC 2357^T), the type species of the genus, detected in this study was ubiquinone-9 (Q-9). Strains SW-3^T and SW-100^T had cellular fatty acid profiles that contained large amounts of straight-chain, unsaturated, and hydroxy fatty acids; the major fatty acids were $C_{18:1}$ ω9c and $C_{16:0}$ (Table 2). There were differences in the proportions of some fatty acids when the two strains were cultivated separately on NA and on MA (Table 2). The cellular fatty acid profiles of strains SW-3^T and SW-100^T were similar to those of some *Acinetobacter* species, although there were differences in the proportions of some fatty acids (Table 2). However, on the basis of the difference in the fatty acid profiles, it may be difficult to differentiate those *Acinetobacter* species as well as strains SW-3^T and SW-100^T. The DNA G+C contents of strains SW-3^T and SW-100^T were 44.1 and 41.9 mol%, respectively.

Phylogenetic Analysis

Almost complete 16S rRNA gene sequences of strains SW-3^T and SW-100^T comprising 1,493 nt (approx. 96% of the *Escherichia coli* 16S rRNA gene sequence) were determined in this study. The 16S rRNA gene sequences of strains SW-3^T and SW-100^T were 95.7% similar. Comparative 16S rRNA gene sequence analyses showed that the two strains are phylogenetically closely related to *Acinetobacter* species of the *Moraxellaceae*. In the phylogenetic tree based on neighbor-joining algorithm, strains SW-3^T and SW-100^T fell within the radiation of the cluster comprising *Acinetobacter* species (Fig. 1). Strain SW-100^T exhibited the highest 16S rRNA gene sequence similarity value to *A. towneri* DSM 14962^T (98.0%); this relationship was supported by a bootstrap resampling value of 100% (Fig. 1). Strain SW-100^T exhibited 16S rRNA gene sequence similarity levels of 94.6–96.6% to the type strains of the other *Acinetobacter* species. Levels of 16S rRNA gene sequence similarity between strain SW-3^T and the type strains of all validly published *Acinetobacter* species ranged from 94.4% (with *A. calcoaceticus*) to 96.5% (with *A. grimontii*). Strains SW-3^T and SW-100^T exhibited 16S rRNA gene

similarity levels of 93.5–96.9% to the fifteen *Acinetobacter* genomic species that were shown in the study of Carr *et al.* [8].

DNA-DNA Relatedness

DNA-DNA hybridization was performed to determine the genomic relatedness between strains SW-3^T and SW-100^T and between the two strains and the type strains of some phylogenetically related *Acinetobacter* species and *Acinetobacter* genomospecies. The mean DNA-DNA relatedness value between strains SW-3^T and SW-100^T was 9.2%, indicating that the two strains are members of two different genomic species [26]. Strains SW-3^T and SW-100^T exhibited mean levels of DNA-DNA relatedness of 7.3–13.4% and 8.5–16.7%, respectively, to the following reference strains; *A. calcoaceticus* KCTC 2357^T, *A. baumannii* KCCM 40203^T, *A. junii* KCCM 40207^T, *A. radioresistens* KCCM 40171^T, *A. lwoffii* KCCM 40172^T, *A. johnsonii* LMG 999^T, *A. towneri* DSM 14962^T, *Acinetobacter* genomospecies TU13 (LMG 993), *Acinetobacter* genomospecies 10 (NCIMB 9019), and *Acinetobacter* genomospecies 11 (NCIMB 8250).

DISCUSSION

The 16S rRNA gene sequence analyses revealed that strains SW-3^T and SW-100^T have the closest phylogenetic affiliations to the genus *Acinetobacter* (Fig. 1). However, chemotaxonomic data, including respiratory lipoquinone and cellular fatty acid, have scarcely been known for members of the genus *Acinetobacter*. The respiratory lipoquinone of strains SW-3^T and SW-100^T was the same to that of the type strain of *A. calcoaceticus*, the type species of the genus *Acinetobacter*, although they were not compared with those of the other *Acinetobacter* species. The fatty acid profiles of the two isolates were similar to those of the type strains of some *Acinetobacter* species. Strains SW-3^T and SW-100^T were similar in most phenotypic properties, but they differed phylogenetically and genetically. The two isolates were differentiated from some phylogenetically related *Acinetobacter* species in some phenotypic characteristics (Table 1). The phylogenetic and genetic discriminations were sufficient to categorize strains SW-3^T and SW-100^T as two species that are distinct from *Acinetobacter* species with validly published names [21, 26]. Therefore, on the basis of the data presented, strains SW-3^T and SW-100^T should be classified in the genus *Acinetobacter* as two distinct novel species, for which the names *Acinetobacter marinus* sp. nov. and *Acinetobacter seohaensis* sp. nov. are proposed, respectively. To our knowledge, these two species are the first reported of novel *Acinetobacter* species that were isolated from a marine environment, although many *Acinetobacter* strains have been isolated from marine environments without detailed identification.

Description of *Acinetobacter marinus* sp. nov.

Acinetobacter marinus (ma.rin'us. L. masc. adj. *marinus* of the sea, marine).

Cells are coccobacilli, 0.8–1.1×1.5–2.0 μm on MA. Nonspore-forming. Nonmotile. Colonies on MA are circular, smooth, glistening, slightly convex, milky-white in color, and 0.8–1.0 mm in diameter after 3 days of incubation at 30°C. Colonies on NA are circular to slightly irregular, smooth, glistening, raised, cream-colored, and 2.0–4.0 mm in diameter after 3 days of incubation at 30°C. Optimal growth temperature is 30–37°C; growth occurs at 10 and 40°C, but not at 4°C and above 41°C. Optimal pH for growth is 6.0–8.0. Growth is observed at pH 5.0, but not at pH 4.5. Optimal growth occurs in the presence of 0–2% (w/v) NaCl. No growth occurs in the presence of more than 8% (w/v) NaCl. Growth does not occur under anaerobic conditions on MA. Tweens 20, 40, 60, and 80 are hydrolyzed. Aesculin, casein, hypoxanthine, starch, tyrosine, urea, and xanthine are not hydrolyzed. Nitrate is not reduced. Acetate, benzoate, and succinate are utilized as carbon and energy sources; L-arabinose, D-cellobiose, D-glucose, D-fructose, lactose, maltose, D-mannose, D-xylose, L-glutamate, and formate are not. The predominant ubiquinone is Q-9. The major fatty acids are $C_{18:1}$ ω9c and $C_{16:0}$. The DNA G+C content is 44.1 mol% (determined by HPLC). Other characteristics are given in Table 1. Isolated from the sea water of Wando-gun of the Yellow Sea in Korea. The type strain is strain SW-3^T (=KCTC 12259^T=DSM 16312^T).

Description of *Acinetobacter seohaensis* sp. nov.

Acinetobacter seohaensis (seo.ha.en'sis. N.L. fem. adj. *seohaensis* of Seohae, the Korean name of the Yellow Sea in Korea where the organism was isolated).

Cells are coccobacilli, 1.0–1.2×1.5–2.0 μm on MA. Nonspore-forming. Nonmotile. Colonies on MA are circular to slightly irregular, smooth, slightly raised, milky-white in color and 0.8–1.0 mm in diameter after 3 days of incubation at 30°C. Colonies on NA are circular to slightly irregular, smooth, raised to umbonate, cream-colored, and 2.0–3.0 mm in diameter after 3 days of incubation at 30°C. Optimal growth temperature is 30–37°C; growth occurs at 10 and 40°C, but not at 4°C and above 41°C. Optimal pH for growth is 6.0–8.0. Growth is observed at pH 5.5, but not at pH 5.0. Optimal growth occurs in the presence of 0–2% (w/v) NaCl. No growth occurs in the presence of more than 4% (w/v) NaCl. Growth does not occur under anaerobic conditions on MA. Tweens 20, 40, 60, and 80 are hydrolyzed. Aesculin, casein, hypoxanthine, starch, tyrosine, urea, and xanthine are not hydrolyzed. Nitrate is not reduced. Acetate, benzoate, and succinate are utilized

as carbon and energy sources; L-arabinose, D-cellobiose, D-glucose, D-fructose, lactose, maltose, D-mannose, D-xylose, L-glutamate, and formate are not. The predominant ubiquinone is Q-9. The major fatty acids are $C_{18:1}$ ω9c and $C_{16:0}$. The DNA G+C content is 41.9 mol% (determined by HPLC). Other characteristics are given in Table 1. Isolated from a sea water of Wando-gun of the Yellow Sea in Korea. The type strain is strain SW-100^T (=KCTC 12260^T =DSM 16313^T).

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