**Introduction**

Bacteria inhabiting the marine environment include halophiles and non-halophiles. Only the halophiles are considered to be autochthonous organisms, i.e. true marine bacteria. The non-halophilic ones tolerate the salinity of seawater but are not considered to be of marine origin since they do not require Na⁺ for growth and are always found in a non-saline environment. Most halophilic marine bacteria are either mesophilic or psychrophilic Gram-negative rods that require 70–700 mM Na⁺ for optimal growth and yield in laboratory media (Baumann & Baumann, 1971). A large proportion of these bacteria are facultative anaerobes that can ferment glucose and other carbohydrates for anaerobic growth. Marine bacteria of this type are currently placed in the genera *Vibrio* (Baumann et al., 1984), *Photobacterium* (Baumann & Baumann, 1984), *Listonella* (MacDonell & Colwell, 1985) and *Colwellia* (Deming et al., 1988) of the family *Vibrionaceae* (Baumann & Schubert, 1984). These bacteria generally constitute 10–50% of heterotrophic bacteria from coastal and oceanic seawater samples that grow on ordinary plate media used in marine bacteriology (Simidu & Tsukamoto, 1985). They are also found to be closely associated with many kinds of marine organisms from plankton to fish (Cerdà-Cuéllar et al., 1997; Liston, 1956; MacDonald et al., 1986; Nair et al., 1988; Onarheim et al., 1994; Simidu et al., 1969, 1971; Sochard et al., 1979; Yoshimizu et al., 1976). Some species are found as symbionts in specialized luminous organs of marine fish and invertebrates (Lee & Ruby, 1994; Leisman et al., 1980; McFall-Ngai & Ruby, 1991; Reichelt et al., 1977; Ruby & Asato, 1993; Ruby & Morin, 1978), whereas quite a few other species are well-known pathogens for humans or marine animals (Blake et al., 1980; Egidus et al., 1986; Hada et al., 1984; Holt et al., 1994; Love et al., 1981; Schiewe et al., 1981). These halophilic, facultatively anaerobic, Gram-negative rods are presently known to include both nitrogen fixers and...
denitrifiers (Guerinot & Colwell, 1985; Guerinot et al., 1982; Shieh & Lin, 1992, 1994; Shieh & Liu, 1996; Shieh & Yang, 1997; Shieh et al., 1989, 1990; West et al., 1985), although Bergey's Manual of Systematic Bacteriology defined in 1984 that they include neither of the two (Baumann & Baumann, 1984; Baumann & Schubert, 1984; Baumann et al., 1984).

Production of gas during glucose fermentation is a characteristic commonly found among the non-halophilic, facultatively anaerobic, Gram-negative rods belonging to either Enterobacteriaceae (Brenner, 1984) or Aeromonadaceae (Colwell et al., 1986). It is, therefore, intriguing why the halophilic, facultatively anaerobic, Gram-negative rods that also possess this property are not prevalent. Vibrio gazogenes, Vibrio furnissii (formerly Vibrio fluvialis biovar 2), Vibrio mytili, Photobacterium histaminum, Photobacterium ilipiscarium, Photobacterium phosphoreum and Listonella damsela (reclassified as Photobacterium damsela subsp. damsela) are the only cases among the more than forty validly described species of these bacteria that produce gas during the fermentation of glucose (Baumann & Baumann, 1984; Baumann & Schubert, 1984; Baumann et al., 1984; Brenner et al., 1983; Holt et al., 1994; Okuzumi et al., 1994; Onarheim et al., 1994; Pujalte et al., 1993; Urakawa et al., 1999).

A new strain of halophilic, facultatively anaerobic, Gram-negative rod has been recovered in this laboratory from a sediment sample collected in a seagrass bed in Nanwan Bay, Kenting National Park, Taiwan. The present study shows that the new strain can achieve anaerobic growth by fermenting glucose and other carbohydrates with the production of various organic acids and H₂ and CO₂. Evidence presented in the study also shows that the strain represents a new species of Vibrio, for which the name Vibrio aerogenes is proposed.

**METHODS**

**Culture media.** Polypeptone-yeast (PY) broth contained the following ingredients (l⁻¹ denitized water): 3 g Polypeptone (Nihon Seiyaku); 1 g Bacto yeast extract (Difco); 25 g NaCl; and 5 g MgCl₂·6H₂O. The medium was adjusted to pH 7.8. Bacto agar (Difco) was added to this medium at 3.5 and 15 g l⁻¹, for the preparation of stab and plate media, respectively. Polypeptone-yeast-nitrate (PYN) broth was prepared by adding KNO₃ at 0.1 g l⁻¹ to PY broth. Tryptone broth contained 10 g tryptone (Sigma), 25 g NaCl and 5 g MgCl₂·6H₂O dissolved in 1000 ml denitized water and adjusted to pH 7.8. Polypeptone-yeast-carbohydrate (PYC) stab media were prepared from two parts. The first contained 3 g polypeptone, 1 g Bacto yeast extract, 25 g NaCl, 5 g MgCl₂·6H₂O, 0.24 g Tris (Sigma), 0.03 g bromothymol blue and 10 g Bacto agar dissolved in 900 ml denitized water and adjusted to pH 7.8. The second contained 5 g glucose (or any carbohydrate) dissolved in 100 ml denitized water. The two parts were autoclaved separately and mixed at about 50 °C. Glucose-mineral (GM) medium was also made up of two parts. Part 1 contained 0.54 g NH₄Cl, 25 g NaCl, 2 g MgCl₂·6H₂O, 3 g K₂SO₄, 0.2 g K₂HPO₄, 0.01 g CaCl₂, 0.005 g FeCl₃·6H₂O and 3 g (ca. 25 mmol) Tris dissolved in 900 ml denitized water and adjusted to pH 8.0, while part 2 contained 5 g glucose dissolved in 100 ml denitized water. The two parts were autoclaved separately and mixed at room temperature. However, GM-II medium differed from GM medium in that part 1 contained 5.4 g l⁻¹ (ca. 25 mM) 3-(N-morpholino)-2-hydroxypropanesulfonic acid (MOPSO; Sigma) instead of Tris and was adjusted to pH 7.0. A modified PY plate medium containing CaCl₂ (0.1 g l⁻¹) and Tween 80 (0.1%) was used for the lipase test. Four other modified PY plate media containing casein (4 g l⁻¹), DNA (2 g l⁻¹), gelatin (5 g l⁻¹) and starch (5 g l⁻¹), respectively, were also used to test the hydrolysis of these substrates. In addition, three stab media modified from Thorneley's semi-solid arginine medium (Smibert & Krieg, 1994) were used for the tests of arginine dehydrogenase, and lysine and ornithine decarboxylases. They contained the following ingredients (1 l⁻¹ denitized water): 2 g Bacto peptone (Difco); 25 g NaCl; 5 g MgCl₂·6H₂O; 0.01 g phenol red; 3 g Bacto agar (Difco); and 5 g l-arginine, 5 g l-lysine or 5 g l-ornithine. All three media were adjusted to pH 7.0. All the pH adjustments of the above-mentioned media were made with HCl (10 M), KOH (10 M) or both.

**Bacterial isolation.** Nanwan Bay of Kenting National Park is located on the south coast of Taiwan, where fringing reefs are widely distributed. Thalassia hemprichii and Halodule uninevris are two seagrass species found to co-habit in a seagrass bed of this bay. Non-rhizosphere sediment samples were collected from the seagrass bed in the morning at low tide. Some wet mass (2 g) of each sediment sample was vigorously shaken in sterile NaCl-Tris buffer (30 g NaCl and 0.24 g Tris in 1 l denitized water, pH 8.0) containing Tween 80 (2 p.p.m.), and the shaken solutions were decimally diluted with the same buffer. A volume (1 ml) of each dilution (10⁻⁵–10⁻⁷ times) was transferred to a minisealless tube (16 mm x 10 cm) containing GM medium (5 ml) in which an inverted Durham insert had been placed. All culture tubes were set in anaerobic jars (Difco) and incubated at 25 °C in the dark for 7 d. The tubes were aerated with Ar for 7 d, after which the cultures were aerated with Ar before incubation. Those cultures that developed visible turbidity and produced gas (accumulated in Durham inserts) were streaked (one loopful) on PY plate medium and the plates were incubated at 25 °C in the dark for 2–3 d under air. Individual colonies appearing on the plates were picked off and purified by successive streaking on PY plates. One bacterial strain, capable of growth with gas production in GM medium under Ar, was isolated for the present study using the above-mentioned procedures. This bacterium was designated as strain FG1¹ and its PY stab cultures were kept at 20 °C under aerobic conditions for maintenance.

**Bacterial growth.** Inoculum cultures of strain FG1¹ were routinely grown in PY broth. Late exponential to early stationary phase cultures (one loopful) were inoculated into tubes containing 5 ml of the test media to determine the effects of various factors on growth. The cultures were statically incubated under either aerobic or anaerobic conditions. Anaerobic conditions were developed by flushing the culture tubes for more than 5 min with Ar gas of 99.9% purity. The tubes were sealed with rubber stoppers that were pierced with two hypodermic needles; one needle was connected to the pipeline of Ar gas for flushing while the other was used for exhausting. Each tube was shaken vigorously on a mixer during the flushing process, after which the needles were pulled out of the stoppers. The OD₅₆₀ of each culture was measured using a Spectronic 20

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W. Y. Shieh and others
growth was determined in modified GM-II media containing various compounds as sole carbon and energy sources for GM-II medium containing 0–10% of NaCl. Utilization of growth around the discs were noted after incubating the broth culture (0...

Antibiotic susceptibility. Susceptibility to various antibiotics was determined with standard 6 mm discs (Difco). The discs were placed on PY plate medium that had been spread with broth culture (0.1 ml) of strain FG1\(^{T}\). Inhibition zones of growth around the discs were noted after incubating the plates at 25 °C for 24–30 h. They were interpreted as either susceptible or resistant with reference to standard data (NCCLS, 1990).

DNA base composition. This determination essentially followed the procedure described by Shieh & Liu (1996).

Cellular fatty acids. Fatty acids in cells from early stationary phase cultures grown in GM-II medium were extracted, saponified and methylated according to Suutari et al. (1990). GLC analysis of the fatty acid methyl esters was performed on a GC-14A (Shimadzu) equipped with an FID and a fused silica capillary column (Shieh & Jean, 1998).

Fermentation products. Strain FG1\(^{T}\) was anaerobically cultivated in GM-II medium under Ar. Cultures grown for 24–60 h were centrifuged to remove the cells. The supernatant samples, after filtering through a Nucleopore membrane (pore size, 0.22 μm), were analysed for organic acids produced during fermentative growth of the cultures by an HPLC equipped with an Interaction Ion-300 column (Shieh & Jean, 1998). Production of CO\(_2\) and H\(_2\) gases in the headspace of the GM-II culture system was, meanwhile, detected by GC using a GC-14A equipped with a thermal-conductivity detector and with either connected columns of Porapak Q and N (both 3 mm × 2 m; for CO\(_2\) analysis) or a column of molecular Sieve 5A (3 mm × 2 m; for H\(_2\) analysis) at 70 °C. Production of acetoin or butanediol in GM-II cultures was determined by the Voges-Proskauer reaction (Smibert & Krieg, 1994).

16S rRNA gene sequencing and phylogenetic dendrogram construction. A method similar to that of Hiraishi (1992) was used for PCR amplification and sequencing of the 16S rRNA gene of strain FG1\(^{T}\). The 16S rDNA sequence was aligned manually with those of reference bacterial strains available in the GenBank database. The aligned positions from 33 to 1398 (by Escherichia coli sequence J01859 numbering) that were unambiguous and available for the sequences used for the phylogenetic analysis. Distance matrices were calculated with the PHYLP program package using the Jukes & Cantor (1969) model by assuming a transition/transversion ratio of 2:0; a neighbour-joining phylogenetic dendrogram was then constructed according to the estimated evolutionary distances (Felsenstein, 1989).

Nucleotide sequence accession numbers. The accession numbers for the sequences used to construct the phylogenetic dendrogram are as follows: *Vibrio aestuarii* ATCC 35048\(^T\), X74689; *Vibrio alginolyticus* ATCC 17749\(^T\), X74690; *Vibrio anguillarum* ATCC 19264\(^T\), X16895; *Vibrio campellii* ATCC 25920\(^T\), X74692; *Vibrio carlidiarum* ATCC 35084\(^T\), X74693; *Vibrio choleae* ATCC 14035\(^T\), X74695; *Vibrio cincinnatiensis* ATCC 35912\(^T\), X74698; *Vibrio dialuticus* HE800\(^T\), X99762; *Vibrio diazotrophicus* ATCC 33466\(^T\), X74701; *Vibrio fischeri* ATCC 7744\(^T\), X74702; *Vibrio fluvialis* NCTC 11327\(^T\), X76335; *Vibrio furnissii* ATCC 35016\(^T\), X74704; *Vibrio gazogenes* ATCC 29988\(^T\), X74705; *Vibrio halotolerans* IAM 14596\(^T\), X74690; *Vibrio harveyi* ATCC 14126\(^T\), X74706; *Vibrio hollisae* ATCC 33564\(^T\), X74707; *Vibrio logei* ATCC 15832, X74708; *Vibrio mediterranei* CIP 103203\(^T\), X74710; *Vibrio metschnikowii* NCTC 11170, X74712; *Vibrio mimicus* ATCC 33653\(^T\), X74713; *Vibrio mytili* CECT 632\(^T\), X99761; *Vibrio natriegens* ATCC 14048\(^T\), X74714; *Vibrio navarrensis* CIP 103381\(^T\), X74715; *Vibrio nereis* ATCC 25917\(^T\), X74716; *Vibrio nigripulchritudo* ATCC 27043\(^T\), X74717; *Vibrio ordalii* ATCC 33509\(^T\), X74718; *Vibrio parahaemolyticus* ATCC 70069\(^T\), X74719; *Vibrio proteolyticus* ATCC 33658\(^T\), X74720; *Vibrio salmonicida* ATCC 33657\(^T\), X74721; *Vibrio mimicus* ATCC 336553, X74722; *Vibrio parahaemolyticus* ATCC 70068\(^T\), X74723; *Vibrio natriegens* ATCC 14048\(^T\), X74724; *Vibrio natriegens* ATCC 14048\(^T\), X74725; *Vibrio mimicus* ATCC 33653, X74726; *Vibrio natriegens* ATCC 14048\(^T\), X74727; *Vibrio natriegens* ATCC 14048\(^T\), X74728; *Vibrio natriegens* ATCC 14048\(^T\), X74729; *Vibrio natriegens* ATCC 14048\(^T\), X74730.
RESULTS

Bacterial growth

Strain FG1 grew over a pH range of 5.0–8.5, with optimal growth at about pH 7.0. The strain grew significantly over a temperature range of 20–35 °C, and most rapidly at 30–35 °C. Growth was weak at 15 and 38 °C, weak or negligible at 40 °C and was not observed at all at 5 and 42 °C. Both aerobic and anaerobic growth in GM-II medium were accompanied by a large decrease in medium pH (ca. 2 pH units) during the exponential phase of growth (Fig. 1). Fig. 2 shows the effect of NaCl on growth. Strain FG1 grew aerobically in GM-II medium containing 1–6% NaCl (ca. 0±17–1±0 M); growth was most rapid at 4% NaCl (ca. 0±68 M) and absent at 0%. Substitution of KCl (2–5%) for NaCl did not support growth (not shown).

Bacterial characterization

Strain FG1 was Gram-negative according to the staining and KOH testing methods. It produced flat, circular, off-white colonies on PY plate medium after 1–2 d incubation. Cells in late exponential to early stationary phase of growth in PY broth culture were straight or slightly curved, motile rods that were 0.6–0.8 µm in diameter by 2–3 µm in length. Electron microscopy revealed that they normally possessed two sheathed flagella at one pole of the cell (Fig. 3). Strain FG1 was halophilic and unable to grow in the absence of NaCl. It fermented glucose and other carbohydrates with production of gas. Catalase was positive but oxidase was negative. The strain did not accumulate any appreciable amount of PHB. It was not susceptible to the vibriostatic agent O129 at 10–150 µg. Additional biochemical and physiological characteristics and the ability to utilize various compounds as sole carbon or nitrogen sources are given in the description of Vibrio aerogenes sp. nov. given below.

Fermentation products

Anaerobic fermentative growth of strain FG1 in GM-II medium resulted in formation of a variety of organic acids and the gases CO2 and H2. Acetate and lactate generally constituted more than 70 mol% of the organic acid products. Other organic acids that have been detected included formate, malate, oxaloacetate, propionate, pyruvate and succinate. No evidence indicated the production of acetoin or butanediol in these fermentative cultures.

Antibiotic susceptibility

Strain FG1 was susceptible to ampicillin, chloramphenicol, colistin, gentamicin, nalidixic acid, polymyxin B and streptomycin but resistant to the other test antibiotics (Table 1).

Cellular fatty acids and DNA base composition

Strain FG1 contained C12:0 as the most abundant cellular fatty acid (50.9–75.3 mol%). The other cellular fatty acids present at levels greater than 3 mol%
Fig. 3. (a) Electron micrograph of strain FG1\(^T\) showing two thick flagella at one pole of the rod-shaped cell. Scale bar, 1 \(\mu\)m. (b) Two sheathed polar flagella on one cell of strain FG1\(^T\). One flagellum has lost the distal part of its sheath exposing the inner core. Scale bar, 0.5 \(\mu\)m.

included C11:0 (46–62 mol%), C14:0 (6.5–14.6 mol%), C16:0 (5.1–9.4 mol%), iso-C16:0 (3.5–8.2 mol%) and C18:1\(^\Delta\) (7.0–10.6 mol%).

Strain FG1\(^T\) had a G+C content of 45.9 mol%.

16S rDNA-based phylogenetic analysis

An almost complete 16S rDNA sequence (ca. 89%; estimated by comparison with the E. coli sequence J01859) of strain FG1\(^T\) was obtained. Phylogenetic analysis based on the 16S rDNA sequence comparisons showed that the strain is a member of the genus *Vibrio* in the gamma subclass of the Proteobacteria. The dendrogram in Fig. 4 shows the phylogenetic position of strain FG1\(^T\) within the radiation of the genus *Vibrio* and some related bacteria. The levels of 16S rDNA sequence similarity between strain FG1\(^T\) and the known *Vibrio* species were never greater than 96.2% (not shown); the highest similarity level found (96.2%) was observed with *V. mytili*.

**DISCUSSION**

The mesophilic strain FG1\(^T\) could be categorized as a slightly halophilic bacterium (Kushner & Kamekura, 1988) since it required NaCl for growth with optimum growth at 4.0% (ca. 0.51–0.68 M). The requirement for NaCl could not be substituted by KCl, indicating that the strain required Na\(^+\) and not Cl\(^-\) for growth and it also showed that the Na\(^+\) requirement was not for osmotic function.

Strain FG1\(^T\) grew significantly in GM-II medium under both aerobic and anaerobic conditions, which indicated that the strain is a facultative anaerobe and does not require vitamins or other organic growth factors. The anaerobic growth in GM-II medium was accompanied by a remarkable decrease in medium pH (ca. 2 pH units) during the exponential phase of growth, regardless of the large buffer content (25 mM MOPSO) in the medium. This indicated that the strain achieved anaerobic growth in the medium by fermenting glucose with the production of considerable amounts of acids. Detection of various organic acids including acetate, lactate, succinate, formate, malate, oxaloacetate, propionate and succinate from the anaerobic GM-II cultures further indicated that strain FG1\(^T\) is a mixed acid fermenter. Similar acidification occurring in the aerobic GM-II cultures during bacterial growth was possibly due to the creation of an oxygen-depleted microenvironment in such static cultures.

**Table 1. Susceptibility of strain FG1\(^T\) to various antibiotics**

<table>
<thead>
<tr>
<th>Antibiotic</th>
<th>Disc content</th>
<th>Inhibition zone diameter (mm)</th>
<th>Reaction*</th>
</tr>
</thead>
<tbody>
<tr>
<td>Ampicillin</td>
<td>10 (\mu)g</td>
<td>25</td>
<td>S</td>
</tr>
<tr>
<td>Chloramphenicol</td>
<td>30 (\mu)g</td>
<td>30</td>
<td>S</td>
</tr>
<tr>
<td>Clindamycin</td>
<td>2 (\mu)g</td>
<td>6</td>
<td>R</td>
</tr>
<tr>
<td>Colistin</td>
<td>10 (\mu)g</td>
<td>20</td>
<td>S</td>
</tr>
<tr>
<td>Erythromycin</td>
<td>15 (\mu)g</td>
<td>12</td>
<td>R</td>
</tr>
<tr>
<td>Gentamicin</td>
<td>10 (\mu)g</td>
<td>24</td>
<td>S</td>
</tr>
<tr>
<td>Kanamycin</td>
<td>30 (\mu)g</td>
<td>13</td>
<td>R</td>
</tr>
<tr>
<td>Lincomycin</td>
<td>5 (\mu)g</td>
<td>6</td>
<td>R</td>
</tr>
<tr>
<td>Nalidixic acid</td>
<td>30 (\mu)g</td>
<td>20</td>
<td>S</td>
</tr>
<tr>
<td>Novobiocin</td>
<td>30 (\mu)g</td>
<td>12</td>
<td>R</td>
</tr>
<tr>
<td>Penicillin G</td>
<td>10 U</td>
<td>15</td>
<td>R</td>
</tr>
<tr>
<td>Polymyxin B</td>
<td>300 U</td>
<td>20</td>
<td>S</td>
</tr>
<tr>
<td>Streptomycin</td>
<td>10 (\mu)g</td>
<td>18.5</td>
<td>S</td>
</tr>
<tr>
<td>Tetracycline</td>
<td>30 (\mu)g</td>
<td>12</td>
<td>R</td>
</tr>
</tbody>
</table>

*S, susceptible; R, resistant.
Strain FG\textsuperscript{T} is characterized as a mesophilic, slightly halophilic, facultatively anaerobic, Gram-negative rod that is motile by sheathed polar flagella and unable to accumulate PHB, indicating that the strain is probably a species of \textit{Vibrio} in the family \textit{Vibrionaceae} (Baumann & Schubert, 1984; Baumann et al., 1984). A 16S rDNA-based phylogenetic analysis has revealed that the sequence of strain FG\textsuperscript{T} indeed falls inside the cluster made up of the representative sequences of the known \textit{Vibrio} species (Fig. 4). Data analysis also supports the establishment of a new species since the levels of 16S rDNA sequence similarity between strain FG\textsuperscript{T} and all known species of \textit{Vibrio} are never greater than 96.2\%.

Thus, the name \textit{Vibrio aerogenes} is proposed for this new bacterium. \textit{V. aerogenes} possesses some phenotypic characteristics quite uncommon among the vibrios, including production of gas from glucose, two polar flagella on each cell, resistance to O\textsubscript{129} and a negative oxidase reaction (Holt et al., 1994). \textit{V. gazogenes} and \textit{V. metschnikovii} are the only two \textit{Vibrio} species previously reported to be negative for the oxidase reaction. Only the former produces gas from glucose. However, \textit{V. gazogenes} is not resistant to O\textsubscript{129} and has only single polar flagellum on each cell (Baumann et al., 1984; Harwood, 1978). Moreover, it produces red to orange colonies on plate media. Colonies produced by \textit{V. aerogenes}, on the other hand, are off-white. More detailed characteristics useful for

**Fig. 4.** 16S rDNA-based phylogenetic dendrogram showing the position of strain FG\textsuperscript{T} within the radiation of the \textit{Vibrio} species and some reference bacteria. Scale bar, evolutionary distance (\textit{K}_\text{sub}) of 0.01.
**Table 2.** Phenotypic characteristics useful for differentiating *Vibrio aerogenes* FG1T from previously described oxidase-negative and/or gas-producing species in the family *Vibrionaceae*


<table>
<thead>
<tr>
<th>Characteristic</th>
<th>1</th>
<th>2</th>
<th>3</th>
<th>4</th>
<th>5</th>
<th>6</th>
<th>7</th>
<th>8</th>
<th>9</th>
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<tbody>
<tr>
<td>Gas from glucose</td>
<td>+</td>
<td>+</td>
<td>+</td>
<td>+</td>
<td>-</td>
<td>+</td>
<td>+</td>
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<tr>
<td>Oxidase</td>
<td>-</td>
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<td>-</td>
<td>+</td>
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<td>+</td>
<td>ND</td>
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<td>+</td>
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<td>O/129 sensitivity (150 µg)</td>
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<td>+</td>
<td>+</td>
<td>+</td>
<td>+</td>
<td>+</td>
<td>ND</td>
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<tr>
<td>Red–orange pigment</td>
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<td>+</td>
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<td>+</td>
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<td>PHB accumulation</td>
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<td>Nitrate reduction</td>
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<td>+</td>
<td>ND</td>
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<tr>
<td>Sheathed polar flagella</td>
<td>+</td>
<td>+</td>
<td>ND</td>
<td>+</td>
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<td>Amylase</td>
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<td>+</td>
<td>ND</td>
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<td>Gelatinase</td>
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<tr>
<td>Lipase</td>
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<td>ND</td>
<td>+</td>
<td>ND</td>
<td>ND</td>
<td>-</td>
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<tr>
<td>Arginine dihydrolase</td>
<td>+</td>
<td>-</td>
<td>+</td>
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<td>+</td>
<td>ND</td>
<td>ND</td>
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<tr>
<td>Growth at:</td>
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<td>4 °C</td>
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<tr>
<td>35 °C</td>
<td>+</td>
<td>+</td>
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<td>+</td>
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<td>-</td>
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<tr>
<td>Growth in 10% NaCl</td>
<td>-</td>
<td>+</td>
<td>-</td>
<td>+</td>
<td>-</td>
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<td>Cellobiose</td>
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<td>Sucrose</td>
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<td>Mannose</td>
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<td>Xylose</td>
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<td>Mannitol</td>
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<td>Inositol</td>
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<td>Citrate</td>
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differentiating *V. aerogenes* from *V. gazogenes* and other oxidase-negative or gas-producing species in the family *Vibrionaceae* are listed in Table 2.

*V. aerogenes* is presently found only in the marine sediment. However, the requirement for NaCl and a moderate temperature indicates that this species may also occur in shallow coastal and oceanic seaways.

**Description of *Vibrio aerogenes* sp. nov.**

*Vibrio aerogenes* {a.e.ro’ge.nes. Gr. masc. n. aer air; Gr. v. gennanio to produce; M.L. adj. aerogenes gas-producing).

Straight to slightly curved rod. Cells stain Gram-negative and are 0.6–0.8 µm wide by 2–3 µm long. Swarming does not occur on solid agar media while motility normally occurs by means of two sheathed, polar flagella when grown in liquid media. Colonies produced on agar media are flat, circular, off-white and non-luminescent with an entire margin. The species is a facultative anaerobe capable of both aerobic and anaerobic fermentative growth. Acid and gas are produced from fermentation of glucose and other carbohydrates such as cellobiose, galactose, inositol, mannitol, mannose, sucrose and xylose. However, d-arabinose, dulcitol, lactose, sorbitol and trehalose are not fermented. Voges–Proskauer reaction is negative. Accumulation of PHB inside the cells is not observed. Catalase, arginine dihydrolase, amylase, caseinase, DNase, gelatinase and lipase are positive while oxidase, agarase, and lysine and ornithine decarboxylases are negative. Indole is produced. Nitrate is reduced to nitrite but not further to N2O or N2. The species is mesophilic, growing well at 20–35 °C but not at all at 4 or 42 °C. It is able to grow in a mineral medium containing glucose and ammonium or nitrate salts. It grows at NaCl levels of 1–7% with optimal growth at about 4% while no growth occurs at 0 and 10% NaCl. It is resistant to the vibriostatic agent O/129 (10 or 150 µg per disc). The most abundant cellular fatty acid is C12:0. The following compounds are utilized as sole carbon and energy sources: cellobiose, galactose, glucose, sucrose, mannose, xylose, citrate, fumarate, pyruvate, inositol, mannitol, l-alanine, l-aspartate, l-glutamate and l-threonine. The following compounds
are not utilized as sole carbon and energy sources: D-arabinose, lactose, melibiose, trehalose, acetate, β-hydroxybutyrate, DL-malate, malonate, tartrate, adonitol, dulcitol, sorbitol, L-arginine, L-glycine, L-lysine, L-methionine, L-ornithine, L-phenylalanine, L-tryptophan and L-valine. The species utilizes ammonium, nitrate and the L-form amino acids alanine, arginine, aspartate, glutamate, lysine, methionine, ornithine, phenylalanine, threonine, tryptophan and valine as sole nitrogen sources for growth. The DNA G+C composition is about 46–0 mol%. The species is found in coastal sediment. The type strain is FG1T, which has been deposited in the American Type Culture Collection as strain ATCC 700797 and in the Culture Center for Research and Collection, Taiwan as strain CCRC 17041T.

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REFERENCES


