Arcobacter anaerophilus sp. nov., isolated from an estuarine sediment and emended description of the genus Arcobacter

T. S. Sasi Jyothsna,¹ K. Rahul,² E. V. V. Ramaprasad,² Ch. Sasikala² and Ch. V. Ramana¹

¹Department of Plant Sciences, School of Life Science, University of Hyderabad, Hyderabad-500046, India
²Bacterial Discovery laboratory, Centre for Environment, Institute of Science & Technology, Jawaharlal Nehru Technological University Hyderabad, Hyderabad-500085, India

Two strains (JC83, JC84ᵀ) of obligately anaerobic, H₂S-producing bacteria were isolated from estuarine sediment samples collected from Gangasagar, West Bengal, India. Cells were Gram-stain-negative, non-motile rods. Both strains were positive for oxidase, negative for catalase, hydrolysed casein, reduced nitrate and utilized citrate. Both strains grew chemoorganoheterotrophically with optimal pH of 7–8 (range 7–10) and at 30 °C (range 25–37 °C). C₁₆:₁ω7c, C₁₈:₁ω7c, C₁₈:₀ and C₁₂:₀ were the major fatty acids of both strains with minor amounts of C₁₄:₀, C₁₂:₀ 3-OH and C₁₈:₀. Polar lipids of both strains included diphosphatidylglycerol, phosphatidylethanolamine, phosphatidylglycerol, phosphatidylserine, phosphatidylcholine, phosphatidylinositol, an unidentified aminolipid (AL2), an unidentified phospholipid (PL2) and an unidentified lipid (L3). MK-6 was the major respiratory quinone. The DNA G+C content of strains JC83 and JC84ᵀ was 25.0 and 24.6 mol%, respectively. The strains showed DNA reassociation >85% (86.0 ± 0.5%) (based on DNA–DNA hybridization). Based on 16S rRNA gene sequence analysis, both strains were identified as belonging to the family Campylobacteraceae of the class Epsilonproteobacteria with Arcobacter marinus CL-S1ᵀ (95.4% sequence similarity) as their closest phylogenetic neighbour. On the basis of morphological, physiological and chemotaxonomic characteristics as well as phylogenetic analysis, strains JC83 and JC84ᵀ are considered to represent a novel species, for which the name Arcobacter anaerophilus sp. nov. is proposed. The type strain is JC84ᵀ (=KCTC 15071ᵀ=MTCC 10956ᵀ=DSM 24636ᵀ). An emended description of the genus Arcobacter is provided.

Abbreviations: DPG diphosphatidylglycerol; ME, minimum-evolution; MP, maximum-parsimony; NJ, neighbour-joining; PC, phosphatidycholine; PE, phosphatidylethanolamine; PG, phosphatidylglycerol; PI, phosphatidylinositol; PS, phosphatidylserine.

The GenBank/EMBL/DDJB accession numbers for the 16S rRNA, gyrB, rpoB and hsp60 gene sequences of strain JC84ᵀ are FR686494, HE609033, HF968432 and HF968431, respectively.

One supplementary table and four supplementary figures are available with the online version of this paper.

Tidal flat sediments are the most productive coastal marine ecosystems (Alongi, 1998) and are characterized by intense heterotrophic and phototrophic activity (Poremba et al., 1999). As a consequence, high microbiological activity and microbial diversity is expected in these sediments (Webster 1999). As a consequence, high microbiological activity and microbial diversity is expected in these sediments (Webster 1999). As a consequence, high microbiological activity and microbial diversity is expected in these sediments (Webster 1999). As a consequence, high microbiological activity and microbial diversity is expected in these sediments (Webster 1999). As a consequence, high microbiological activity and microbial diversity is expected in these sediments (Webster 1999). As a consequence, high microbiological activity and microbial diversity is expected in these sediments (Webster 1999). As a consequence, high microbiological activity and microbial diversity is expected in these sediments (Webster 1999). As a consequence, high microbiological activity and microbial diversity is expected in these sediments (Webster 1999). As a consequence, high microbiological activity and microbial diversity is expected in these sediments (Webster 1999). As a consequence, high microbiological activity and microbial diversity is expected in these sediments (Webster 1999). As a consequence, high microbiological activity and microbial diversity is expected in these sediments (Webster 1999).

At the time of writing, the genus Arcobacter (Vandamme et al., 2005) comprised 17 recognized species, eight of which have been described recently: Arcobacter defluvii (Collado et al., 2011), Arcobacter trophiarum (De Smet et al., 2011), Arcobacter molluscorum (Figuera et al., 2011a), Arcobacter ellisii (Figuera et al., 2011b), Arcobacter bivalviorum and Arcobacter venerupis (Levican et al., 2012), ‘Arcobacter cloacaee’ and ‘Arcobacter suis’ (Levican et al., 2013). Members of the genus Arcobacter were isolated from various environments, (Jørgensen, 1982). Hence, an attempt was made to isolate sulfate-reducing bacteria from the tidal sediment samples of Gangasagar, India. Gangasagar, which is located in the Sundarbans (a mangrove ecosystem) of West Bengal, is an estuary where the river Ganges meets the Bay of Bengal. The two new isolates belonging to Campylobacteraceae reported in this communication were isolated from these samples. The family Campylobacteraceae comprises three genera, namely Arcobacter, Campylobacter and Sulfurospirillum.
many from plant and animal associations (Vandamme et al., 1991; Figueras et al., 2011a, b; Levican et al., 2012). All reported members of the genus Arcobacter are motile by means of a single polar flagellum and are capable of growing under microaerobic conditions. Arcobacter butzleri, Arcobacter skirrowii, Arcobacter cryaerophilus, Arcobacter nitrofigilis (Vandamme et al., 1992) and Arcobacter halophilus (Donachie et al., 2005) show anaerobic growth, Arcobacter cibarius (Houf et al., 2005), Arcobacter mytili (Collado et al., 2009) and Arcobacter defluvii (Collado et al., 2011) show weak or poor growth, while Arcobacter marinus (Kim et al., 2010) cannot grow under anaerobic conditions. In this communication, we report two novel members of the genus Arcobacter, strains JC83 and JC84T, isolated from two estuarine sediment samples collected from Gangasagar, Kolkata, West Bengal, India (GPS position of the sample collection site is 21° 88’2” N 088° 164° E).

The initial enrichment for strains JC83 and JC84T was carried out in Postgate’s C broth (Postgate, 1984) supplemented with lactate (0.35 %, w/v) as the carbon source, NH4Cl (0.1 %, w/v) as the nitrogen source and FeSO4 .7 H2O (0.05 %, w/v) as the electron acceptor in fully filled screw cap test tubes (10 × 100 mm) and incubated at 28 ± 2 °C. Black coloured enrichments were obtained after 10 days of incubation, indicating the presence of sulfate-reducing bacteria. Purification of the bacteria was achieved by repeated streaking of the culture on agar slants in the above medium sealed with suba-seal and flushed with argon gas to maintain anaerobic conditions (Lakshmi et al., 2011a, b). After 5 days of incubation, small, round transparent colonies were observed. The colonies initially formed a black zone around them and the whole medium turned black within the next 2 days of growth. Both strains grew well in nitrate medium containing (per litre) beef extract (3 g), peptone (5 g) and KNO3 (1 g) under anaerobic conditions. The colonies on nitrate medium were tiny, round and pale yellow. As the strains could not withstand either refrigeration at 4 °C or lyophilization, pure cultures were maintained by frequent subculturing in broth or in agar slants under anaerobic conditions with subculturing done every 15 days in nitrate medium.

Genomic DNA was extracted and purified from strains JC83 and JC84T according to the method of Marmur (1961) and the G+C content of the DNA was 25.0 and 24.6 mol%, respectively, as determined by HPLC (Mesbah et al., 1989). Cell material for 16S rRNA gene sequencing was taken from a colony. DNA was extracted and purified by using a Qiagen Cell material for 16S rRNA gene sequencing was taken from a colony. DNA was extracted and purified by using a Qiagen DNA extraction kit. Recombinant Tag polymeuser (Genei) was used for PCR, which was carried out using primers F27 (5’-GTTTGATCCTGGCAG-3’) and R1489 (5’-TACCTTGTTACGACTTCA-3’) [positions 11–27 and 1489–1506, respectively, according to the Escherichia coli 16S rRNA gene numbering system of the International Union of Biochemistry (Brosius et al., 1978; Lane et al., 1985)]. PCR amplification was performed as described by Imhoff & Pfennig (2001) and Imhoff et al. (1998). 16S rRNA gene sequencing was performed using the BigDye Terminator v1.1 Sequencing kit (Applied Biosystems) in a 3730-DNA-Analyser (Applied Biosystems) as specified by the manufacturer. For sequencing, the primers F27, F790 (5’-GATAACCTTGAGTCC-3’) and R1489 were used. Identification of phylogenetic neighbours and calculation of pairwise 16S rRNA gene sequence similarity were achieved using the EzTaxon-e server (Kim et al., 2012). The CLUSTAL W algorithm of MEGA 5.2 was used for sequence alignments and MEGA 5.2 (Tamura et al., 2011) software was used for phylogenetic analysis of the individual sequences. Distances were calculated by using the Kimura correction in a pairwise deletion manner (Kimura, 1980). Neighbour-joining (NJ), maximum-parsimony (MP) and minimum-evolution (ME) methods in the MEGA 5.2 software were used to reconstruct phylogenetic trees. Percentage support values were obtained using a bootstrap procedure.

The results of phylogenetic analysis of the 16S rRNA gene sequences (~1390 bp) suggested that strains JC83 and JC84T were monophylectic with members of the genus Arcobacter including the type species, but they took up the most diverse phylogenetic position within the family Campylobacteraceae [the NJ tree is shown as Fig. 1; ME and MP trees had similar topologies (data not shown); the NJ tree including the type strains of additional species is shown in Fig. S1, available in IJSEM Online], and the sequence similarities with the nearest phylogenetic members are in agreement with the EzTaxon-e server result. The 16S rRNA gene sequence similarity between strains JC83 and JC84T was 99.6%. EzTaxon-e server search analysis revealed that both strains (JC83 and JC84T) were related most closely to members of the genus Arcobacter; highest sequence similarity was observed with A. marinus CL-S1T (95.5%).

To further confirm the genus affiliation of strains JC83 and JC84T, the housekeeping genes rpoB, gyrB and hsp60 were also amplified and sequenced. The rpoB gene was amplified using the primers CamrpoB-L (5’-CAATTATTTGATGACAC-3’) and RpoB-R (5’-GTTCATGTTGNNACCAT-3’), the gyrB gene was amplified with the primers gyrB-Arc-7F (5’-GTTTAYCATTGAAGGTGG-3’) and gyrB-Arc-14R (5’-CTAGATTITTTCAACATTTAAT-3’), and the hsp60 gene was amplified with the primers hsp60F (5’-TTGAACCTTAAAAAGGGTCCAG-3’) and hsp60R (5’-TCCATCAACATGTCACCT-3’), and these were sequenced according to the methods described by Collado et al. (2009, 2011) and Figueras et al. (2011a).

The concatenated sequences of housekeeping genes rpoB, gyrB and hsp60 were first aligned by CLUSTAL X and the alignments were improved by removing poorly aligned regions within each gene sequence by using the online program Gblocks (Castresana, 2000). A phylogenetic tree of the concatenated sequences was constructed using CLUSTAL W within MEGA 5.2 (Tamura et al., 2011) and distances were calculated with default parameters. The robustness of the tree topologies was evaluated by 100 bootstrap replications. Bootstrap values were calculated...
using the NJ and MP methods in MEGA 5.2 to compare and validate the tree topologies reconstructed by the different algorithms. The concatenated gene sequences of gyrB (586 bp), hsp60 (513 bp) and rpoB (573 bp) of strains JC83 and JC84T were compared with those of the type strains along with additional strains in the genus Arcobacter, and the improved Gblocks concatenated sequences consisted of 1444 homologous positions. The constructed phylogenetic NJ tree (Fig. S2) clearly confirmed the genus affiliation of strains JC83 and JC84T but distinct from other members of the genus Arcobacter.

The taxonomic relationship between strains JC83 and JC84T was examined further using genome DNA–DNA hybridization. Genomic relatedness was determined by the membrane filter technique (Seldin & Dubnau, 1985; Tourova & Antonov, 1987) using a DIG High Prime DNA labelling and Detection Starter kit II (Roche). Hybridization was performed with three replications for each sample (control: reversal of the strains used for binding and labelling) and a mean DNA–DNA relatedness of >85% (86.0 ± 0.5%) between strains JC83 and JC84T was observed.

All morphological, physiological and biochemical analyses, unless otherwise mentioned, were performed for both strains JC83 and JC84T separately in nitrate medium containing (per litre) beef extract (3 g), peptone (5 g) and KNO₃ (1 g) under anaerobic incubation conditions. A. nitrofigilis DSM 7299T (representing the type species of the genus Arcobacter), A. marinus DSM 24769T (=CL-S1T), A. molluscum DSM 18005T (=LA31BT) and A. halophilus DSM 18005T (=LA31BT) were used for comparative taxonomic analysis owing to their phylogenetic closeness to strains JC83 and JC84T. On nitrate agar, anaerobically grown colonies of strains JC83 and JC84T were pale yellow, circular, convex and with entire margin. On Postgate’s C medium, colonies were tiny, circular and cream coloured which become milk white on exposure to air. Morphological properties (cell shape, cell size, motility) as observed under phase-contrast light microscopy (Olympus BH-2) indicated that individual cells of both strains were rods (1.0–2.0 μm long, 0.1–0.3 μm wide; Fig. S3) and non-motile. The absence of motility in both strains was confirmed through the hanging-drop method, and negatively stained cells observed by transmission electron microscopy (H-7500; Hitachi) indicated the absence of flagella.

Growth and tolerance of the strains at different temperatures, pH and salinity were determined in nitrate broth. Growth was measured turbidometrically at 540 nm from cultures which were centrifuged (15 000 r.p.m. for 15 min) and resuspended in distilled water. Anaerobically grown broth cultures were pale yellow, slightly gelatinous and sedimented at the bottom of the tube after 2 days of growth. NaCl was not required for growth of the two strains, although optimum growth was at 1–3% and they
were able to tolerate up to 6% (w/v). Both strains showed optimum growth at pH 7.0–8.0 (range pH 7–10) and 30 °C (range 25–37 °C). Chemoorganoheterotroph was the only growth mode and both strains utilized a limited range of organic molecules (Table S1).

Phenotypic characterization was performed following the proposed minimal standards for describing new species of the family Campylobacteraceae (Ursing et al., 1994). Growth was not possible on blood agar and no growth was observed in the presence of triphenyltetrazolium chloride (0.04%). Oxidase was positive while catalase was negative for both strains. Oxidases are uncommon among strict anaerobic bacteria, although the presence of membrane-bound functional cytochrome bd-type oxidases in a few strict anaerobes helps in catalysing the reduction of low levels of dioxygen (Das et al., 2005; Baughn & Malamy, 2004; Lemos et al., 2001). Both strains hydrolysed casein and reduced nitrate. Both strains produced H2S in Postgate's C medium, iron-bisulphite-pyruvate medium and sulfide-indole-motility agar. The dsrAB gene, which codes for sulfite reductase, could not be amplified using the primer set dsr1F (5′-AC[C/G]CAGTGAAGCACG-3′) and dsr4R (5′-GTGAGCAGTACCGCA-3′; Sasi Jyothsna et al., 2008), while this gene was positive for Desulfovibrio psychrotolerans JS1T, which was used as a positive control. Gelatin was not liquefied and indole was not produced from L-tryptophan by both strains. Lipase, amylase and urease activities were negative for both strains. Acid from glucose and acid phosphatase were not produced. Indoxyl acetate was hydrolysed by both strains.

For cellular fatty acids, anaerobically grown cells were harvested when growth of the cultures attained 70% of maximal optical density (at their late exponential growth phase). Forty milligrams of bacterial cells was subjected to saponification and the methyl esters were analysed by GC (Agilent 6890) with Sherlock MIS software (Microbial ID; MIDI 6.0 version; peak identification was done based on the RTSSA6 database) [Sasser (1990); http://www.midi-inc.com/]. Analysis was outsourced to the Royal Research Laboratories, Secunderabad, India. C16:1ω7c (25%), C18:1ω7c (34.4%), C16:0 (20.5%) and C12:0 (7.3%) were the major (>5%) cellular fatty acids of strain JC84T. Minor (≤5%, >1%) amounts of C12:0 3-OH (3.4%), C14:0 (3.5%) and C16:0 (23%) were also present. The fatty acid profile of strain JC83 was qualitatively identical to that of strain JC84T. The fatty acid profile of strain JC84T was similar to those of A. nitrofigilis DSM 7299T (=Cl1T), A. marinus DSM 24769T, A. molluscorum LMG 25693T and A. halophilus DSM 18005T, which were analysed in parallel, further differentiated strains JC83 and JC84T from their nearest phylogenetic neighbours (Fig. S4).

Quinones from strains JC83 and JC84T were extracted with a chloroform/methanol (2:1, v/v) mixture, purified by TLC and analysed by HPLC as described (Tamaoka et al., 1983; Hiraishi & Hoshino, 1984; Hiraishi et al., 1984) and were compared with those extracted from A. nitrofigilis DSM 7299T, A. marinus DSM 24769T, A. molluscorum LMG 25693T and A. halophilus DSM 18005T. Both novel strains had MK-6 (97%) and MMK-6 (3%) as respiratory quinones. The quinone compositions of strains JC83 and JC84T were not in line with that of A. nitrofigilis DSM 7299T, which had MK-6 (50%), MK-7 (46%) and MK-8 (4%). However, the quinone compositions of A. marinus DSM 24769T, A. molluscorum LMG 25693T and A. halophilus DSM 18005T were similar to those of strains JC83 and JC84T.

The distinct polar lipid profile (Fig. S4), quinone composition and other phenotypic traits (Table 1), including lack of motility, obligate anaerobic growth, negativity for catalase, positivity for casein hydrolysis, ability to produce H2S and citrate utilization, allows a clear separation of strains JC83 and JC84T from the type strains of recognized species in the genus Arcobacter. Based on their molecular and phenotypic distinctiveness, strains JC83 and JC84T are considered to represent a novel species of the genus Arcobacter, for which the name Arcobacter anaerophilus sp. nov. is proposed.

Description of Arcobacter anaerophilus sp. nov.

Arcobacter anaerophilus [a.n.a.e.ro’phi.lus. Gr. pref. an not; Gr. masc. n. aer air; N.L. masc. adj. philus (from Gr. masc. adj. philos) friend, loving; N.L. masc. adj. anaerophilus not air-loving].

International Journal of Systematic and Evolutionary Microbiology 63
**Table 1.** Characteristics that differentiate strain JC84<sup>T</sup> from all other species of the genus *Arcobacter*

<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>
<th>10</th>
<th>11</th>
<th>12</th>
<th>13</th>
<th>14</th>
<th>15</th>
<th>16</th>
<th>17</th>
<th>18</th>
</tr>
</thead>
<tbody>
<tr>
<td>Motility</td>
<td>-</td>
<td>+</td>
<td>+</td>
<td>+</td>
<td>+</td>
<td>+</td>
<td>+</td>
<td>+</td>
<td>+</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>+</td>
<td>+</td>
<td>-</td>
<td>+</td>
<td>+</td>
<td>+</td>
</tr>
<tr>
<td>Growth in/on:</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Air at 37 °C</td>
<td>-</td>
<td>-</td>
<td>+</td>
<td>+</td>
<td>+</td>
<td>+</td>
<td>+</td>
<td>+</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>+</td>
<td>+</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
</tr>
<tr>
<td>CO&lt;sub&gt;2&lt;/sub&gt; at 37 °C*</td>
<td>-</td>
<td>-</td>
<td>+</td>
<td>+</td>
<td>+</td>
<td>+</td>
<td>+</td>
<td>+</td>
<td>V(+)</td>
<td>+</td>
<td>+</td>
<td>+</td>
<td>-</td>
<td>-</td>
<td>+</td>
<td>+</td>
<td>-</td>
<td>-</td>
</tr>
<tr>
<td>CO&lt;sub&gt;2&lt;/sub&gt; at 42 °C*</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>V(+)</td>
<td>-</td>
<td>-</td>
<td>(+)</td>
<td>-</td>
<td>-</td>
<td>ND</td>
<td>ND</td>
<td>-</td>
<td>-</td>
</tr>
<tr>
<td>100% Argon†</td>
<td>+</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
</tr>
<tr>
<td>Blood agar</td>
<td>-</td>
<td>+</td>
<td>+</td>
<td>+</td>
<td>+</td>
<td>+</td>
<td>-</td>
<td>-</td>
<td>+</td>
<td>+</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
</tr>
<tr>
<td>NaCl (0.5%)</td>
<td>+</td>
<td>+</td>
<td>-</td>
<td>-</td>
<td>+</td>
<td>+</td>
<td>+</td>
<td>+</td>
<td>+</td>
<td>+</td>
<td>+</td>
<td>+</td>
<td>+</td>
<td>+</td>
<td>+</td>
<td>+</td>
<td>+</td>
<td>+</td>
</tr>
<tr>
<td>NaCl (4%)</td>
<td>+</td>
<td>+</td>
<td>+</td>
<td>+</td>
<td>+</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
</tr>
<tr>
<td>Glycine (1%)</td>
<td>+</td>
<td>-</td>
<td>+</td>
<td>+</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>V(+)</td>
<td>-</td>
<td>-</td>
<td>+</td>
<td>+</td>
<td>ND</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
</tr>
<tr>
<td>Resistance to cefoperazone (64 mg l&lt;sup&gt;-1&lt;/sup&gt;)</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
</tr>
<tr>
<td>Enzyme activities</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Catalase</td>
<td>-</td>
<td>+</td>
<td>-</td>
<td>-</td>
<td>+</td>
<td>+</td>
<td>+</td>
<td>+</td>
<td>(+)</td>
<td>V(+)</td>
<td>+</td>
<td>+</td>
<td>+</td>
<td>+</td>
<td>+</td>
<td>+</td>
<td>+</td>
<td>+</td>
</tr>
<tr>
<td>Urease</td>
<td>-</td>
<td>+</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
</tr>
<tr>
<td>Nitrate reduction</td>
<td>+</td>
<td>+</td>
<td>+</td>
<td>+</td>
<td>-</td>
<td>-</td>
<td>+</td>
<td>+</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>+</td>
<td>+</td>
<td>-</td>
<td>-</td>
<td>+</td>
<td>+</td>
</tr>
<tr>
<td>Indoxyl acetate hydrolysis</td>
<td>+</td>
<td>+</td>
<td>+</td>
<td>+</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>V(-)</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
</tr>
<tr>
<td>H&lt;sub&gt;2&lt;/sub&gt;S production</td>
<td>+</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>ND</td>
<td>ND</td>
<td>ND</td>
<td>ND</td>
<td>ND</td>
<td>ND</td>
<td>ND</td>
<td>ND</td>
<td>ND</td>
<td>ND</td>
</tr>
<tr>
<td>Casein hydrolysis</td>
<td>+</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>ND</td>
<td>ND</td>
<td>ND</td>
<td>ND</td>
<td>ND</td>
<td>ND</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
</tr>
<tr>
<td>DNA G+C content (mol%)</td>
<td>24.6</td>
<td>28–31</td>
<td>28</td>
<td>35</td>
<td>29</td>
<td>ND</td>
<td>ND</td>
<td>28–31</td>
<td>28–29</td>
<td>29</td>
<td>26.8–27.3</td>
<td>26.9</td>
<td>28.5</td>
<td>28.5</td>
<td>ND</td>
<td>ND</td>
<td>ND</td>
<td>ND</td>
</tr>
</tbody>
</table>

*Microaerobic conditions.
† Obligate anaerobic growth under 100% argon.
‡ Growth on glycine, which was previously not observed by Donachie et al. (2005).
Gram-stain-negative, non-spore-forming, non-motile, non-flagellated rods. Cells are 1.0–2.0 μm long and 0.1–0.3 μm wide. On nitrate agar, anaerobically well grown colonies are pale yellow, circular, smooth with entire margin. Anaerobic broth culture is pale yellow, gelatinous and sediments at the bottom. Catalase-negative, oxidase-positive; utilizes citrate, hydrolysae casein, reduces nitrate and produces H₂S. Amylase, urease and gelatinase are not produced. Growth occurs on limited carbon sources and good growth occurs on complex media. NaCl is not essential for growth, but good growth occurs with 1–3 % (w/v) NaCl; tolerates up to 6 % NaCl. The following compounds are oxidized in the Biolog GN2 test system: L-arabinose, D-arabitol, cellobiose, lactose, D-mannose, melibiose, acetate, inosine and phenylethylamine. Major polar lipids include DPG, PE, PG and PS; PC, an unidentified aminolipid (AL2), an unidentified phospholipid (PL2), PI and an unidentified lipid (L3) are also present. Major fatty acids include C₁₆:₁ω₂₇c, C₁₈:₁ω₇c, C₁₆:₀ and C₁₂:₀. Minor amounts of C₁₂:₀ 3-OH, C₁₄:₀ and C₁₈:₀ are also present. MK-6 is the major quinone and MMK-6 is present in minor amounts.

The type strain is JC84T (=KCTC 15071T=MTCC 10956T=DSM 24636T), which was isolated from a sediment sample of Gangasagar estuary, West Bengal, India. The DNA G+C content of the type strain is 24.6 mol%. JC83, isolated from the same region, is a second strain of the species. The DNA G+C content of strain JC83 is 25 mol%.

**Emended description of the genus Arcobacter**

Vandamme *et al.* 1991, 1992

The description is as given previously (Vandamme *et al.*, 1991, 1992) with the following modifications. Some species are obligate anaerobes and lack motility. Cells of some species are filamentous and are up to 7 μm long. Catalase is absent in some species. Hydrolysis of casein is variable and H₂S is produced by some species. The type species of the genus has MK-6 and MK-7 as major quinones with minor amounts of MK-8 while some species have MK-6 as the major respiratory quinone with minor amounts of MMK-6. C₁₆:₁ω₂₇c and C₁₈:₁ω₇c are the major fatty acids with minor amounts of C₁₂:₀ 3-OH, C₁₄:₀ C₁₄:₁ω₆c and C₁₈:₁ω₉c also present in some species. All species tested have PG and PE as major polar lipids. In addition, some species have DPG, PE, PG and PS; an unidentified aminolipid (AL1, AL2), unidentified phospholipids (PL1, PL2) and unidentified lipids (L1, L2 and L3). The DNA base composition ranges from 24.6 to 31 mol% G+C.

**Acknowledgements**

We thank BCCM/LMG Bacteria Collection for providing *A. molluscum* 1998-3T (=LMG 25693T) on an exchange basis. We thank Professor J. Euzéby for his expert suggestion regarding the correct species epithet and Latin etymology. T. S. S. J. acknowledges UGC and K. R. and E. V. V. R. acknowledge CSIR, New Delhi, for the award of a post-doctoral and senior research fellowship, respectively.

Facilities used under the DBT-CREB, DST-FIST and UGC-CAS from the University of Hyderabad are acknowledged.

**References**


