The genus *Oceanicola*, which belongs to the order *Rhodobacterales* of the class *Alphaproteobacteria*, was first proposed by Cho & Giovannoni (2004). Currently, the genus includes five species, *Oceanicola granulosus* (the type species), *Oceanicola batsensis*, *Oceanicola nanhaiensis*, *Oceanicola marinus* and *Oceanicola pacificus*, which are Gram-negative, non-motile, *poly-β*-hydroxybutyrate (PHB)-accumulating and contain large amounts of unsaturated cellular fatty acids. *O. granulosus* strains HTCC2516T and HTCC2523 and *O. batsensis* HTCC2597T were isolated from the Bermuda Atlantic Time-series Study (BATS) site by means of a high-throughput culturing method based on dilution-to-extinction in an oligotrophic seawater-based medium (Cho & Giovannoni, 2003). *O. nanhaiensis* SS011B1-20T was isolated from sediment samples from the South China Sea (Gu et al., 2007). *O. marinus* was obtained from seawater collected off the coast of Taiwan (Lin et al., 2007). *O. pacificus* was isolated from a deep-sea pyrene-degrading consortium (Yuan et al., 2009). All strains of this genus were isolated from the ocean. JLT1210T was isolated from the Beibu Gulf in the South China Sea during voyage no. 908 on the ship Dongfanghong 2 in July 2006, strain JLT1210T was obtained after direct plating of seawater dilutions. It was incubated in a rich organic (RO) medium (Yurkov et al., 1999) at room temperature (28 °C) for 10 days. RO medium contained the following (g l−1): yeast extract, 1.0; Bacto Peptone, 1.0; sodium acetate, 1.0; KCl, 0.3; MgSO4·7H2O, 0.5; CaCl2·2H2O, 0.05; NH4Cl, 0.3; K2HPO4, 0.3; and NaCl, 20.0. This medium was supplemented with a mixture of vitamins and trace element solution.

Cell morphology was examined by transmission electron microscopy (JEM 2100 HC). The Gram reaction was performed using the method described by Gerhardt et al. (1994). Colonies were pale yellow, 0.5–3.0 mm in diameter, uniformly circular, convex and opaque after growth on marine agar RO at 28 °C for 3 days. Cells stained Gram-negative and were short rods, 0.7–2.1 μm long and 0.4–1.1 μm wide, with polar or subpolar flagella, dividing by binary fission (Fig. 1).

During our surveys of the biodiversity of bacteria in the South China Sea during voyage no. 908 on the ship Dongfanghong 2 in July 2006, strain JLT1210T was obtained after direct plating of seawater dilutions. It was incubated in a rich organic (RO) medium (Yurkov et al., 1999) at room temperature (28 °C) for 10 days. RO medium contained the following (g l−1): yeast extract, 1.0; Bacto Peptone, 1.0; sodium acetate, 1.0; KCl, 0.3; MgSO4·7H2O, 0.5; CaCl2·2H2O, 0.05; NH4Cl, 0.3; K2HPO4, 0.3; and NaCl, 20.0. This medium was supplemented with a mixture of vitamins and trace element solution.

Abbreviation: PHB, *poly-β*-hydroxybutyrate; RO, rich organic medium. The GenBank/EMBL/DDBJ accession number for the 16S rRNA gene sequence of strain JLT1210T is EU581832.
respectively. The temperature range for growth of strain JLT1210T was 10–45 °C (optimum 28 °C), the pH range for growth was pH 7.0–9.5 (optimum 7.8–8.5) and the NaCl concentration range for growth was 0.5–7.0 % (w/v) (optimum 1.5–3.0 %).

Oxidase activity of strain JLT1210T was tested according to the method of Smibert & Krieg (1994) and catalase activity was determined by using 3% (v/v) hydrogen peroxide solution (Dong & Cai, 2001). Nitrite and nitrate reduction were tested in RO medium by growing the strain in the presence of NO_2^- and NO_3^-, respectively (Dong & Cai, 2001). Denitrification was tested by growing the strain anaerobically in the presence of NO_2^- and NO_3^- (Zumft, 1992).

Hydrolysis of starch, gelatin and Tween 80, and urease activity were determined as described by Dong & Cai (2001). H_2S production was tested in RO medium supplemented with 0.01 %L-cysteine, the indicator being a strip of paper impregnated with lead acetate placed in the neck of the tube (Clarke, 1953; Mata et al., 2002). Methyl red and Voges–Proskauer tests were performed by using methyl red and Barritt’s reagent (Barritt, 1936; Mata et al., 2002). Accumulation of PHB was determined by the Sudan Black staining method (Smibert & Krieg, 1994). Growth on sole carbon and nitrogen sources was examined according to the procedures described by Williams et al. (1983) on RO medium without organic compounds at 28 °C for 7–14 days.

The isolate was positive for oxidase and catalase activities. Biochemical tests were positive for aerobic nitrate reduction, and PHB and H_2S production. Gelatin, starch, Tween 80 and casein hydrolysis, indole production, aerobic nitrite reduction, anaerobic nitrate reduction, methyl red and Voges–Proskauer tests, and urease activity were negative.

The strain utilized a wide range of substrates, including hexoses, oligosaccharides, organic acids and amino acids, as sole carbon sources. Antimicrobial susceptibility tests were performed by using the disc-diffusion plate (Kirby–Bauer) method according to Fraser & Jorgensen (1997) and Andrews (2008), and with the antimicrobial compounds suggested by Mata et al. (2002).

### Table 1. Fatty acid composition (%) of strain JLT1210T and type strains of related Oceanicola species

<table>
<thead>
<tr>
<th>Fatty acid</th>
<th>1</th>
<th>2</th>
<th>3</th>
<th>4</th>
<th>5</th>
<th>6</th>
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<td>10:0</td>
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<td>–</td>
<td>–</td>
<td>–</td>
<td>–</td>
<td>–</td>
</tr>
<tr>
<td>10:0 3-OH</td>
<td>0.2</td>
<td>0.1</td>
<td>1.5</td>
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<td>–</td>
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<tr>
<td>11:0 3-OH</td>
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<td>–</td>
<td>–</td>
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</tr>
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<td>2.0</td>
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<td>–</td>
<td>–</td>
</tr>
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<td>–</td>
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<td>–</td>
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<tr>
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<td>–</td>
<td>–</td>
<td>–</td>
<td>0.4</td>
<td>–</td>
</tr>
<tr>
<td>12:0 3-OH</td>
<td>1.8</td>
<td>1.6</td>
<td>–</td>
<td>2.1</td>
<td>–</td>
<td>–</td>
</tr>
<tr>
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<td>–</td>
<td>–</td>
<td>–</td>
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</tr>
<tr>
<td>14:0</td>
<td>3.8</td>
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<td>1.5</td>
<td>0.2</td>
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<td>–</td>
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<td>iso-16:0</td>
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<tr>
<td>16:0</td>
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<tr>
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<tr>
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<td>–</td>
<td>–</td>
<td>–</td>
<td>1.3</td>
</tr>
<tr>
<td>17:0</td>
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<td>0.4</td>
<td>0.4</td>
<td>1.5</td>
<td>–</td>
<td>6.6</td>
</tr>
<tr>
<td>18:1o7c</td>
<td>25.7</td>
<td>81.2</td>
<td>62.8</td>
<td>31.0</td>
<td>49.1</td>
<td>32.1</td>
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<tr>
<td>18:0</td>
<td>0.9</td>
<td>1.3</td>
<td>0.9</td>
<td>2.4</td>
<td>1.0</td>
<td>7.3</td>
</tr>
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<td>8.1</td>
<td>–</td>
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<tr>
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<tr>
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<td>–</td>
<td>–</td>
<td>0.2</td>
<td>0.4</td>
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</tr>
</tbody>
</table>

*Summed feature 3 comprises iso-15:0 2-OH and/or 16:1o7c; summed feature 7 comprises 19:0 cyclo o10c and/or 19:1o6c.

Fig. 1. Electron micrographs of negatively stained cells of strain JLT1210T observed by transmission electron microscopy. Bars, 0.5 µm.
The chemotaxonomic study of strain JLT1210T included analysis of its cellular fatty acids. Cellular fatty acid analysis was carried out as described by Komagata & Suzuki (1987). Isoprenoid quinones were analysed as described by Hiraishi et al. (1998) using a UPLC-Q-TOF-MS spectrometer and the electrospray ionization method (Romano et al., 2006). The predominant fatty acid was summed feature 3 (iso-C15:0 2-OH and/or C16:1ω7c: 40.5%); significant amounts of C18:1ω7c (25.7%), C16:0 (16.2%), C14:0 (3.8%), C12:0 (2.4%), C12:0 3-OH (1.8%), C17:1ω8c (1.7%), C15:0 (1.6%) and C17:0 (1.1%) were also present. Fatty acid profiles of members of the genus Oceanicola are listed in Table 1.

Genomic DNA was extracted using the method of Marmur (1961) from cells grown in RO medium for 2 days at 28°C, washed and resuspended in TE buffer [10 mM Tris-HCl (pH 7.5), 1 mM EDTA (pH 8.0)]. Purity was assessed using A280/A260 and A230/A260 ratios (Johnson, 1994). Phylogenetic analyses based on 16S rRNA gene sequences were performed as described by Lin et al. (2007). The 16S rRNA gene was amplified with universal bacterial primers corresponding to Escherichia coli positions 8F (5′-AGA GTTTGATCCTGCTCAG) and 1492R (5′-GGTACCTTGTTACGACTT) (Embley, 1991). The 16S rRNA gene sequence of strain JLT1210T was compared with those available in GenBank using the BLAST program (NCBI) to determine the approximate phylogenetic affiliation. The 16S rRNA gene sequence of strain JLT1210T was aligned with those of related Oceanicola species and phylogenetic trees were constructed using the neighbour-joining and maximum-parsimony methods of the MEGA software (Kumar et al., 2004; Fig. 2). Nearly complete 16S rRNA gene sequences (1386 bp) were determined for strain JLT1210T and used for phylogenetic analyses. The closest neighbours were O. nanhaiensis SS011B1-20T (96.5% similarity), O. batsensis HTCC2597T (96.4%) and O. granulosus HTCC2516T (93.2%).

The genomic DNA G+C content of strain JLT1210T was estimated from the midpoint value (Tm) of the thermal denaturation profile, as described by Mandel et al. (1970). The DNA G+C content of strain JLT1210T was 72.8 mol% and the predominant respiratory ubiquinone was Q-10.

**Description of Oceanicola nitratireducens sp. nov.**

Oceanicola nitratireducens (ni.tra.ti.re-du.ens. N.L. n. nitras -atis nitrate; L. part. adj. reducens leading back, bringing back and in chemistry converting to a different
Cells are Gram-negative, non-motile, short rods with polar or subpolar flagella. Small (about 0.5–3.0 mm) straw yellowish colonies are produced on RO after incubation at 28 °C for 3–5 days. Colonies are smooth, convex and opaque. Growth occurs at 10–37 °C, at pH 7.0–9.5 and in 0.5–7.0 % NaCl; optimum growth occurs at 28 °C, at pH 7.8–8.5 and in 1.5–3.0 % NaCl. Positive for oxidase, catalase and nitrate reduction. Negative for hydrolysis of gelatin, starch, Tween 80 and casein, aerobic nitrite reduction, anaerobic nitrate reduction, methyl red and Voges–Proskauer tests, and indole production. The utilization of various substrates as sole carbon sources, antimicrobial susceptibility and other differentiating characteristics are listed in Table 2. The following substrates are utilized: maltose, sucrose, D-fructose, D-galactose, dextrin, trehalose dihydrate, cellobiose, citric acid, sodium D-gluconate, glycerol, mannitol, L-(−)-malic acid, acetic acid, ethanol, L-threonine, L-alanine, L-glycine and L-arginine. Sensitive to ciprofloxacin, cephalothin, tetracycline, gentamicin, chloramphenicol and cefotaxime. The predominant fatty acid is summed feature 3 (iso-C15 : 02-OH and/or C16 : 1 ω7c); significant amounts of C18 : 1 ω7c, C16 : 0, C14 : 0, C12 : 0, C12 : 0 3-OH, C17 : 1 ω9c, C15 : 0 and C17 : 0 are also present. The predominant respiratory ubiquinone is Q-10.

The type strain is JLT1210 T (LMG 24663 T = CGMCC 1.7292 T), isolated from surface water of the Beibu Gulf in the South China Sea. The DNA G+C content of the type strain is 72.8 mol% (Tm method).

Acknowledgements

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References


