Belliella buryatensis sp. nov., isolated from alkaline lake water

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Two bacterial isolates from water of the alkaline brackish Lake Solenoe (Buryatia, Russia), 2C and 5C T, were characterized by using a polyphasic taxonomic approach. The strains were small, non-motile, Gram-stain-negative rods that formed small orange–red colonies on the surface of marine agar. Studies based on 16S rRNA gene sequences showed that the strains were related closely to Belliella pelovolcani CC-SAL-25 T (98.7 % sequence similarity). The G + C content of the DNA was 38–40 mol%. DNA–DNA hybridization values between strains 2C and 5C T and B. pelovolcani CC-SAL-25 T were 56–58 mol%. A menaquinone with seven isoprene units (MK-7) was the major respiratory quinone. The fatty acid profiles were slightly different from that of B. pelovolcani CC-SAL-25 T. The novel strains could be distinguished from the phylogenetically closest species B. pelovolcani CC-SAL-25 T based on matrix-assisted laser desorption ionization time-of-flight mass spectra of whole cells and a range of physiological and biochemical characteristics. The data obtained suggest that strains 2C and 5C T represent a novel species of the genus Belliella, for which the name Belliella buryatensis sp. nov. is proposed. The type strain is 5C T (=VKM B-2724 T =KCTC 32194 T).

The phylum Bacteroidetes is one of the most abundant bacterial groups of microbial communities in freshwater and marine ecosystems, cold-water habitats and habitats with high salinity (Dorador et al., 2009). Based on metagenomic data, the abundance of Bacteroidetes is about 10–20 % in alkaline lake sediments (Xiong et al., 2012; Zaitseva et al., 2014; Glaring et al., 2015). Bacteroidetes has an important role in the marine carbon cycle, involved in the degradation of high-molecular-mass compounds such as cellulose, agar and chitin (Cottrell & Kirchman, 2000; Kirchman, 2002; Abell & Bowman, 2005).

The family Cytophagia of the phylum Bacteroidetes. At present the family consists of 15 genera of bacteria including the genus Belliella (http://www.bacterio.net/-classifgenerfami-les.html#Cyclobacteriaceae), which have been isolated from alkaline, saline and thermal habitats. At the beginning of our research, the genus Belliella was represented by two recognized species. Belliella baltica was isolated from surface water of the Baltic Sea (Brettar et al., 2004) and Belliella pelovolcani was isolated from a rare mud-volcano in Wandan, Pintung County, Taiwan (Arun et al., 2009). Two novel species of the genus Belliella have more recently been isolated and described. Belliella kenyensis (Akhwale et al., 2015) and Belliella aquatica (Zhong et al., 2015) were isolated from sediment of alkaline Lake Elmenteita in the Kenyan Rift Valley and surface water of saline Lake Tuosu in China, respectively.

During investigations into the biodiversity of the culturable microbial community of water from Lake Solene,
Republic of Buryatia, Russia, five strains were isolated from orange-red colonies forming on plate count agar (PCA, pH 8; Difco) and marine agar (MA, pH 8; Difco). Subsequent analysis of the incomplete 16S rRNA gene sequences of these strains indicated that they belonged to the genus Belliella.

In the present study, two strains, designated 2C and 5CT, were subjected to a polyphasic taxonomic analysis and were shown to represent a novel species of the genus Belliella.

Lake Solonoe is situated at 50° 23’ 75° N 106° 42’ 22” E in the south of the Republic of Buryatia. It is a shallow, brackish, alkaline lake typical for the Transbaikalian region, with interannual fluctuations of water depth, temperature, pH and mineralization. A sample of the surface water was used as inoculum. At the time of sampling (2010), the water was characterized by the following parameters: temperature 25.6 °C, pH 9.7, salinity 9270 mg dm⁻³, O₂ 5.9 mg dm⁻³, with main ions (mg dm⁻³) Cl⁻ (3766), HCO₃⁻ (400), CO₃²⁻ (1400) and Na⁺ (3380) (Egorova et al., 2011).

Strains 2C and 5CT were isolated by a standard tenfold dilution plating technique on PCA. Subsequently, the strains were routinely cultured on PCA and/or MA. To produce biomass for chemotaxonomic and genomic analyses, strains were grown on a medium similar to marine broth (MB) (NaCl (0.022), Na₂SiO₃ (0.004), NaF (0.002), (NH₄)NO₃ (0.001)), based on the recommended standard tests as described by Gerhardt et al. (1981). Cellular fatty acids, matrix-assisted laser desorption ionization time-of-flight (MALDI/TOF) MS analysis and DNA–DNA hybridization.

Cell morphology, motility and determination of cell size were studied using a light microscope (AxioStar plus, x 1000; Carl Zeiss) equipped with phase-contrast optics and AxioVision software, with cultures grown for 48–72 h at 28 °C on MA. Gram-staining was performed as described by Gerhardt et al. (1981).

The pH range for growth was determined in MB that was adjusted prior to sterilization to various pH values (5.3, 6.6, 8.1, 8.5, 9.0, 9.5, 10.8, 11.5) using appropriate biological buffers (Na₂HPO₄ + NaH₂PO₄, KCl/H₂BO₃ + NaOH, Na₂CO₃ + NaHCO₃, NaHCO₃ + NaOH) (Dawson et al., 1986). Verification of the pH values after autoclaving revealed only minor changes. Growth at various temperatures (21–60 °C) adjusted with a gradient thermostat was measured in MB. The salinity range and optimum were tested in MB which contained 0.1, 0.5, 1, 2.5, 5, 10, 15 and 20 % (w/v) NaCl and also in the same medium without NaCl. The growth intensity was assessed in triplicate by measuring the optical density (wavelength, 560 nm) with a Cecil CE1021 spectrophotometer (Cecil Instruments) in all the above experiments.

Phenotypic and biochemical characteristics were determined based on the recommended standard tests as described by Gerhardt et al. (1981) and with the use of API 20E and API 50CH kits (bioMérieux), according to the manufacturer’s instructions, except that 0.9 % NaCl solution was used for inoculation (API 20E). Catalase and oxidative activities were tested with a 3 % (v/v) H₂O₂ solution and 1 % (w/v) tetramethyl-p-phenylenediamine, respectively.

Resistance to antibiotics was determined by the diffusion method using discs (Scientific Research Center of Pharmacotherapy), according to the manufacturer’s instructions. Discs contained (μg): ampicillin/subactam (10/10), ticarcillin (75), ticarcillin/clavulanic acid (75/10), piperacillin (100), piperacillin + tazobactam (100/10), cefepime (30), imipenem (10), meropenem (10), ciprofloxacin (5), ceftazidime (30), amikacin (30), gentamicin (10, 120), tobramycin (10), colistin (50), cotrimoxazole (trimethoprim/sulfamethoxazol) (1.25/23.75), erythromycin (15) and clindamycin (2). This corresponds to the ATB-PSE 5 antibiotic set (bioMérieux).

The presence of flexirubin-type pigments was investigated as described by Reichenbach et al. (1981). Cellular pigments were extracted with acetone/methanol (7:2, v/v), from cultures grown in MB. Absorption spectra were determined with a scanning UV/visible spectrophotometer (Shimadzu UV-mini at 300–700 nm; Shimadzu Scientific Instruments).

Lipoquinones were extracted and purified according to Collins (1985) and analysed by MS for strain 5CT only.

Cellular fatty acids were analysed by GC/MS using an Agilent HP 6890 gas chromatograph (Agilent Technologies) with HP MSD 5973N quadrupole mass spectrometer as a detector in SIM and SCAN mode. Chromatography of methyl esters of fatty acids and trimethyl-silyl ethers was performed using an HP-5MS column with an internal diameter of 0.25 mm. The percentage composition of the mixture was calculated from the peak areas. Qualitative analysis was performed on the basis of comparison of retention times and mass spectra against the complete NIST08L database and the standard mixture of bacterial fatty acids [Bacterial Acid Methyl Ester (CP Mix; Supelco)]. Cells were collected in the exponential growth phase (48 h) after growth in MB at 28 °C. Assays were performed in duplicate.

For MALDI/TOF MS analyses of whole cells, strains were grown for 48 h on R2A medium (M962; HiMedia). Surface cultures (~5–10 μg) were transferred with a thin sterile spatula into a plastic tube containing 50 μl of the freshly prepared 50 % aqueous acetonitrile solution (Sigma-Aldrich) supplemented with 2.5 % trifluoroacetic acid and mixed thoroughly. The suspension (0.8 μl) was then applied to a polished steel MALDI sample target, mixed
with an equal volume of the matrix solution (z-cyano-4-
hydroxycinnamic acid in 50 % aqueous solution of aceto-
nitrile containing 2.5 % trifluoroacetic acid), and air-dried
at room temperature.

Mass spectra were registered in linear mode with delayed
ion extraction on an Autoflex Speed mass spectrometer
(Bruker Analytics). Spectra were recorded in positive ion
mode using delay time of 350 ns, and an accelerating vol-
tage of 20 kV. The mass range was 2–20 kDa. The Bruker
Bacterial Test Standard (Bruker Daltonics) mixture was
used for external calibration; spectra resolution was
±2 Da (200 p.p.m.). The resulting spectra of each strain
were obtained by summation of the spectra registered at
10–15 points of the analyte upon 500 laser shots. Mass
spectra were processed using the Flex analysis 3.3 and
Biotyper 3.0 software packages (Bruker Daltonics).

Genomic DNA was extracted and purified by using the
CTAB method (Wilson, 1997) with an additional purifi-
cation step with RNase, proteinase K and three deprotein-
ization cycles (Mesbah et al., 2011). The DNA G+C
content was determined by the thermal denaturation
method (Owen & Pitcher, 1985; De Bartolomeo et al.,
1991) using a DU800 spectrophotometer (Beckman Coul-
ter) with a temperature-controlled cell. For DNA–DNA
relatedness experiments, the purified DNA was disrupted
with an ultrasonic disintegrator to obtain a fragment
length of 400–800 bp. Hybridization reactions were carried
out as described by Rossello-Mora et al. (2011) and calcu-
lations of percentage relatedness were performed as
described by De Ley et al. (1970). Each experiment was
repeated at least twice.

Amplification and sequencing of the 16S rRNA gene were
performed as described previously (Anan’ina et al., 2007).
Sequencing was carried out on a Genetic Analyzer 3500xl
(Applied Biosystems) according to the manufacturer’s rec-
ommendations. Alignment of 16S rRNA gene sequences
from strains 2C and 5C\textsuperscript{T} and closely related organisms
retrieved from GenBank (http://www.ncbi.nlm.nih.gov)
and Ez-Taxon 2.1 (http://www.eztaxon.org) was achieved
using CLUSTAL W software (Thompson et al., 1994) and
corrected manually. Phylogenetic and evolutionary ana-
lyses were conducted in MEGA6 (Tamura et al., 2013).

Strains 2C and 5C\textsuperscript{T} were isolated from surface water of Lake
Solenoe. They formed small, convex, circular, smooth,
orange-/red-pigmented colonies with a diameter of up to
2.5 mm on PCA and MA after 48 h at 30 °C. Cells were
Gram-stain-negative, aerobic, rod-shaped, non-motile and
non-spore-forming. Cells of strain 2C were 1.9–2.1 μm in
length and 0.4 μm in diameter, whereas those of 5C\textsuperscript{T} were
2.0–5.0 μm in length and 0.4 μm in diameter. They were
positive for oxidase and catalase activity. The strains grew
on MB at pH 7.0–10.0. Optimal pH for growth was 8.5–
9.0 (2C) and 8.0–8.5 (5C\textsuperscript{T}). Growth occurred in the presence
of up to 5 % (w/v) NaCl and at an optimum of 0–2.5 (w/v)
NaCl for both strains. Optimal temperatures for growth
were 25–33 °C in a temperature range from 21 to 39 °C.
The fatty acid profiles of strains 2C and 5C\textsuperscript{T} were dominated
by branched-chain fatty acids (iso-C\textsubscript{15} : 0, iso-C\textsubscript{15} : 1, anteiso-
C\textsubscript{15} : 0, iso-C\textsubscript{17} : 10:9c) and C\textsubscript{16} : 1ω6c, as reported for other
representatives of the genus (Brettar et al., 2004; Arun
et al., 2009; Akhwale et al., 2015; Zhong et al., 2015) and
were most similar to that of \textit{B. pelovolcani} CC-SAL-25\textsuperscript{T}
(Table 1). With regard to the major fatty acids, the pro-
portion of iso-C\textsubscript{15} : 0 in strains 2C and 5C\textsuperscript{T} was similar to
those observed in \textit{B. pelovolcani} CC-SAL-25\textsuperscript{T} and \textit{B. kenyensis}
No.164\textsuperscript{T}, but nearly twofold higher than in \textit{B. baltica}
BA 134\textsuperscript{T} and \textit{B. aquatica} TS-T86\textsuperscript{T}. The propor-
tion of iso-C\textsubscript{15} : 1 was two- to threefold lower than those
reported for \textit{B. baltica} BA 134\textsuperscript{T} and \textit{B. aquatica}
TS-T86\textsuperscript{T}. In general, the composition of major fatty
acids of strains 2C and 5C\textsuperscript{T} was similar to those of
\textit{B. pelovolcani} CC-SAL-25\textsuperscript{T} and \textit{B. kenyensis} No.164\textsuperscript{T}.

The flexirubin test was negative: the strains did not show
a change in colour after alkalinization with KOH. The
spectrum of the acetone–methanol extracts of both strains
showed a broad peak with a maximum around 470–
500 nm that is typical for carotenoids.

The predominant respiratory quinone was a menaquinone
with seven isoprene units (MK-7).

The 16S rRNA gene sequences of strains 2C and 5C\textsuperscript{T} were a
continuous stretch of 1427 and 1419 bp, respectively. The
strains showed highest 16S rRNA gene sequence similarity
to \textit{B. pelovolcani} CC-SAL-25\textsuperscript{T} (98.7 %) and \textit{B. kenyensis}
No.164\textsuperscript{T} (97.4 %), followed by \textit{B. baltica} BA134\textsuperscript{T} (96.1 %)
and \textit{B. aquatica} TS-T86\textsuperscript{T} (95.9 %). Phylogenetic analysis
using clustering via the neighbour-joining method showed
that the strains clustered within the same phylogenetic
group as the above type strains, formed a separate subclus-
ter with \textit{B. pelovolcani} CC-SAL-25\textsuperscript{T} and \textit{B. kenyensis}
No.164\textsuperscript{T}, but formed a distinct phyletic line (Fig. 1). The
topologies of the maximum-parsimony and maximum-lik-
elihood trees were similar to that of the neighbour-joining
tree (data not shown).

The G+C content of the DNA of strains 2C and 5C\textsuperscript{T} was
40.1 ± 0.12 and 38.2 ± 0.5 mol%, respectively.

According to the results of DNA–DNA hybridization
experiments, levels of similarity between strains 2C and
5C\textsuperscript{T} and its closest phylogenetic neighbour, \textit{B. pelovolcani}
CC-SAL-25\textsuperscript{T}, were 58.0 ± 2.5 %, which is considered
the lower limit for members of the same species
(Wayne et al., 1987). Based on DNA binding data and
high 16S rRNA gene sequence similarity, we suggest that
strains 2C and 5C\textsuperscript{T} belong to the same species (Stackeb-
randt & Goebel, 1994).

In addition to genotyping, we performed MALDI/TOF MS of
whole bacterial cells. This is a rapid and efficient method for
the identification and typing of micro-organisms (De Bruyne et al., 2011; Welker & Moore, 2011). Protein identification studies have shown that many of the signals of the bacterial mass patterns are derived from ribosomal or other abundant bacterial proteins (Sauer & Kliem, 2010).

In the present study, the protein peaks (unambiguously detectable) with a relative intensity $\geq 1\%$ were registered in the range of 2000–20000 $m/z$ (mass/charge). According to the mass spectra obtained there were 34 peaks of different relative intensities registered for all strains (Table 2). These peaks may serve as biomarkers either of members of the genus Belliella or at least the species group studied.

The spectra of strains 2C and 5CT were more similar to each other than to that of B. pelovolcani CC-SAL-25T. 2C and 5CT shared approximately 60 % of defined peaks and only 26 % with B. pelovolcani CC-SAL-25T. The unique peaks for strain 2C were registered at $m/z$ 3440, 3511, 5818 and 6876; the unique peaks for 5CT were at $m/z$ 2094, 6600, 9078, 9092 and 9726. By contrast, B. pelovolcani CC-SAL-25T had 17 unique peaks, the majority of which had significant intensities unlike the unique peaks for strains 2C and 5CT. Thus, the novel strains could be easily distinguished from the phylogenetically closest B. pelovolcani CC-SAL-25T based on MALDI/TOF MS.

Results of the API tests are given in detail in Tables S1 and S2 (available in the online Supplementary Material) for strains 2C, 5CT, B. pelovolcani CC-SAL-25T and B. baltica BA134T. In the API 50CH test system, strain 2C produced acid from 18 substrates and nine were weakly positive; 5CT produced acid from 22 substrates. The number of substrates used by strains 2C and 5CT was less than that reported for B. pelovolcani CC-SAL-25T (33), but greater than for B. baltica BA134T. All strains produced acid from L-arabinose, D-galactose, D-glucose, salicin, cellulbiose, maltose, lactose, melezitose, sucrose, trehalose, raffinose and starch. In the API 20E test system (Table S2) strains 2C and 5CT were positive for $\beta$-galactosidase and tryptophan deaminase, but negative for lysine decarboxylase, ornithine decarboxylase, urease, citrate utilization, acetoacetoin production, indol production and H2S production.

### Table 1. Cellular fatty acid compositions of strains 2C and 5CT and the type strains of reference strains

<table>
<thead>
<tr>
<th>Fatty acid</th>
<th>1</th>
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<th>3</th>
<th>4</th>
<th>5</th>
<th>6</th>
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<tr>
<td>C14:0</td>
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<td>0.76</td>
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<td>ND</td>
<td>&lt;1.0</td>
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<tr>
<td>C15:0</td>
<td>3.94</td>
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<td>3.83</td>
<td>0.5</td>
<td>1.7</td>
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<tr>
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<td>2.03</td>
<td>3.56</td>
<td>1.5</td>
<td>ND</td>
<td>1.3</td>
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<tr>
<td>Branched-chain</td>
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<td></td>
<td></td>
</tr>
<tr>
<td>iso-C14:0</td>
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<td>0.64</td>
<td>0.79</td>
<td>1.3</td>
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<tr>
<td>iso-C15:0</td>
<td>35.82</td>
<td>35.34</td>
<td>36.60</td>
<td>31.2</td>
<td>30.6</td>
<td>12.5</td>
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<tr>
<td>iso-C15:1G</td>
<td>6.77</td>
<td>7.88</td>
<td>5.61</td>
<td>1.9</td>
<td>22.9</td>
<td>10.07</td>
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<td>iso-C16:0</td>
<td>3.5</td>
<td>2.28</td>
<td>2.48</td>
<td>2.2</td>
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<tr>
<td>iso-C16:1</td>
<td>1.64</td>
<td>1.24</td>
<td>0.86</td>
<td>1.7</td>
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<tr>
<td>anteiso-C15:0</td>
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<td>5.73</td>
<td>4.9</td>
<td>2.9</td>
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<td>iso-C17:1v8c</td>
<td>2.59</td>
<td>3.61</td>
<td>2.82</td>
<td>ND</td>
<td>ND</td>
<td>ND</td>
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<td>1.10</td>
<td>1.61</td>
<td>1.06</td>
<td>6.4</td>
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<td>1.2</td>
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<td>C16:1v5c</td>
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<td>C17:1v6c</td>
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<td>2.90</td>
<td>3.09</td>
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<td>4.6</td>
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<tr>
<td>C16:0 3-OH</td>
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<td>0.55</td>
<td>0.32</td>
<td>2.5</td>
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<tr>
<td>C15:0 3-OH</td>
<td>1.10</td>
<td>1.59</td>
<td>0.85</td>
<td>4.2</td>
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<td>C15:0 2-OH</td>
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<td>3.11</td>
<td>1.2</td>
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<td>ND</td>
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<tr>
<td>C17:0 3-OH</td>
<td>2.30</td>
<td>2.98</td>
<td>2.50</td>
<td>6.7</td>
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<td>13.1</td>
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<td>2.35</td>
<td>2.13</td>
<td>2.31</td>
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</table>
Gelatin was hydrolysed by 2C but not by 5C<sup>T</sup>. Arginine dihydrolase activity was determined for 2C only. Strains 2C and 5C<sup>T</sup> differed from <i>B. pelovolcani</i> CC-SAL-25<sup>T</sup> in the spectrum of substrates used in carbohydrate metabolism, and a set of tested enzyme activities.

In relation to antibiotics, strains 2C and 5C<sup>T</sup> showed minor differences from <i>B. pelovolcani</i> CC-SAL-25<sup>T</sup>. Sensitivity to gentamicin (120 µg) was detected in both strains, but to amikacin in 5C<sup>T</sup> only (Table S3).

On the basis of a comparative analysis of phenotypic and genotypic properties, strains 2C and 5C<sup>T</sup> could be differentiated from the closely related <i>B. pelovolcani</i> CC-SAL-25<sup>T</sup> and other strains of the genus <i>Belliella</i>. A summary of the main differential characteristics is presented in Table 3.

![Phylogenetic tree](image)

**Fig. 1.** Neighbour-joining (Saitou & Nei, 1987) phylogenetic tree showing the position of strain 5C<sup>T</sup> among related species. The percentage of replicate trees in which the associated taxa clustered together in the bootstrap test (1000 replicates) is shown next to the branches (Felsenstein, 1985). Evolutionary distances were computed using the method of Jukes & Cantor (1969). Bar, 0.01 changes per nucleotide position.

<table>
<thead>
<tr>
<th>5C&lt;sup&gt;T&lt;/sup&gt;</th>
<th>2C</th>
<th>&lt;i&gt;B. pelovolcani&lt;/i&gt; CC-SAL-25&lt;sup&gt;T&lt;/sup&gt;</th>
</tr>
</thead>
<tbody>
<tr>
<td>2094, 6600, 9078, 9092, 9726</td>
<td>3440, 3511, 5818, 6876</td>
<td>2708, 3168, 3255, 3662, 3717, 4065, 4136, 4405, 4606, 4788, 5417, 6334, 6356, 6642, 6669, 9558, 12975</td>
</tr>
<tr>
<td>2070, 2086, 2108, 2164, 2208, 2216, 2410, 2549, 2703, 3161, 3212 (±)</td>
<td>3246, 3275, 3311, 3350, 3670, 4108, 4152, 4320, 4801, 4847, 5316, 5642, 5670, 6160, 6319, 6404, 6449, 6542, 6619, 6647, 7050, 7152, 7323, 7895, 8090, 8258, 9598, 9637, 9690, 11336, 11540, 12805, 12940</td>
<td>2552, 2602, 3141, 3325, 3338, 3403, 3424, 3546, 3615, 3691, 3822, 4503, 4560, 4807, 4820, 5024, 5037, 5136, 5202, 5404, 6226, 6278, 6489, 6510, 6802, 7227, 7377, 7640, 8998, 10047, 10070, 12268, 10628, 12449</td>
</tr>
</tbody>
</table>

Table 2. Mass values (<i>m/z</i>) specific for the <i>Belliella</i> strains
The name *Belliella buryatensis* sp. nov. is proposed to accommodate these strains.

**Description of *Belliella buryatensis* sp. nov.**

*Belliella buryatensis* (bu.ry.a.ten’sis. N.L. fem. adj. *buryatensis* pertaining to Buryatia, the geographical origin of the type strain).

Cells are Gram-stain-negative, aerobic, rod-shaped, non-motile and non-spore-forming, 2.0–5.0 μm in length and 0.4 μm in diameter. Good growth occurs after 48 h of incubation on MA at 30°C. Oxidase- and catalase-positive. Colonies on MA and PCA at 30°C are small, convex, circular, smooth, orange-/red-pigmented, 1.5–2.5 mm in diameter and non-fluorescent. Cellular pigments present are carotenoids, but not flexirubin. Optimal temperature for growth is 30–33°C. Optimal pH for growth is 8.0–8.5; growth occurs at pH 7.0–10.0. Growth occurs in the range 0–5% (w/v) NaCl; optimal growth occurs with 0–2.5% (w/v) NaCl. The main respiratory quinone is MK-7. The predominant fatty acids (% of the total) are iso-C_{15} : 0, iso-C_{15} : 1, anteiso-C_{15} : 0, C_{16} : 1 °c6c and iso-C_{17} : 1 °c9c. The following carbon sources are oxidized: glycerol, L-arabinose, D-galactose, D-glucose, D-fructose, methyl α-D-glucopyranoside, salicin, cellobiose, maltose, lactose, melibiose, sucrose, trehalose, raffinose, starch, turanose, potassium 2-ketogluconate, methyl α-D-mannopyranoside, amygdalin, arbutin and aesculin. Gentiobiose and L-arabitol are oxidized weakly. The following substrates are not oxidized: N-acetylglucosamine, D-adenitol, D-mannitol, D-sorbitol, D-fucose, D-arabitol, erythritol, D-arabinose, D-ribose, xylose, D-mannose, L-sorbose, L-rhamnose, dulcitol, inositol, xylitol, inulin, glycosgen, D-lyxose, D-tagatose, D-fucose and potassium gluconate. Positive for β-galactosidase and tryptophan deaminase; negative for arginine dihydrolase, lysine decarboxylase, ornithine decarboxylase, citrate utilization, H2S production, urease, indole production, acetoin production and gelatinase. Differential features are given in Table 3. The detailed fatty acid profile is given in Table 1.

The type strain is 5C^T (VKM B-2724^T = KCTC 32194^T), which was isolated from water of the alkaline brackish Lake Solenoe (Buryatia, Russia). 2C is a second strain of the species.

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**References**


